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An Analysis of Motivic Development in Schoenberg's Op. 11, No. 2 By Yuchen (Sally) Xi

Abstract

Looking back on composer Arnold Schoenberg's early composition and the development of 19th-century music, the transition from tonality to atonality is not abrupt, but rather a gradual process when composers retain some conventions in tonal music and begin experimenting with new techniques and ideas. This article unfolds the motivic development in Schoenberg's early piece Op. 11, No. 2, thereby reflecting that despite the use of new compositional techniques including pc-sets, Schoenberg can still ensure the coherence of the piece and illustrate the "tension—resolution" relationship, which are ideas retained from tonal music.

I. Introduction

"Schoenberg and Atonality" by Charles Rosen summarizes the development of Arnold Schoenberg's atonal music (298-327). In the 19th century, composers began to take a break from previous practices to enhance the expressiveness of music through explorations in chromaticism, less symmetrical forms, and exotic combinations of chords to break the hierarchy of notes. Rosen writes, "Every new style seems at once to make nonsense of the preceding one, and yet to retain part of it selectively as essential to its own operations" (301). Although Schoenberg departed from the convention of cadential tonic arrival, the conventions in tonal music, such as the idea of "tension-resolution" are presented in Schoenberg's music through different approaches, one of which is the motivic development (Rosen, 298-327).

Before 1915, when he first used twelve-tone serialism in his composition the Four Orchestral Songs Op. 22, Schoenberg explored various ways in which the hierarchy of notes and chords can be abandoned while the coherence of the piece is still maintained. As he wrote in "Analysis of the Four Orchestral Songs Opus 22", "about 1908 I had taken the first steps-also with songs-into that domain of composition [...] whose distinguishing characteristic is the abandonment both of a tonal center and of the methods of dissonance-treatment that had been customary up to that time. It was this latter feature, as I subsequently ascertained, that occurred if the perception of a dissonance could be ideally equated to that of a consonance" (1-21).

During this time, he composed *Drei Klavierstücke*, Op 11 consisting of three short pieces for solo piano. The pieces, written in ternary form and with tonal harmonic progressions as one might interpret, represent one of the first attempts of Schoenberg with atonal compositional techniques such as motivic development using unordered pitch sets. Hence, it is worthwhile to analyze and study the pieces to gain a whole picture of Schoenberg's evolution from tonality to atonality. Also, because Op. 11 was composed in his transition from tonality to atonality, it is still controversial whether the piece is atonal or not. As the composer did not provide clarification on this, the tonality of the piece remains open for interpretation. This paper will focus on Op. 11, No. 2 and analyze it from an atonal perspective.

As early atonal compositions preserve practices from tonal music, this transition from tonality to atonality is evolutionary and continuous rather than abrupt, as concluded by Howard

Octave complementation	<+8,+6> <+8,-6> <-8,+6> <-8,-6> <+6,+8> <+6,-8> <-6,-8> <-6,-8> <+6,+4> <+6,-4> <-6,+4> <-6,-4> <+4,+6> <+4,-6> <-4,-6> <-4,-6>
Expansion/shrink by one interval	<+5,+6> <+5,-6> <-5,+6> <-5,-6> <+6,+5> <+6,-5> <-6,-5> <-6,-5> <+3,+6> <+3,-6> <-3,+6> <-3,-6> <+3,+5> <+3,-5> <-3,-5> <-3,-5> <+4,+5> <+4,-5> <-4,+5> <-4,-5> <+5,+4> <+5,-4> <-5,-4> <-5,-4>

In the same article, Boss also explained the concept of “a chain of motives”, a term coined by Schoenberg that refers to the generation of new motives through the overlapping of the previous ones, although the technique is not explicitly employed in Op. 11, No. 2 (Boss, 125-149).

II. Motivic Development

As concluded in Cinnamon’s article, the writer agrees that the structure of the piece can be roughly divided into three sections—a large exposition, rapid development, and an abbreviated recapitulation, as shown in Table 2. My analysis agrees with Cinnamon Figure 7, with the exception that I included a C3 section to show the repeated occurrence of the motive as follows.

The exposition can be further divided into sections A, B, and C, with new materials introduced in sections A and C and section B acting as a development and transition of the ideas.

Table 2 The tertiary form of Schoenberg’s Op. 11, No. 2

Exposition (1-39)										
A			B	A'			C			
A ₁	A ₂	A ₁ '	B ₁	A ₃	A ₄	A ₅ '	C ₁	C ₂	C ₁ '	C ₃
1-4	4-13	13-15	16-19	20-22	23-25	26-28	29-30	31-32	33-38	39

Development (40-54)				Recapitulation (55-66)					
				A	B	A	C		Coda
B ₁ '	A ₂ '	C ₃ '	C ₁ '	A ₁ '	B ₁ '	A ₂ '	C ₁ '	C ₃ '	A ₂ '
40-4 2	43-47	48-49	50-54	55-58	59-60	61-62	63-64	65	65-66

Below, the writer will analyze in detail the development of motives using set theory and the repeated occurrence of intervals including minor thirds and major sevenths.

In A_1 (bar 1-4), the beginning of the piece, Schoenberg quickly establishes the tonality of the piece as atonal. While the ostinato on the left hand alternates between D and F, giving the first impression of D-minor, the melody on the right hand plays in a tonal center of Db. The difference in tonality in the melody and the ostinato indicates either polytonality or atonality, where the former is proved wrong with the introduction of new motivic materials in A_2 . While Cinnamon treated the piece as tonal, in this paper, I will interpret it as an atonal composition and focus on analyzing the use of set theory in it.

In the beginning of the piece, the right hand introduces one of the main motives: $\langle +8, -6 \rangle$. The motive reappears in the following phrase in bar 3 as $\langle -6, -2 \rangle$, a pitch reordering of the original motive. They have the same prime form, as will be discussed later.

Figure 2 Bar 1-4: A_1

The motive, after pitch reordering, also appears in bars 4-5 (A_2), further reinforcing it as one of the main motives and allowing listeners to remember its presence.

Figure 3 Bar 4-5: A_2

Two other main motives— $\langle +2, +3 \rangle$ and $\langle +5, -1 \rangle$ are also introduced in A_2 , from bars 4-13, as marked in Figure 3. These motives are developed into more variant forms. For example, $\langle +2, +4 \rangle$ in bar 11 is an intervallic expansion from $\langle +2, +3 \rangle$ while $\langle -2, -9 \rangle$ in bar 8 is an octave complementation of $\langle -2, +3 \rangle$.

A, B, D: <+2,+3> G#, C#, C: <+5,-1>

F, Eb, Gb, <2, 0> A, B, D: <+2,+3> D, E, G#: <+2,+4>

Figure 4 Bars 4-11: A₂

The ending of A₂ in bar 13 is marked by a fermata, after which the melody from bars 1-4 and the motive <+8,-6> reoccurs. Section B functions as a short transition from section A to section C, where motivic materials brought up in section A are developed into more remote forms. As can be seen in the below figure, the motives are developed from the original presentation into more remote forms after intervallic expansion and octave complementation. Moreover, they are more closely packed, with the accompaniment continuously playing chromatic movements to build up tension and prepare listeners for the introduction of new materials in section C. This tendency toward chromaticism when transitioning between musical materials, as concluded by Rosen, had been applied in the late Romantic period by composers including Liszt (326). Hence, he argues that the use of chromaticism in Schoenberg's music is a continuation of the previous compositional devices in tonal music.

C#, A, Eb: <+8,-6> Eb, D, A: <-1,-5> G, D, Eb: <-5,+13> E, D, B: <2, 3>

Chromaticism

Figure 5 Bars 16-19

This tendency toward chromaticism is also evident almost throughout the development of the piece, namely bars 40-47, pushing toward the climax. Specifically, aside from the chromatic movement of the tremolos and 32nd notes, the motive $\langle +5, -13 \rangle$ is also transposed chromatically upward, whereas $\langle +2, +3 \rangle$, in its most original form, is transposed upward as well, thereby heightening the tension and pushing toward the climax of the piece.

The image displays three systems of musical notation for piano, covering bars 42-47. The first system shows the right and left hands with dynamic markings *f*, *p*, *f*, and *pp*. Orange boxes highlight specific melodic phrases in both hands, with a label $\langle +5, -13 \rangle$ and the text "Chromatic transposition of the motive on both hands". The second system includes a *rit.* marking and a *tr.* (trill) marking, with a dynamic marking of *ff* and a label $\langle +2, +3 \rangle$. The third system features a *ff* dynamic, a *non legato* instruction, and a *rit.* marking. Orange boxes continue to highlight the $\langle +2, +3 \rangle$ motive across the systems.

Figure 6 Bars 42-47

The recapitulation of the piece follows the ternary form in the exposition as well, though it is abbreviated. Following the reoccurrence of the main melody and the motive $\langle +8, -6 \rangle$ in its most original form, section B reappears before the repetition of section C. In the coda, the motives $\langle +2, +3 \rangle$ and $\langle +5, -1 \rangle$ reappear in their original forms, putting an end to the piece.

A, B, D:
 $\langle +2, +3 \rangle$



Figure 8 Bars 65-66

III. Musical Coherence and Resolution

One advantage of sticking to tonality or a tonal center is that they ensure the coherence of the piece as composers would frequently end a phrase on the tonic or dominant through the use of cadences. This, indeed, is one of the challenges Schoenberg faced when attempting to treat all consonances and dissonances equally.

In Op. 11, No.2, the coherence of the piece is maintained through the repeated and consistent use of several unordered pitch sets. Specifically, three unordered pitch class sets are used prominently in the main motives and themes of the piece. The table below shows their prime forms and subsets. The 01457 set and its subsets agree with Cinnamon's Figure 14.

Table 3 prominent unordered pc sets and subsets used in the piece

Sets	Significant subsets
(01468)	(0247) (0248) (0157) (0148) (0146)
(01457)	(0145) (0147) (0157) (0237) (0236)
(01347)	(0236) (0347) (0147) (0137) (0134)

The following two figures show how the main motives of the piece all belong to the same sets of (0,1,4,6,8) and (0,1,4,5,7), which ensures the coherence of the piece as the motives all derive from the same pc sets, similar to the conclusion in Cinnamon's Figure 22.



Figure 9 Bar 2



Figure 10 Bar 4

G#, C#, C, A: (0,1,4,5)

Figure 11 Bar 5

rit. *ff* *ff* *non legato* *rit.* *mf*

Eb, A, B, D: (0,1,4,6)

Bb, E, F#, A: (0,1,4,6)

C, Ab, D, G: (0,1,5,7)

(0,1,4,6) (0,1,4,8) (0,1,5,7) (0,1,5,7)

(0,1,3,4,7) (0,1,4,6) (0,1,4,6)

Figure 12 Bars 45-49: C₂-C₃

Interestingly, the main motive in section C, as shown in the figure below, is characterized not by the interval sets, but by the unordered pitch sets and the 4/4 time signature. As a result, even though the intervals between the notes are not consistent, the fact that the notes belong to the same subset creates a sense of coherence and relation between the pitches, thereby making the motive recognizable.

(0,1,5,7)

Figure 13 Bars 29: C₃

Figure 14 Bars 35-36: C₁

Another difficulty in composing atonal music, presenting the idea of tension-resolution without using harmonic progressions, is also achieved through unordered pitch sets and the repetition of certain intervals. First, the repeated use of the unordered pitch set (0,1,2,3,6,8) marks the ending of sections, for example in bar 39, the end of the exposition, and the end of the piece). Hence, it can be deferred that the pc set (0,1,2,3,6,8) acts similar to a perfect cadence in tonal music, as its presence forms a resolution.

Figure 15 Bar 39, end of exposition

Figure 16 Bar 66, end of the piece

Aside from the aforementioned pc-sets, the emphasis on intervals including major seventh and minor thirds over others also expresses the idea of tension-resolution. The ostinato on the left hand is one prominent feature of the piece. The interval of the ostinato is mainly minor thirds, but it expands and develops to M3, m7, and a tritone during the exposition and development of the piece.

Figure 17 Bar 1: ostinato on the left hand with an interval of m3

Figure 18 Bar 20: ostinato on the left hand in an interval of M3

Figure 19 Bar 23: ostinato on the left hand, presenting intervals of m7, M3, and a tritone



Figure 20 Bar 26: ostinato on the left hand in an interval of m3



Figure 21 bar 55: ostinato on the left hand with an interval of m3

Horizontally, the ostinato mainly in the interval of minor thirds establishes the importance of the interval m3, similar to the tonic in tonal music, thus providing a sense of resolution as the piece concludes with a m3 movement from Ab to F on the soprano, as shown in the figure below.



Figure 22 Bar 66: the ending of the piece

Similarly, examining the piece vertically, one can identify the repeated occurrences of the interval major seventh in a large number of chords, as evident in bars 9, 66, and many other bars. With the last chord containing a major seventh of Gb to F, Schoenberg again attempts to provide a sense of resolution.

From the above discussion, it can be seen that despite the absence of the hierarchy of notes and tonality, Schoenberg presents the idea of “tension-resolution” through the consistent and repeated occurrence of minor thirds and major sevenths. These intervals replace the function of a tonic, hence allowing the dissonances to be kept throughout the piece.

Though not manifested through harmonic progression, the idea of “tension-resolution” is achieved by the form and motivic development of the piece; and though the piece abandoned the tonic or a tonal center, the coherence is achieved as the motives used are from the same pc sets. This paper examines Schoenberg’s early composition Op. 11, No. 2 using set theory and prominent intervallic and structural features. It agrees with Cinnamon’s conclusion on the structure and sets used in Op. 11, No. 2. As an extension, the paper treats the piece as atonal and examines its motivic development. It analyzes the structure and the motivic development, as well as how Schoenberg demonstrates certain compositional conventions in tonal music, for instance, the “tension-resolution” idea using unordered pc-sets and the repetition of certain intervals. Overall, Op. 11, No. 2 provides listeners a glance at Schoenberg’s gradual transition from tonality to atonality of incorporating certain common practices of the time with new compositional techniques.

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Impact of Consumer Psychology in Digital Marketing of Healthy Food Products By Saina Prasanna

Abstract

In today's marketing world, digital marketing has become a powerful tool globally to advertise a number of goods that can address health concerns and promote a healthier lifestyle through diet. Consumer psychology plays the important role of aiding in designing a marketing campaign, since the study of consumer psychology allows for the implementation of several psychological principles that are able to attract and persuade leads into becoming regular customers. This paper explores how people are influenced by consumer psychology principles embedded in digital marketing campaigns of larger corporations, who have the common goal of marketing their healthy product to the general public. This paper delves deeper into the topics of color psychology, cognitive bias framing, reciprocity effect and addresses the challenges that digital marketing campaigns face when promoting healthier foods using consumer psychology.

Introduction

Did you know that Chobani's digital marketing campaigns garner over 50K views online (Latterly, 2024)? That's what earned Chobani 1.75 billion dollars via in-store sales in 2023 (Bloomberg, 2024). Brands use digital marketing to establish and communicate with an audience, to build loyal relationships with consumers to increase purchases and generate income (Market Tactics, 2024). Companies also use consumer psychology to build relationships with potential consumers by tailoring what is marketed to the viewer's preferences, beliefs, and emotions. This paper explores how consumer psychology aids in digitally marketing healthier foods to consumers and evaluates the extent to which digital marketing is effective, through two experiments and a case study.

Overview

Digital marketing has rapidly become one of the most versatile methods of conducting marketing to promote a business's goods or services. Digital marketing is a "large umbrella" term that consists of using search engines, social media, websites, and advertisements to attract and reach potential consumers with ease (Hubspot, 2024). Marketers use digital marketing to establish a connection between the brand and the consumer, which further encourages the consumer to purchase the product. For example, social media targeting can be an effective tool for creating ads that are aimed toward certain demographics based on age, interests, gender, location, and popular trends (Inspira, 2023). Content creation on Instagram, Facebook, and TikTok allows for businesses to monitor and construct stories, reels, captions, and ads that will captivate a person's attention. This not only allows for an increase in purchases but also an increase in impression and click rates via a viewer, subscriber, or influencer. This could result in the brand being shared on their platform or with the people they know, further pushing that brand's content towards the algorithm (Hubspot, 2024). Through digital marketing, small

businesses are able to scale up and compete with more renowned brands in a more cost-effective manner by having control over where and how much money is being invested into their content creation.

Digital marketing offers numerous benefits for both marketers and consumers through strategies such as email marketing and SEO marketing. For example, to build a loyal relationship with a consumer and further promote their goods or services, brands often use email marketing and send their subscriber personal and relatable content to foster connections such as giveaways or notices for when an anticipated product is about to release or to address general pain points a consumer is experiencing (My Emma, 2019). Digital marketers also offer insights into certain pain points and how their product can solve those. SEO marketing or search engine optimization marketing may come into play when a business desires to reach its target demographic by tracking common keywords searched by their target audience to convey their wants, providing insights on what to include on their website to improve their website's ranking on a search engine such as Google (Acadium, 2019). Digital marketing is also measurable in the sense that the number of views and purchases from a post, ad or website can be monitored through digital analytics software, allowing for digital marketing campaigns to make adjustments to their strategies accordingly (Hubspot, 2024). Digital marketing is so vastly used amongst brands due to how all of us, globally, are shifting to a digital world in which social media and search engines have become a necessity to our day-to-day lives. It allows marketers to reach audiences, and promote products and services in unprecedented ways (ScienceDirect, 2021)

Consumers tend to make choices about which products to buy before they purchase them by weighing the pros and cons of the product such as the price or practicality. Consumer psychology focuses on discovering what motivates a person to make a purchase, whether that be emotion, thoughts, or beliefs. The goal of applied consumer psychology in marketing and other fields is to cater to the target demographic in a more efficient manner (USC Dornsife, 2023). SEO marketing often utilizes consumer psychology by tracking what keywords consumers most frequently search for allowing them to highlight those keywords on their website. SEO marketers may also use consumer psychology by taking advantage of the fact that consumers often look at other people's experiences to make decisions (Thimpress, 2022); this can be done by including positive reviews of their products or services on their websites. In email marketing, marketers may appeal to their audience through emotions such as FOMO (fear of missing out) to persuade the target audience to purchase a product that is available for a limited amount of time (Spiralytics, 2022). Consumer psychology uses different phenomena such as color psychology, the reciprocity effect, and cognitive framing to create a marketing campaign for a product that captivates people's attention.

Color Psychology

Color psychology delves into the wonder of how human emotion is influenced by color; it is the study of how different shades, colors, and hues can influence a consumer's mood and attract them towards a certain post or ad of a product resulting in a purchase. Color psychology is

also used to convey a brand's message or core values to its consumers. Ninety-three percent of consumers make purchasing decisions based on color alone (Maybray, 2023). Color can also be used as a defining feature for recognizing a brand by utilizing color in a label or a series of posts. Different colors can be used within a post, website, logo, or packaging to induce different emotions from a viewer to inspire connections, and loyalty and also convince a potential customer to buy a product (Paper Turn Blog, 2024).

Different colors hold different meanings that can be modified to express a brand's message. For example, blue is a color that depicts security, reliability, trust, and logic; orange sparks confidence, creativity, and warmth (Hubspot, 2023). Green is the color for growth, freshness, nature, and nourishment (it is utilized by the brandHelloFresh; their lime green logo symbolizes healthy meal kits) (Hubspot, 2023). The color purple is often used to convey a sense of superiority or royalty, which is why it is often used by luxury brands such as Hallmark (Hubspot, 2023).

Reciprocity Effect

The reciprocity effect is a phenomenon where an individual feels compelled to return a kind favor if they have received one (Psychology Today, 2015). An example would be when small brands and ice cream stores present free samples of their product to an individual compelling the person to buy the product. Giveaways and rewards from a brand also influence a person's feelings such as a sense of gratitude resulting in them wanting to purchase a product from a brand to increase their sales and brand awareness (Wisernotify, 2023). At times, the reciprocity effect increases brand awareness by displaying a brand's great customer service; this can result in loyalty, but also a large number of positive reviews online. Using this effect to offer discounts and "Buy 1 get 1 free" deals influences a potential consumer to purchase a product since they want to return the positive experience they had just received (Wisernotify, 2023). Marketers can build trust and establish a connection to their consumers by using the reciprocity effect since it establishes trust between the brand and the consumer.

Framing Effect

The framing effect is a strategy used by marketers to use wording to its fullest potential to create a message that attracts the target demographic of a certain product (Investopedia, 2023). Framing can dictate decisions and influence people's responses. For instance, a skincare brand will be received more positively if it markets the fact that "70% experience improved skin" rather than "30% don't experience improved skin". Both phrases mean the same thing, but the wording will evoke different emotions which is why brands frame their message in a more positive light. Another example is a small brand specializing in creating products for a healthy diet changing its name from Living LoCarb to Nourish-ology to gain more attraction so that people realize this brand and its products are not only for those who want to pursue a low-carb lifestyle, but for those who want to pursue a healthy lifestyle in general. Marketers use the

framing effect to determine what it is that they want to highlight about their product in a way that evokes a positive response from the consumer resulting in a purchase (Linkedin, 2021).

How Brands Promote Healthier Foods

Experiment 1: Color Psychology

Living Locarb is a woman-owned small business that enforces the power of dietary changes by creating products that are gluten, sugar, and dairy-free and low in carbs. Across their Instagram page, they use vibrant colors such as yellow and orange to attract viewers and potential consumers. Living LoCarb also uses yellow and orange as the base colors of the labeling on their gluten-free chia seed puddings, Saffron Vanilla, and Nutti Nirvana. The color yellow exudes feelings of happiness and spontaneity while orange emits the emotions of liveliness, encouragement, rejuvenation, and vibrancy (Nourish-o-logy, 2024), which are the main messages they are trying to promote. Transforming your diet by using healthier alternatives can lift a person's mood and energy levels. By using vibrant colors on social media to post about the product or certain events where the product is being showcased, not only are the colors of the post radiating the positive energy the brand is trying to convey, but the post is also more eye-catching and bound to get a viewer's attention easier.

Since Living Locarb is a small business that needs to grow its following, I decided to conduct an A/B test to see how changing the main colors of the brand would impact its business. I created two Instagram posts promoting the chia seed puddings, where one post's background remained as a gradient of yellow to orange (figure 1) to enforce the brand's message of happiness and vibrancy and then created another post that had the same features aside from the background which was now a forest green (figure 2). Forest Green is a color that emanates tranquility, low stress, health, and prosperity (Color Psychology). I hypothesize that people would be more persuaded to purchase the gluten-free and vegan chia seed puddings based on the forest green colored post since the colors directly convey the messages of health and prosperity louder which is what the product is trying to promote rather than the yellow-orange post which mainly conveys feelings of positivity. Within the post, I included a high rating of it to build some trust between the brand and the consumer and also build some credibility of the brand. I also included some keywords such as zero sugar and weight management to showcase the diverse benefits of the product. The post also includes a sense of inclusivity by promoting that the chia seed puddings are dairy-free and vegan so it can be included in many dietary plans even if you have allergies or aversions to certain foods.



Fig 1: yellow-orange post of chia seed puddings Fig 2: Forest green post of chia seed puddings

After creating these posts I conducted a survey and asked thirty-five people which post would convince them to buy the chia seed puddings. The responses I got were overwhelmingly in support of the yellow-orange post with the main reason being the bright colors in that post were more appealing and eye-catching and those bright colors highlight the food more (figure 3). The minority who responded with the forest green post found that post easier on the eyes and easier to read. However, none of the responses for the forest green post mentioned anything in correlation to the message the green was trying to convey for the product. Based on the fact that most people were to decipher the meaning of one post, the yellow-orange post, and not the other, color psychology works to a moderate extent in aiding digital marketing.

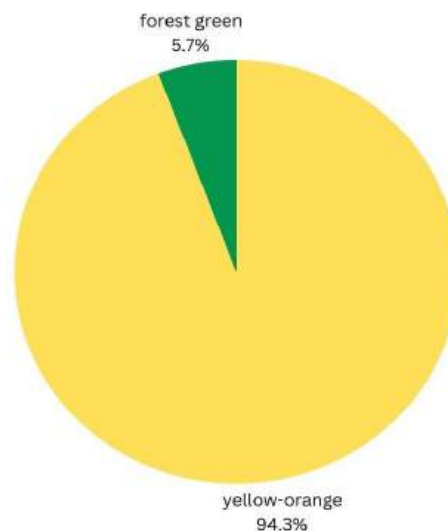


Fig 3: Results of Color Psychology A/B test

The responses I got could have contradicted my hypothesis because people may be more familiar with the association of yellow being correlated to happiness and orange being correlated to liveliness since they are exposed to stimuli that resemble that meaning more often such as the sun, flowers and even in the media where yellow has been established as the color of joy. However, people may be less familiar with the association of green with freshness since it is obscured because of more common meanings such as envy or disgust. For example, waste, slime, and green vegetables are depicted as green to elicit a response of disgust. Forest green having a more diverse range of meanings may have made it more difficult to get one specific meaning, freshness, across to the audience. However, yellow and orange which are bright colors have broader and similar meanings of liveliness, spontaneity, and happiness and for that reason may have been easier to communicate.

Experiment 2: Reciprocity Effect

Small businesses often showcase their products through product demonstrations during events or in front of the stores they are selling them at. These product demonstrations help build a connection between brands and potential clients, but it is also a huge opportunity to showcase their goods and collect ratings or photos of people with the product for their social media profile. Living LoCarb has hosted and participated in many events to showcase their chia seed puddings through sampling.

I acted as a brand ambassador for Living LoCarb to test the outcome of the reciprocity effect. Based on the reciprocity effect, I hypothesized that people would be more inclined to purchase the chia seed puddings after being given a sample. During the event, I gave samples of both the Saffron Vanilla and Nutti Nirvana chia seed puddings to the 30 people who stopped by our booth. Although many samples were given the sales in return were quite low with 4 people purchasing out of the 30 people at the event (figure 4). When those who purchased the chia seed pudding were asked if they would have bought it if there had not been any samples, the results were fifty-fifty. Those who responded with yes, stated that they would have bought it without the sampling since they are already familiar with chia seeds. Those who responded with no, stated they would not have bought it without the sampling because they did not know much about the product before the sampling and assumed that “pudding” referred to an unhealthy dessert. However, none of the responses alluded to purchasing the chia seed puddings due to feeling compelled to do something in return after receiving a kind gesture. For this reason, the reciprocity effect works to a low extent in aiding digital marketing.

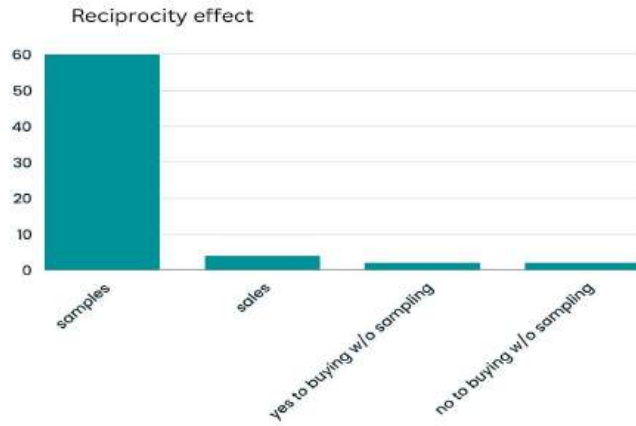


Fig 4: Results of the Reciprocity Effect for Living Locarb

The brand Simply Bhonu is another woman-owned small business that sells chai tea blends that attended a non-sampling event and an event that used sampling in 2023. They found that sampling tends to provide better results if the product isn't well known to the audience, but the difference isn't substantial enough to be considered relevant (figure 5). Another discovery was that a high foot traffic location was more significant than sampling. In addition to this, sampling was more advantageous during small pop-up events where the goal is to spread brand awareness

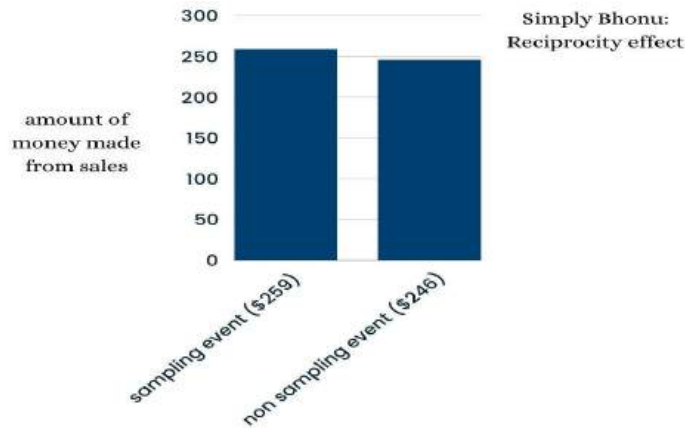


Fig 5: Results for reciprocity effect for Simply Bhonu

Case Study: Framing Effect

HelloFresh is a meal kit service that provides ingredients and recipes for creating meals based on customizable dietary plans and restrictions. One of the brand's main objectives is to aid in reducing food waste, which they accomplish by spreading awareness to their digital audience on Instagram. HelloFresh made a post on their Instagram stating that "7 in 10 people are trying to cut down on food waste," which is the same as saying "3 in 10 people are not trying to cut down on food waste" but it will invoke a different response. By saying the majority of the population

wants to cut down on food waste, the issue of food waste seems a lot more significant since so many people are working to solve the issue. Showcasing the importance of the cause will convince others to participate in finding solutions to the food waste crisis. HelloFresh's solution to food waste is its meal kits.

On the contrary to the previous post, Hello Fresh also wanted to showcase the dire circumstances of food waste by posting "The Top 10 Reasons Why People End Up Throwing Away Unused Food." According to the post, 70 percent report they do so because it goes bad before they can use it, 35 percent report they forgot about the unused food, and 23 percent report that they do not like leftovers. This data focuses on the negative aspects of the food waste issue to elicit a negative reaction from HelloFresh's audience whether that be disappointment or disgust. By invoking these feelings, HelloFresh's followers and other potential consumers will have an incentive to want to fix the issue which again can be done by buying HelloFresh's meal kits.

When selling their meal kits, HelloFresh also showcases the price per serving below the box price since the price per serving appears to be significantly cheaper (Customer Camp, 2023). HelloFresh also commented on the issue of food waste by stating, "Twenty-three percent of people who go grocery shopping wind up purchasing at least 5 food items a week they wish they could get in smaller quantities." (HelloFresh, 2023) They framed their comment in this manner to spread awareness of food waste so people are more inclined to take the cause seriously and support their meal kits. If HelloFresh were to phrase it, "Seventy-seven percent of people who go grocery shopping don't wind up purchasing at least 5 food items a week they wish they could get in smaller quantities," the issue wouldn't come across as urgent and important since most grocery shoppers don't struggle with the issue of wishing they could get items in smaller quantities. These types of carousels are extremely effective due to the interest they generate, which is just over 200 likes greater when compared to other posts.

Conclusion

Digital marketing has taken the world by storm, helping brands build their foundation, enhance their public image, and grow to new heights. Consumer psychology continues to play a massive role in digital marketing to help pave the way towards growth and success with some strategies working better than others. The reciprocity effect aids to a limited extent when digitally marketing healthy foods due to people interpreting sampling as an introduction to the product rather than an act of kindness. Color psychology works to a moderate degree due to certain colors being correlated with an associated meaning that is easier to decipher framing effect aids to a high extent when digitally marketing healthier foods since the message is clear and impactful in order to tailor to people's emotions and beliefs, motivating them to buy the product.

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The Relationship Between High School Funding and Gender Gap in Mathematics

By Paige Baker

Abstract

Debates over how much funding should be allocated to public high schools have fueled numerous studies. The increased focus on gender equality in the past few decades has opened the door to further research on reaching gender equality in the STEM workforce. The combination of educational funding and gender equality allows for a further study on how to improve high school education. Using results from the High School Longitudinal Study of 2009, a national test on mathematics, English, and science, we investigated the role that school funding had on the gender gap in mathematics. We found that an increase in socioeconomic status generally predicts an increase in mathematics score. We found that gender has no significance when it comes to predicting mathematics scores. The most important finding is that the interaction between gender and socioeconomic status holds significance as a predictor for mathematics scores. This finding reveals that the gender gap has not been completely closed and there is still improvement needed in education. Surprisingly, we found that the gender gap in mathematics is larger for students with higher socioeconomic status compared to those with average socioeconomic status.

Introduction

The gender gap in STEM subjects is a widely studied field. The gender gap in the STEM field generally consists of men outperforming women in areas including test scores and STEM interest and engagement (Grosch et al.; Zander et al.). From elementary to college level education, the causes and effects of these gaps have been researched. From socioeconomic conditions to classroom environments, a variety of causes have been suggested (Grosch et al.; Pope and Sydnor; Reardon et al.; Sadler et al.). In a similar fashion, many solutions and possible treatments have been proposed to support closing the gap. One STEM field that has been the subject of many studies is the mathematics gap. As the basis of many STEM related careers, math is an important area of focus to ensure gender equality beyond just education. Starting from a young age, it is important to boost confidence in young girls so they have the skills needed to join the STEM field when they are older. Creating an equal environment in earlier education can help set up girls for future careers in STEM (Grosch et al.).

A variety of causes of the mathematics gap have been proposed by researchers. One of the most frequent hypotheses of the cause of the mathematics gender gap is student location. For one, student location can indicate poverty level which has been known to be another possible cause of the gender gap (Pope and Sydnor; Reardon et al.). Location is also important as due to different educational policies from state to state and different economic conditions in districts, the gender gap varies throughout the United States. For example, states in the South and Mountain West tend to have a greater gender gap favoring boys in math and science (Pope and Sydnor). While Pope and Sydnor (2010) suggest this gap could be a result of traditional gender stereotypes in the areas, a possible separate cause could be due to the wealth status of the areas.

It was found that in wealthier districts and districts with socioeconomic gender inequality, the math gap favors boys (Reardon et al.). These two studies suggest student location as a predictor of the mathematics gender gap and can be used to focus on specific solutions to the gap by area.

In similar fashion, the effects of different levels of funding on testing scores have also been frequently studied. As a result of education being state controlled and partially state funded, there are large differences in education between states (Baker et al.). These differences in education shape each student's learning experience both on the state and local levels. One possible result of these differences is an increased gender gap in mathematics.

As a result of factors such as geographic location, district size, and poverty, per-pupil spending varies from state to state (Baker et al., 2014). This difference in funding results in unequal implementations of beneficial education policies put forth. Under-funded districts typically have to hire teachers with lower qualifications who exhibit higher turnover rates (Adamson and Darling-Hammond). This in turn can hurt the gender gap as it has been shown that teachers have a large impact on girls showing an interest in STEM (Hazari et al.). While there is some disbelief that increased funding of education results in vastly improved test scores (Lips & Watkins, 2008), it is clear there needs to be some sort of change in educational policy to help underperforming students. Whether it is a complete overhaul to school budgets or changing the way schools are funded, change is needed. There are clear effects of district wide poverty on learning. One of the biggest effects is a decreased graduation rate in areas with greater poverty (Baydu et al.). With this connection between an area's socioeconomic status and school performance, it is possible that district funding could also cause a change in the math gender gap.

Our goal with this study is to combine the math gender gap and school funding into one area of research. Using data from the High School Longitudinal Study of 2009 given by the National Center for Education Statistics, we aimed to study the relationship between school funding and the math gender gap. We predicted that districts with higher levels of funding would have a decreased gender gap in math performance. This relationship is an important area of study as it would give cause for states to change both their educational funding and systems. It could also extend to the federal level as the government could also step in to encourage progress.

Literature Review

Educational funding and gender inequality in STEM fields have both been widely studied in their own fields. Results of these studies are important to look into in order to observe possible connections and crossovers between the two fields.

Research has consistently shown that educational funding is not used effectively (Allegretto et al.; Baker et al.; Fahle, and Reardon). Due to the majority of funding responsibility being allocated to the states, there is a large range in educational spending. This largely affects poverty stricken districts as per-pupil spending is much lower in these areas compared to other parts of the country (Baker et al., 2014). With these inequities, it becomes hard for education to be standardized and for effective educational policies to be put into place. National test results have shown differences in test results between districts, especially in areas with racial and

economic segregation (Fahle, and Reardon). This failure can also be seen on a smaller level in lower income districts as they struggle to match increased spending by wealthier districts in higher spending states. These shortcomings and inequality in education can be seen by many states' failures to return to pre-recession level spending (Allegretto et al., 2022). The recession hit education hard, and due to the lack of a large federal presence, states have had a hard time catching up.

Besides funding issues, many states are also underspending relative to their capacity (Allegretto et al., 2022). This means that many states have the ability to increase spending or use their funds more effectively, but are unable to do so due to various reasons. It is possible for states to use less funding to create effective education. It has been made clear that it is not always funding that causes success in education, but effective policy. This is seen in states like Arkansas and South Carolina who do not have as large of a budget as other states in regard to education, but use policy to use it more effectively. This creates a larger focus on education. While studies have found that increasing spending on education does not consistently result in significant increases in standardized test scores (Lips & Watkins, 2008), this could be a result of only looking at improvement in linear models. Looking at improvement as a curve suggests that a certain spending cutoff renders spending effective (Lockwood). The model illustrates that until spending reaches a specific cutoff, there will not be dramatic increases in student test scores.

While girls have been closing the math gap when it comes to average scores, studies have shown that boys have been outperforming girls at the top level (Pope and Sydnor). This trend can be seen across the United States as districts with high performances in math testing have scores favoring men (Reardon et al.). While it is significant to see a decreasing gap in average math test scores, there is still progress to be made. One example of progress is the closing gender gap in high school statistics. One study found that girls had an overall higher GPA and a higher reported academic effort (Sartain, Lauren et al., 2023). The same study found that the math gender gap decreased in honors courses. These results suggest classroom attitudes can change student performances, but do not explain the state and national testing results. With these studies in mind, it is clear that there is reason to study the effects of school funding as a possible cause of the gender gap. Identifying the root of the issue makes it easier to completely solve the problem.

Decreasing the math gender gap is important in the greater STEM field. High school performance has a large impact on college and career choices. It was found that having a STEM career interest at the beginning of high school increased the chance of having the same interest at the end of high school (Sadler et al.). Results from this study showed that half of the girls who showed an interest in STEM at the end of high school were interested at the start of high school. Increasing girl's performance in math could result in increased interest in math and as a result an increased number of girls interested in math careers. The same study found that more boys are attracted to STEM subjects than girls. This is concerning as girls already start with a lower interest in STEM fields and lose interest in STEM over high school at a higher rate than boys (Grosch et al.; Sadler et al.). With this, it is important to keep female test scores high to encourage STEM interests.

During elementary school, girls have reported a lower interest in STEM subjects (Grosch et al., 2022). One possible cause of the lower STEM interest in students is lower self-efficacy and personal interests. At times, it has been found that despite having the same or better grades, girls have lower self-efficacy than boys (Zander et al.). This trend of thinking can start as early as elementary school with girls reporting lower self-efficacy and showing less confidence in future math endeavors. This way of thinking builds over time and can prevent girls from actively pursuing STEM as an interest. It has been found that boys have higher math confidence and were more willing to enter in math competitions than girls were (Grosch et al.). This shows that it is important to start reducing stereotypical gender thinking at a young age before it becomes too ingrained. Identifying school funding as a possible predictor could help equalize the field even more and help more girls retain their interest in the STEM field. Finding every possible factor in gender equality is important in order to minimize the impact of the gender gap..

With these findings, there is a clear possibility of school funding affecting the gender gap in high school math. Concretely identifying the relationship would help prevent harmful consequences down the line in college and in future careers.

Methods

Sample

The data used was collected from the High School Longitudinal Study of 2009 (HSLs).. This was a longitudinal national test given to high schoolers of various backgrounds. Students were first given the test as 9th graders in 2009, and then followed up with as 11th graders in 2012. Students given the test filled out surveys about themselves including their gender and math education background. Parents, school counselors, and school administrators were also given surveys to fill out. Information from these surveys included parental income and school environment. This test also included English and Science sections, but for this study only mathematics results were looked into. The sample taken included results from 23,503 students. Variables:

Both categorical variables and numerical variables were used as predictors. While gender is a categorical variable, for this study it was used as a dichotomous predictor. The HSLs test only gave two options for gender: male and female. Every student who answered the question put themselves in one of those two categories. Female became the indicator variable meaning it holds a value of 1. Male became the reference category meaning it holds the value of 0. Math scores were used as the predicted value. Student's numerical scores on the test were standardized with the mean score being 0 and a standard deviation of 1. The last variable used was socioeconomic status of the student as a reflection of the funding of the school. Students' socioeconomic status, abbreviated to SES, was broken down into 5 quintiles. Students were given a value of 1-5 based on their socioeconomic status. Status was calculated by using their parent or guardian's education, occupation, and income. Each school's urbanity (city, suburban,

town rural) was also taken into account when calculating the socioeconomic status of each student.

Variable	
Gender	Students were given two options: male or female. Males were assigned a value of 0 and females a value of 1.
Socioeconomic Status, SES	Students were broken down into 5 quintiles based on their parents or guardians' education, occupation, and income.
Math Score	Scores were standardized with a mean score of 0 and standard deviation of 1.

Table 1: All variables used in this study are listed and described in the table.

Models

To conduct this study, a linear regression model was built using R version 4.4.1 (R Core Team, 2024) with the stats package. Linear regression models are used to see if there is an association between variables and to quantify it if it's found. To capture the relationship between the variables, a line of best fit, otherwise known as the regression line, can be created. The formula of the line is based on $math_i = \beta_0 + \beta_1 * gender + \beta_2 * disrict_SES$. β_0 refers to the intercept of the equation while β_1 refers to the slope in relation to female students and β_2 the slope in relation to socioeconomic status. The model created in this study is a fitted model rather than a population as sample students were taken.

Model 1 was created by using all students with SES as the predictor and math performance as the outcome. This model allows for a broad assessment on the relationship between the variables. Model 2 was also created with all students, but this time with gender on the x-axis and math performance on the y-axis. This model was created to explore the relationship between just gender and math scores. Model 3 used both socioeconomic status and gender as variables, but did not show their interaction. Again, the math score was the predicted value. Model 4 uses the same variable as Model 3, but shows the interaction between the two predictors. The regression line for this model multiplies socioeconomic status by gender in order to plot the two variables' relationship.

To summarize how well the model predicted the outcome, R^2 was used. This value is used to express how much of the variability of the mathematics scores can be explained by the predictors.

Results

In model 1, socioeconomic status was found significant as a predictor with a p-value of less than .001. On average, math scores increased by 0.29 standard deviations for each quintile of SES. Students in quintile 3 are predicted to have an average math score. Students in quintiles 1 and 2 are predicted to have below average math scores and students in quintiles 4 and 5 above average ones. The value of R^2 for this model is .18, indicating 18% of the variance in math score is explained by student socioeconomic status. Model 1 is reflected in Figure 1 seen below.

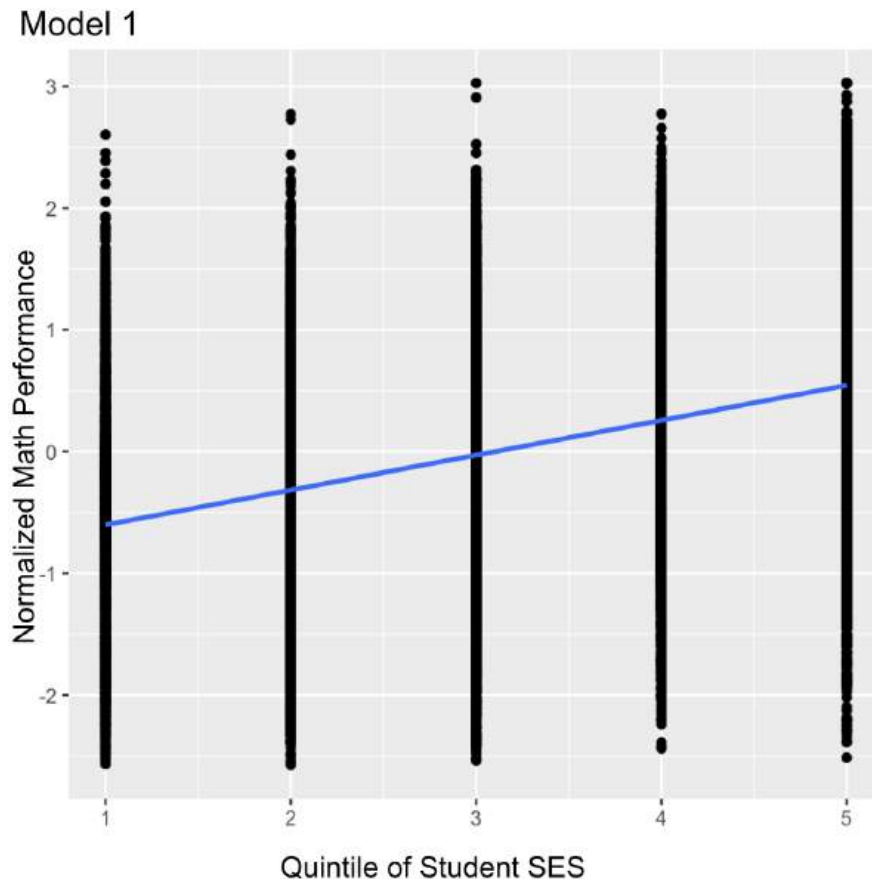


Fig 1: Scatterplot plot constructed using all student data with the regression line overlaid

Model 2 used gender as the sole predictor of math proficiency. Gender was not proven significant in this model with a p-value above .05.

Model 3 uses both gender and socioeconomic status as predictors for math score, but does not plot their interaction. This model holds the same results as Models 1 and 2 with only socioeconomic status significant as a predictor of math proficiency. Gender had a p-value of above .05 while socioeconomic status had a p-value of below .001. When interpreting the math score, one of the two variables is held constant. The slope changes interpretation to the predicted difference between two students with the same value on one of these variables and a one point

difference on the other variable. For example, if a student is male, gender has a value of 0. Their math score is based on their socioeconomic status.

Model 4 uses both socioeconomic status and gender as predictors for math score while including their interaction. In this model, both variables are significant with p-values less than .001. The value of R^2 for this model is .18 with 18% of the variance in math score explained by gender and socioeconomic status. The margins plot of Model 4 can be seen in Figure 2. In quintiles 1 and 2, females outperform males. In quintile 3, the results are roughly the same. In quintiles 3 and 4, males outperform females. The results of all 4 models can be seen in Table 1.

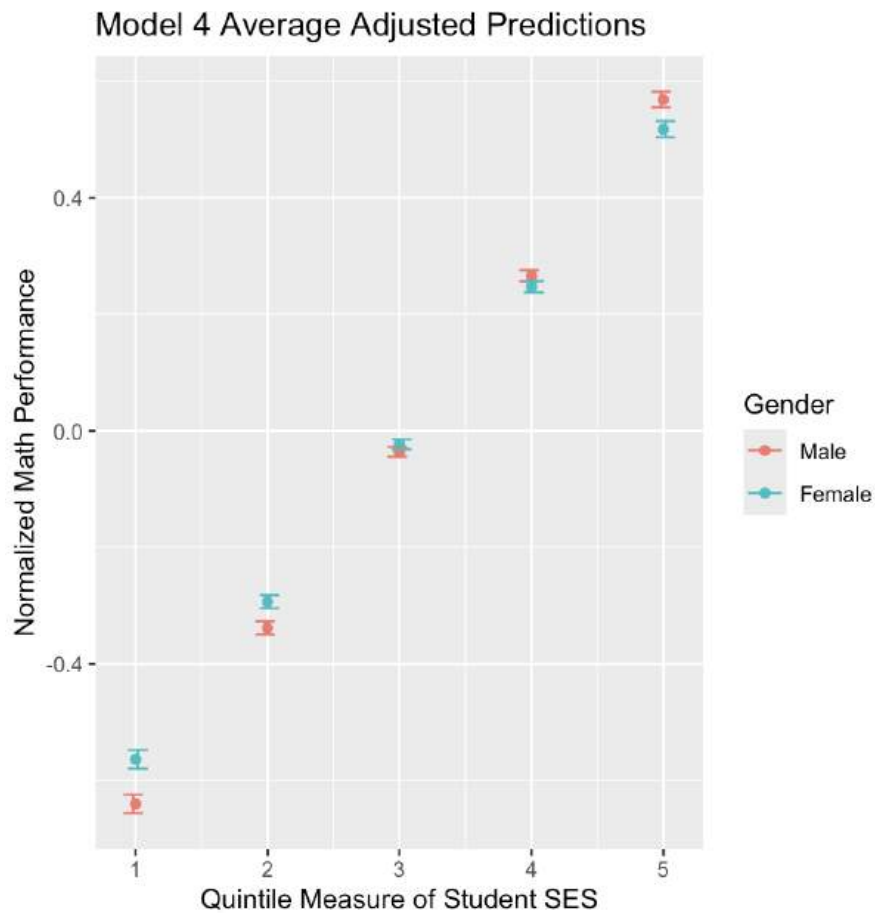


Fig 2: Regression plot using the interaction of socioeconomic status and gender to predict math performance

	Model 1	Model 2	Model 3	Model 4
Intercept	-0.89 *** (0.01)	0.04 *** (0.01)	-0.89 *** (0.02)	-0.94 *** (0.02)
Quintile Student SES	0.29 *** (0.00)		0.29 *** (0.00)	0.30 *** (0.01)
Gender		-0.01 (0.01)	-0.01 (0.01)	0.11 *** (0.03)
Quintile Student SES*Gender				-0.03 *** (0.01)
R ²	0.18	0.00	0.18	0.18
Adj. R ²	0.18	-0.00	0.18	0.18
Num. obs.	21444	21444	21444	21444

*** p < 0.001; ** p < 0.01; * p < 0.05

Table 2: Regression table showing results from all models constructed

Discussion

The significance of socioeconomic status on math scores is further solidified by the results in this study. As seen in Model 1, socioeconomic as the sole predictor has a significant impact on all student's test scores. This supports previous research on the effects of money on education in a broad sense (Allegretto et al.; Baker et al.; Pope and Sydnor). The findings in this study signifies the importance of individual wealth on student performance. As seen in Figure 1, students in higher quintiles of socioeconomic status tend to score higher on math tests. Areas with students in higher quintiles can afford to put more focus on education and as a result have higher proficiency in mathematics. These results highlight the importance of efforts to improve education in lower income areas that are performing below the national average.

The finding that gender has no significance in terms of predicting math scores shows the success of efforts to equalize the STEM fields. These results demonstrate that a student's gender has no effect in predicting their math score. Previous studies have found that the average test scores of boys and girls are the same (Pope & Sydnor, 2010; Sartain, Lauren et al., 2021). These results show the significance of socioeconomic status as a predictor of math score. Without the addition of this variable, there is no gender gap in the national average.

The most important finding of this study is the effect of socioeconomic status on the gender gap in math scores. The previous two findings are both in agreement with previous research, but the new relationship found in this study opens the door for further improvements in education. The connection between socioeconomic status, gender, and math scores shows there is still work to be done in equalizing education. The relationship found indicates that as students' socioeconomic status increases, the gender gap switches. In lower quintiles, female students outperform male students. Male students within higher quintiles are more likely to outscore female students. Future research should be done in order to decrease the gender gap in all quintiles.

One shortcoming of this research is the inability to use direct school budgets. Socioeconomic status of the students was used as an inference of the school's funding as no variable with school budgets was available. Further research should be conducted using the school budgets directly in order to solidify these findings.

Conclusion

The gender gap in math is an issue that people have been trying to solve for decades. Over the years, the gap for the most part has equalized. Despite the mathematics gap persisting at high performance levels, girls and boys typically score around the same, on average (Niederle and Vesterlund; Pope and Sydnor). One area of research that has not been completely looked into is the effect that money has on the gap. The effect of funding on test scores is a well researched area with room for debate. Both gender and socioeconomic status have been looked into separately. Combining the two areas has led to the discovery of a whole separate problem that needs to be addressed.

Socioeconomic status was shown as a significant predictor of math scores as student scores increased with wealth. It has been found that the gender gap in math scores varies with socioeconomic status. While looking at all combined student scores shows no significant differences between genders, adding socioeconomic status as a variable increases the gap. Improvements in education in lower income areas can help decrease this gap. It has been shown that lower income students have the ability to perform well on math tests, so focusing efforts on these schools can both decrease the gap caused by gender and by wealth. Furthermore, a greater focus should be placed on students in higher quintiles in order to decrease the male favoring gender gap. Future research should be done in order to figure out the best way to improve education nationwide.

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The Role of the Glottis in Trumpet Playing By Jayden Peng

Abstract

Conducted in collaboration with California State University at Fullerton's Associate Professor Sycil Mathai, this study investigates the impact of the glottis on brass playing, specifically targeting improper glottal usage and its effects on trumpet performance. The glottis, part of the larynx, significantly influences breath control, articulation, and pitch in brass playing. Improper use of the glottis can lead to excess tension and inefficient playing, especially in the high register.

Our findings suggest that minimizing conscious glottal engagement and instead relying on the tongue for airflow control results in more effective and tension-free playing. Strategies such as practicing high-note passages with a relaxed sensation were explored as possible alternatives to glottal playing in order to mitigate improper glottal use. This project emphasizes the need for enhanced awareness and correction of glottal habits in brass pedagogy, contributing valuable insights for educators and performers.

Exigence

As an aspiring trumpet player myself, there are aspects of my technique that require improvement and continued practice in order to reach a professional standard of proficiency on the instrument. Most issues I've encountered are quite common amongst all types of brass musicians and will naturally resolve over time with standard forms of practice and diligent study of the trumpet. However, there can exist some habits which hinder trumpet playing and are not easily diagnosed since much of what happens during trumpet playing cannot be seen with the naked eye. In this way, brass playing can be challenging compared to a violinist who can visually assess any technical issues that may arise. One such habit was pointed out to me by a friend when I performed for him once during a practice session. He noted that whenever I performed octave slurs, he could hear a grunting noise emitting from my throat. While I was unaware this represented a manifestation of incorrect trumpet technique, I became curious and determined to better understand the underlying causes of this occurrence.

My preliminary research on this particular problem led to an in-depth exploration of the glottis' role while playing the trumpet. This paper aims to summarize my findings in identifying and eliminating improper glottal usage with the hope of improving overall playing techniques and clarifying this less understood issue of trumpet pedagogy.

Purpose of Study

In attempting to diagnose and alleviate my own issues with the glottis in brass playing, it became apparent that a simple answer was not to be found. While there exists some peer-reviewed works and research on the matter, there was not a general consensus- that is, in the world of brass pedagogy- about the role of the glottis and its use while playing trumpet. To bolster my efforts, I partnered with a faculty member at California State University Fullerton,

Associate Professor Sycil Mathai. We agreed that, while many trumpet players experienced similar problems with understanding the impact of the glottis on their playing, there was not much research into the effect of the glottis on brass playing. Our aim was to more thoroughly investigate the topic of the glottis with the hope of helping clarify current brass pedagogy related to this matter.

By increasing awareness of the role of the glottis, more brass players will recognize and correct possible bad habits or playing impediments, or avoid such problems before they arise.

Background

The aforementioned sound created in my throat area whilst playing trumpet was discovered to be caused by the glottis.

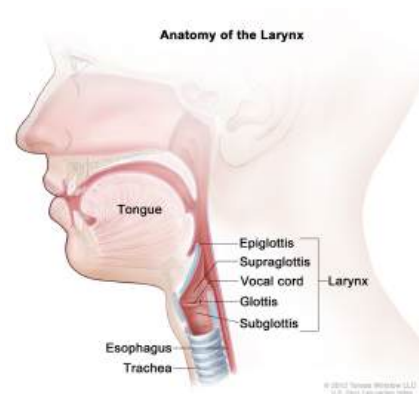


Fig 1: Diagram of location of the glottis in the human anatomy¹

For reference, one can locate and identify the glottis by saying words such as “Punalu’u” and “Ka’a’awa”; the mechanism which closes your throat in order to make the sounds indicated by the apostrophes is the glottis. You may also identify the glottis during a cough: the glottis opens suddenly allowing for a forced exhalation against a closed glottis.

The glottis is the part of the larynx (voice box) which includes the vocal cords and the opening between the vocal cords. The glottis plays an important role in creating sound for speech and singing by allowing the vocal cords to vibrate as air passes through by changing its shape and size to alter pitch and volume. The tension and spacing of the vocal cords, both controlled by the glottis, determine the pitch and volume of the sound produced. The glottis plays a protective role in breathing by regulating airflow and participating in the cough reflex, which helps expel foreign particles from the respiratory tract. Additionally, it should not be mistaken for the epiglottis which helps protect the airways for breathing by closing during swallowing, preventing food and liquid from entering the trachea.

As it relates to trumpet playing and brass playing in general, the glottis contributes to regulating airflow and pressure in activities during brass instrument playing. It affects several functions in playing including: articulation, tone, dynamic control, pitch control, vibrato control, and breath efficiency. In other words, one's overall “sound” on a brass instrument. Proper control

of the glottis allows for efficient use of breath, helping to sustain notes and play longer phrases in music. The glottis is critical in achieving a wide dynamic range and producing clear articulations in musical performance. However, if the glottis is used incorrectly, a brass player might feel his/her throat closing off in the high register, leading to excess tension and negatively impacting their playing. A question arises as to whether or not a brass musician should consciously control glottal movement, or to trust the body's automated control of the glottis to achieve safe and optimal control of the air speed while playing a brass instrument.

Research Question

How does the glottis impact brass playing?

In addition to carefully reviewing previous existing studies and research, Professor Mathai and I focused on an especially elucidating dissertation by Dr. Ryan Chapman titled "A Strategy for Proper Utilization of the Glottis and Implications of Improper Use in Trumpet Performance²⁷". Our interview with Dr. Chapman also provided valuable information about the glottis and how to reduce tension while playing. As a lead trumpet player, Dr. Chapman was extremely insightful in elaborating on the effects of the glottis in the high range. He recalled and shared his own experiences of feeling his throat close off when playing in the upper registers, which partly inspired him to study this topic in his dissertation. The reader should understand that the role of the glottis becomes even more integral when performing in the highest ranges of a brass instrument. The amount of compression inside the body necessary to produce these higher notes is accompanied by the need to properly produce these notes without forcing them. Bad habits, such as improper use of the glottis, not only lead to inefficient playing but can also significantly damage a musician's career due to inadequate technique. I've personally met teachers who have had to take *years* off the instrument due to an injury resulting from too much tension in the lip. Since some of the difficulties that Dr. Chapman's experience before writing his dissertation mirrored my own, I decided to pursue and apply his own techniques of glottal usage in my own trumpet playing.

Findings/Argument

It is important to understand the basic techniques of playing a brass instrument. When a person puts their instrument to his or her lips in preparation to playing it, they are likely "hearing" the exact note they will produce inside their head, or are preparing to set the air required to produce that note. If there exists any excess or unnecessary muscle movement, their playing is likely impeded. The optimal method to play is just to simply breathe in and breathe out. When the player articulates a note (using the tongue to briefly interrupt the air flow inside the mouth so that a punctuation of the tone can be heard; much akin to consonant sounds like "ta" or "tee"), they are making a minor movement that is usually tension-free to start a note. With practice this method is the more effective way to start and play notes.

In contrast, when people consciously use their glottis to start or play notes a lot more tension arises. Kaburagi et al., 2013 in their paper “A morphological and acoustic study on the effect of a trumpet player's vocal tract⁴” concluded that the opening of the glottis, known as the rima glottidis, gets smaller as a trumpet player’s pitch rises. Using the glottis to play “high notes” means forcing your glottis to close even more, causing a smaller hole for air to exit, and creating a lot more tension. Since tension is a major hindrance for the majority of trumpet players, as it makes brass instruments much harder to control and greatly increases one's likelihood of injury, consciously trying to control the action of the glottis should not be a preferred method for playing.

The tongue is much easier to train and control, and can be used to control the airstream. While some brass pedagogues advocate using the glottis to control the airstream, this methodology is, comparatively, inefficient to using the tongue for the same purposes. Since the glottis cannot be seen or easily felt, even trying to use it consciously proves difficult and relies entirely on feeling. While a small number of trumpet players have figured out how to effectively use their glottis to help their playing, trying to teach it to other people is difficult and risky due to aforementioned factors. Phillip Farkas, the esteemed former principal hornist with the Chicago Symphony, advocated for the use of the glottis in his landmark book *The Art of Horn Playing*³. However, a few months after this work was published (a word still read today by brass musicians around the world), Mr. Farkas retracted his statement and ceased using the glottis as a measure to control air speed during brass playing.

Our findings have led to the conclusion that the glottis should be used as minimally and as naturally as possible when playing a brass instrument. Many of the body’s mechanisms are autonomic while playing brass instruments and the glottis is no exception. Most musicians who perform on wind instruments have no issue with the glottis, but some do inadvertently develop bad habits with the glottis as a result of incorrect techniques they are taught or techniques they develop themselves that are unfounded in modern brass pedagogy.

Further Strategies

Dr. Chapman provided information on how to avoid such tension and minimize glottis use. One strategy that he recommended for high-note passages, where improper or proper use of the glottis becomes more readily apparent, is to practice playing while feeling relaxed, then applying that same sensation to the written music- regardless of its difficulty level. Whether this involves performing the music an octave lower or just transposing a few notes down, a player should practice the difficult passage at a point where it is not too difficult for the player to produce a desired sound and not become physically tense. Then, the player should take that feeling of being absolutely relaxed while playing, and transpose it to play the passage as written. It is important to retain that feeling of ease in a player’s head. Dr. Chapman emphasizes the importance of getting into a relaxed headspace, ensuring that playing higher notes does not feel more difficult, but rather the same, as the lower notes.

Further strategies include Max Schlossberg's Daily Drills and Technical Studies for the Trumpet⁵ exercise no. 16. Playing all three parts of the phrase with the same embouchure really emphasizes how close partials are to each other, despite the nearly two-octave jump in between them. Marissa Benedict, renowned studio musician and assistant professor of trumpet at the University of Minnesota Twin Cities, is a proponent of this exercise and has extended this exercise to G6 (two Gs above the treble clef staff). After practicing her extended version for two weeks, I observed a clear change and improvement in my approach to high notes, and without using the glottis. Before my studies with Professor Benedict, performing a concert G pitch (2nd G above the treble clef staff is an extraordinarily high note for a trumpeter to produce) seemed unfathomable to execute, but now I can play it with a full tone, albeit quietly, almost every day. Playing without tension is crucial, and proper use of the glottis makes this endeavor much more possible.

Conclusion

The jumpstart to this project was my own experience with incorrect glottal use, which caused me to want to know more about my playing inadequacies. This project attempted to answer the question, "How does the glottis impact brass playing?" The findings show that the glottis should be used as minimally as possible in brass playing. Excess use of the glottis would cause a surplus of tension, which should be avoided at all costs. An alternative to glottal playing known as tonguing, was explored and shown to be a more effective way to play a brass instrument. This project also explored methods of avoiding tension and glottal use, combining knowledge and techniques from multiple sources. Basic knowledge of the glottis was also provided, both relating to overall anatomy and specifically in brass playing.

Much more in-depth research can potentially be performed using cameras and medical tools like Magnetic Resonance Imaging (MRI) machines. These instruments that can explore inside the body and give us greater access to how the glottis functions exactly during brass playing.

Words of Thanks

This project was a wonderful and insightful experience. I learned so much about human physiology, identified inefficiencies in my playing, discovered ways to improve my playing, and so much more. I want to give a massive thank you to Professor Sycil Mathai for dedicating so much of his time throughout the summer, and for being so supportive and open-minded throughout. I also want to thank Dr. Ryan Chapman for his valuable time and suggestions to this project; without his ideas and prior knowledge, much of this project would not have been possible.

I feel like appreciation is also necessary for those who made this paper readable, the editors: Julia Peng, Ilinca Ciubotariu, and Kelly Hur.

Thanks again to everyone who made this paper possible.

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Nusaybah and the Role of Women in Islam By Sarah Soleman

Abstract

Contrary to popular belief, Muslim women have occupied prominent roles in Islamic history, including early Islam. A prime example of such a Muslim woman is the early companion of the Prophet Muhammad, Nusayaba bint Ka'ab (d.) also known as Umm 'Ammarah. By examining her life, this paper argues that the modern female Muslim archetype requires serious re-examination. In the first section, I briefly detail Nusayabah's life and then move to examine her critical engagement with the Quranic hermeneutics directly with the Prophet Muhammad, the religion's founder. This encounter keys us into the non-passivity and incisiveness of early Muslim women. In the second section, I then discuss Nusayabah's dynamic movement between her role as a mother and warrior, showing that ideals of Muslim femininity were not strictly domestic in character. Instead, early Muslims and succeeding generations made space for and valued public and powerful feats performed by female figures, and held up such women as spiritual role models for all of society. If we take early Islamic women as authentically representative of truly Islamic mores and norms, then the story of Nusaybah patently negates prejudices that pedal the ideal Muslim women as docile and cloistered; and allows us to conceive a renewed and robust account of genuinely Islamic femininity.

Introduction

Popular Western imaginations of Muslim Women, as found in media, literature, and other mediums, have often construed them as objects of rank oppression in need of rescue by Western values and cultures. This widespread stereotype, however, fits into a longer history of Orientalist caricaturing of Muslims - often for the sake of achieving political objectives - and overlooks conveniently both the history of Muslim women who have occupied roles of significant authority and the Quranic scriptural foundations that enabled this dynamic femininity (Zine 1).

A prime example of such a Muslim woman is the early companion of the Prophet Muhammad, Nusayaba bint Ka'ab (d.) also known as Umm 'Ammarah. By examining her life, this paper argues that the modern female Muslim archetype requires serious re-examination. In the first section, I briefly detail Nusayabah's life and then move to examine her critical engagement with the Quranic hermeneutics directly with the Prophet Muhammad, the religion's founder. This encounter keys us into the non-passivity and incisiveness of early Muslim women. In the second section, I then discuss Nusayabah's dynamic movement between her role as a mother and warrior, showing that ideals of Muslim femininity were not strictly domestic in character. Instead, early Muslims and succeeding generations made space for and valued public and powerful feats performed by female figures, and held up such women as spiritual role models for all of society. If we take early Islamic women as authentically representative of truly Islamic mores and norms, then the story of Nusaybah patently negates prejudices that pedal the

ideal Muslim women as docile and cloistered; and allows us to conceive a renewed and robust account of genuinely Islamic femininity.

Nusaybah and the Quran

The existing biographical information on Nusaybah is regrettably scant but enough to portray a picture that fails to fit into popular notions of Islamic femininity as purely passive and unthinkingly submissive. This section will look at Nusaybah's intellectual incisiveness and confidence, particularly as exemplified in her engagement with the Quran.

Born in 595 CE in the city of Medina, Nusaybah bint Ka'ab was a prominent member of the local Ansari tribe and among the earliest women to embrace Islam after the Prophet Muhammad migrated to the city in 622 CE. Following her acceptance of the faith, Nusaybah took on the role of being a teacher for the women in her immediate community (Suleiman).

Nusaybah, like many women belonging to the Ansari tribe, was not shy of asking questions, even those concerning the Quran - believed to be God's literal Speech to the Muslim community delivered through the Prophet Muhammad. In a hadith, or verified historical report¹, Nusaybah one day approached the Prophet stating the following: "I see that everything is for the men and I have not seen women mentioned for a thing" (Tirmidhī). By asking this, she was likely alluding to the Quran's frequent descriptions of paradise that contain male-centered pleasures. The fact that Nusaybah felt comfortable raising this concern indicates that early Muslim women like herself, though reverent, did not engage in Divine Revelation uncritically. On the contrary, they were constantly seeking to refine their understanding of the new revelation. They understood that apparent meanings, like those of the descriptions of paradise's delights, required nuanced engagement and explanation, not just mindless submission.

Her concern received not only the attention of the Prophet but became the object of direct Divine attention. In response to her concern, the following verse of the Quran was reportedly revealed to the Prophet (Hamishi).

"Surely for Muslim men and Muslim women, believing men and believing women, devout men and devout women, truthful men and truthful women, patient men and patient women, humble men and humble women, charitable men and charitable women, fasting men and fasting women, men who guard their private parts and women who guard [their private], and men who remember Allah often and women who remember [Allah often]—for all of them Allah has prepared forgiveness and a great reward. [Al-Ahzaab 33:35].

In this verse, the Quran responds to Nusaybah's inquiry with a similarly stark answer: God's reward does not discriminate based on gender, no matter the action or the virtue. This is particularly captured in the Quran's repetition of each word in the female plural form for each virtue (believing men *and* believing women, devout men *and* devout women, etc.). This is

¹ found in the collection of Al-Tirmidhi, a prominent medieval Islamic historian.

withstanding the fact that the male plural form, linguistically, encompasses both men and women (Sadiqi). As such, the Quran did not simply dismiss Nusaybah's concern but responded with full attention to her grievance and an emphatic insistence on the parity of both genders in paradise.

In short, Nusaybah's particularly frank relationship with the Prophet and the Quranic scripture is indicative of the disposition of early Islamic women: reverential but curious and critical, they were dynamic individuals who clearly would not fit into gender prejudices widespread in the Western imaginary today.

A Valorous Mother

The above-mentioned verse comes in the Quranic chapter "The Parties," a chapter that deals primarily with the themes of maternity and martiality. Fittingly, these two qualities are nowhere better embodied than in the personality of Nusaybah. This section will examine her participation in the famous military encounter between the Meccans and the early Muslims known as the Battle of Uhud. In this battle, Nusaybah not only exhibited military prowess but moved fluidly between her role as a mother and warrior, entirely negating the notion that early Muslim women abided by an "Islamic" cult of domesticity (Shehata 4). Instead, they moved fluidly between their maternal and public roles, many times becoming universal role models of excellence for all Muslims.

Considered one of the most pivotal battles in Islamic history, the Battle of Uhud occurred in 625 AD near Medinah. While Muslim victory initially seemed imminent, the course of the battle shifted dramatically in favor of the Meccans when Muslim archers prematurely left their posts. Soon wounded and in a deadly situation, the Prophet Muhammad was left exposed on the battlefield, fighting for his life alongside a mere ten soldiers, one of whom was Nusaybah (Suleiman).

Though initially tending to wounded soldiers by providing bandages and water, when the battle took a dire turn Nusaybah fluidly shifted into a warrior. Without hesitation, she stepped up to defend the Prophet. Armed with a sword she found on the desert floor but lacking a shield, she positioned herself in front of the Prophet, adeptly taking down incoming horsemen and illustrating a military prowess that could only have been acquired by prior training (Ghazanfar). Her bravery was so remarkable that amidst the battle the Prophet has been quoted to have said "Who is it that could do what you do?" (Suleiman). Despite her valor, Nusaybah did not emerge unscathed; she sustained thirteen wounds, including a severe shoulder injury that would take years to heal (Younis).

More striking, however, is the fluid continuity existing between martial prowess and maternal compassion within Nusaybah. During the battle, her son was injured prompting her to drop her sword and rush to his aid, demonstrating her maternal care even in the heat of combat. Moments later, she swiftly killed the man who had wounded her son (Ghazanfar). This is wholly different from what we might observe in modern Western conceptions of femininity (and caricatures of Islamic femininity), where the military and motherly spheres are strictly segregated and immiscible. But, as embodied in Nusaybah, the fusion of the two was considered the ideal in

early Islam. The Prophet Muhammad's abovementioned testament to her seeming omnipresence on the battlefield - "wherever I turned...I saw her fighting for me" - suggests that she epitomizes not only a female model of bravery but a universal one (Shehata 4).

In all, the integrated existence of a martial and maternal spirit in the example of Nusaybah shatters modern prejudices regarding Islamic femininity. Though most notable, Nusaybah's participation in the Battle of Uhud was not her last military expedition. She would go on to participate in the Battle of Yamama in 632 AD - where she lost a hand - and the Battle of Hunayn in 600 AD (Suleiman). Moreover, after the Prophet's lifetime, her piety and prowess on the battlefield were the object of praise of other early Islamic leaders such as Abu Bakr Al Siddique and Umar bin Al-Khattab (Titilola). The fact that such eminent male leaders extol her personality reinforces further that Nusaybah's example was an ideal that was aspired to not just among women but across both genders.

Conclusion: The Quran and History

In sum, this paper has shown how the little-known story of Nusaybah provides a glimpse into the nature of early Islamic femininity. Contrary to pervasive stereotypes that construe Islam as a religion that necessarily domesticizes and limits women, Nusaybah's story - in what little is recorded - paints a radically different picture, showing a woman who, though pious and reverent, was intellectually curious and confident; possessed a unique martial prowess; and was simultaneously a compassionate mother.

Some may object that this model of femininity is anomalous when compared with the longer timeline of Islamic history. However, a serious survey of the historical record shows that she is far from an exception. In another hadith report, the closest wife of the Prophet, Aisha, praised the women of the Ansari tribe "for they did not let their shyness prevent them from deeply understanding the religion" ("Sahih"). A critical inquisitiveness about Islamic scripture and theology was a part of the broader culture of early Muslim women in Medina. Furthermore, female participation in martial roles can be found in other women such as Khawla bint Al-Azwar, who was famous for delivering a group of women to their Byzantine captors (Ghazanfar). Of course, female martial participation was not the norm in early Islam or later Islamic history. But the idea of a brave woman, when manifested in instances like Nusaybah and Khawla, was always valorized and never demeaned as unbecoming of their gender or overly masculine. Rather, their bravery is a manifestation of their being Muslim - and thus, indirectly, their femininity.

To conclude, I have argued that this paper has shown the need to critically challenge popular constructions of Islamic femininity, ranging from the Muslim woman as an object of rank oppression to the Muslim woman as a Victorian lady with an Islamic twist. Such notions are modern in their origin and are, in fact, more a reflection of Western political anxieties concerning Islam and Muslims than they are the philosophical-moral values of Muslims themselves. Indeed, to recover authentic Islamic femininity requires a return to the historical archives of early Islam and the rich 1400-year-old tradition that sprung forth from it.

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Examining the Impacts of Guided Physical Exercise and Sports on Children with Autism: A Review By Sophia Zheng

Introduction

Autism spectrum disorder in children is characterized as a neurological and developmental disorder by the DSM-V. Commonly observed behaviors in autistic children include but are certainly not limited to: deficiency in social and emotional reciprocity, failure to initiate or respond to social interactions, abnormalities in eye contact and other forms of nonverbal communication, repetitive motions and phrases, and highly fixated interests and focus (DSM-V, American Psychological Association). Due to these unique behaviors, autoethnographic research has highlighted the presence of social stigma and ableism toward the neurodiverse community, positioning special needs children and their families as “outsiders” (McMahon et al., 2019). Therefore, it is important to recognize the need for increased resources for ASD intervention and tolerance of differences.

Popular evidence-based therapeutic interventions for autism include cognitive behavioral therapy (CBT) and applied behavioral analysis (ABA) (Naglieri & Goldstein, 2013). While these methods have credibility within the professional community, psychologists such as Muller recognize the immense importance of sports and physical activity interventions for autism and similar disorders. He views participation in normal, daily activities that involve community participation as equally important as professional treatment. By giving people with ASD opportunities for everyday interactions, people can promote awareness and celebrate their differences (Muller et al., 2008). Moreover, professional intervention services might not be easily accessible for many communities. Studies have proven the existence of socioeconomic disparities in intervention service usage. Researchers measured utilization of professional services such as occupational therapy, speech therapy, daycare, and psychiatry while considering the family’s educational background, primarily language, insurance usage, and the mother’s race/ethnicity. They concluded that children whose first language is not English entered such services at a later age and received less time for individual services. In addition, families who used public insurance experienced significantly less individual intervention services compared to those who used private insurance (Nguyen et al., 2016). These disparities highlight the importance of lower-cost and more accessible forms of autism therapy, such as guided physical activity, playtime, and everyday socializing.

Sports and physical activity have been proven many times to have significant positive effects on all facets of wellness for youth. Such benefits include improving gross motor skills, preventing obesity, and developing teamwork and leadership skills at a young age (Merkel, 2013). Moreover, team sports have been shown to improve adolescents’ mental health. Participation in team sports such as soccer, basketball, hockey, etc. were found to be strongly correlated with improved mental health, especially for boys. The researchers concluded that factors such as self-esteem, sense of communal connection, and social acceptance contributed to better mental health outcomes in adulthood (Easterlin et al., 2019). Current knowledge about the

positive impacts of sports should also be applied to neurodiverse populations in order to gain a better understanding of how its benefits differ among populations and to encourage schools and communities to offer physical activity programs to autistic children whose families might not have access to expensive therapeutic interventions.

Motor Skills and Physical Fitness

Increased physical activity is necessary for autistic children because studies have shown that children with ASD were more likely to have a higher BMI than those without. On top of that, children with ASD were less likely to be physically active compared to neurotypical children (McCoy et al., 2016). This highlights the importance of promoting physical activity among the population, since sports and exercise have been proven to reduce BMI and motor skills. Additionally, researchers reached similar conclusions when observing the effects of guided physical activity for autistic children. When researchers implemented specific programs directed toward promoting physical activity among children, namely SPARK and ICPL, they found that children on both treatments improved their gross motor skills, agility, balance, coordination, and reaction time compared to the control group (Hassani et al., 2020).

The SPARK program focuses on promoting engaging and moderate levels of physical activity in young children (Dowda et al., 2005). Children who were part of the ICPL program were also found to have significantly improved fine motor skills (Hassani et al. 20xx). Moreover, research has shown that fine motor skills are correlated with higher performance in reading, language, and math in elementary school, further highlighting the importance of motor skill development for both neurotypical and neurodivergent children (Suggate et al., 2019). As sports programs directed towards autistic children grow in number, professionals and program directors should recognize the important motor skill benefits certain physical activity routines can bring. Moreover, there is promising evidence that the SPARK and ICPL programs are effective in developing motor skills, thus warranting future research and implementation in other populations.

Experts have also found structured training programs geared towards autistic children to be highly effective in improving various physical skills. The study consisted of a structured exercise program that met 3 days per week, for 13 weeks, and 60 minutes per session. Training consisted of warmups, stations (balance beam, barriers, strength exercises, hand-eye coordination), and cool-downs. By the end of the program, the 12 male participants improved their running speed and agility, 1-foot balance, jumping skills, and foot-hand coordination (Akyuz et al., 2016). Because children with ASD are more likely to have a sedentary lifestyle and be more at risk for heart disease, it is also important for sports/physical activity programs to promote overall fitness and wellness. An easy, low-cost method of exercise can be simply walking. A case study conducted on 5 young adults with ASD found that when they set goals for themselves to walk over 10,000 steps every day, they were consistently able to meet their goal (LaLonde et al., 2014). These conclusions are significant because walking 10,000 steps every day is considered enough to incur many health benefits, such as lowering blood pressure and

increasing exercise capacity (Iwane et al., 2000). Interventions such as nature walks, hikes, and tours are prospective cheap and effective ways of helping children with ASD become less sedentary and reap the benefits of daily exercise.

Social and Behavioral Benefits

Physical activity and sports can enrich multiple facets of people's lives outside of physical fitness. One such benefit of guided physical activity, and especially sports, is developing social skills and improving mental health throughout adulthood (Easterlin et al., 2019). When researchers implemented structured physical activity programs and regular physical activity programs for children with ASD and compared the results, they discovered that those who participated in the structured group improved significantly in eye contact, group participation, and relationship building skills. This was because the structured activities were designed to promote social interaction and implemented a reward system. Moreover, children were also given their own individual schedule cards to promote independence at the same time (Zhao & Chen, 2018). While another group of professional researchers did not find that their exercise-intervention group led to improvement in eye contact, they did find significant improvements in attention deficit, emotional reactivity, sleep disturbance, and stereotypical behaviors (Toscano et al., 2022). Many other research projects have concluded that structured and guided physical activities can help decrease some degree of stereotypical behaviors in children with ASD. One example of an activity that has been proven to deliver positive behavioral improvements in children with ASD is horseback riding. Children who experienced horseback riding therapy had improvements in irritability, communication, and social behaviors. The research suggests that horseback riding therapy is a viable intervention to address verbal and social behaviors commonly associated with ASD (Gabriels et al., 2018).

At the same time, other interventions involving physical activity are being developed by professionals. For instance, the TEACCH model emphasizes direct teaching of skills, adaptation of the environment to make the recipient feel safe, and usage of structured teaching experiences. An example of structured teaching could be implementing daily routines and minimizing distractions. By doing so, the TEACCH program aims to improve adolescents' and children's social skills while helping them see social interaction in a positive light (Van Bourgondien & Schopler, 1996). The TEACCH program has been successfully implemented in studies before. Researchers implemented the program on one group of autistic children, while the control group underwent regular physical activity teaching. The experimental (TEACCH) approach involved trained staff and was designed in an organized fashion. After the programs were finished, children who experienced TEACCH exhibited significant improvement in maladaptive and idiosyncratic behaviors compared to the control group (Panerai, Ferrante, & Zingale, 2002). These studies demonstrate how organized teaching programs that specialize in autism can not only get children with ASD involved with physical activity, but also help them develop social skills and reduce stereotypical behaviors.

Conclusion

Sports and physical exercise have been shown to confer numerous benefits for all children. Partaking in exercise can improve motor skills, promote physical health, prevent disease, and foster social skills. Structured physical activity and sports programs have significant potential as interventions for autism in children. Numerous studies have proven that physical activity helps adolescents and kids with ASD develop motor skills, improve physical health, foster socialization, and address behaviors commonly associated with ASD. Programs in particular such as therapeutic horseback riding and the TEACCH model demonstrate potential specific interventions for ASD. These fields warrant future research and studies, which can promote awareness within professional and educational communities. More schools, towns, and local communities should incorporate these two interventions, as well as structured physical activities in general. However, physical exercise should not be considered as a substitute for other scientifically backed forms of intervention such as ABA and CBT. Rather, physical activity can be treated as a fun activity with significant benefits instead of a ‘treatment’. Considering the growing awareness surrounding autism acceptance, implementing guided physical exercise/sports programs can prove effective in helping kids with ASD.

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The Ethics and Legal Treatment Which Should Govern Artificial Intelligence

By Aarav Das

Abstract

In the status quo, there are no solid laws and regulations on consumer artificial intelligence products/services. While some regulations are in place, one area is still very unstable: blame. As an example, when an autonomous vehicle violates a law, there is no specific party that is at fault. The company did not directly influence the occurrence of the violation at hand, and the victim is not at fault either; the car being an inanimate object cannot be blamed-- or can it be? The following paper details a method that can be universally applied to distinguish natural humans and artificial beings: the ability to perform evolution; following this is a method to enforce laws and penalize artificial intelligence, whose implementation the government could enforce.

The Problem

“The asphalt is now red and smells like corroded copper,” would be the first thing one would notice if the stench of a human corpse hadn’t gotten to them first. Looking up, one sees a driverless autonomous taxi with a squashed human underneath its tire. Other than wondering about which poor custodian is to clean up this mess, the next best thing to do is to assign blame. Someone must be held accountable for this disastrous mess, but who?

As no one is driving this car other than an Artificial Intelligence model, the one to blame is the Artificial Intelligence Model; however, jailing or assigning the death penalty to AI means literally nothing. How are we to hold AI accountable for negative actions if the same methods that would work on humans are completely obsolete on AI?

Identifying AI

Before determining any legal rule for punishment of AI, we need to redefine AI and create an element which we can apply universally to differentiate AI and Human. Using ‘the ability to have emotions’ as a differentiating factor as we used to is now useless due to it being theoretically possible for AI to have this ability.

Our origins, where we came from, determine the abilities that we have and what we can theoretically gain. We, humans, got here through evolution, but AI did not pop into existence and evolve from some microorganism; we made AI. Using this as a differentiating element is also not great however, as AI can also make AI. One thing AI cannot do is gain more abilities without thinking about it. We, humans, and other living beings on earth got to the state we are in through the evolutionary process; but no one ever intentionally evolved to the state that we are in now. If AI is to evolve, it would have to think about it, visualize the destination, and perform modifications. When translating this rule into *legal mumbo-jumbo*, we get a differentiating element that can be applied universally to distinguish a robot/AI, and living being. A proposed statement which could be applied in a court of law would be similar to, “Any being that cannot

perform evolution in a subconscious manner is considered an Artificial Being; A being that can perform evolution subconsciously, without intention, is considered a Natural Being.” This proposed standard will be referenced as IEADS (Independent Evolutionary Ability Distinguishing System) onwards.

Holding AI accountable

Upon a brief analysis of the AI manslaughter scenario initially brought up, we see the car is responsible for the manslaughter, and we can infer that the car is an artificial being. As traditional punishing methods that normally work on humans, simply will not work on AI, we need to take advantage of a weakness that AI has. As it was brought up that, theoretically, AI *has* emotions, we can attempt to manipulate these emotions. Unlike natural beings, AI does not have emotion natively. AI can *theoretically* have emotion if we give it the ability to have emotions through simulating a brain. In the [natural] human body, emotions and feelings such as pleasure and sadness are all simply biochemical reactions which occur in the brain. Removing this system in a human would bring this respective human to lack the ability to experience emotion. (An example of this are painkillers such as Ibuprofen or acetaminophen, which mess up these reactions in the brain to make one feel something completely different than one would normally.) If we add some sort of simulated reaction in an artificial being which it is forced to consult on every output, we give AI emotions.

The advantage of these emotions occurring in an artificial being is that they can be manipulated. By forcibly implanting some sort of negative memory of a possible wrongdoing and/or its punishment, we can force trauma into the brain of an artificial being. While this sounds like a theory that would be successful, there are an infinite number of things that an artificial being can do wrong, and it will take forever to program a link between every single one of these and a negative emotion, not to even mention the amount of storage space that would demand. A solution to this problem can be found through consulting a book from 1950: the Handbook of Robotics 56th edition from I, Robot.

The three laws of robotics are written by Issac Asimov; these laws dictate what a robot may and may not do. “1: A robot may not injure a human being or, through inaction, allow a human being to come to harm; 2: A robot must obey the orders given it by human beings except where such orders would conflict with the First Law; 3: A robot must protect its own existence as long as such protection does not conflict with the First or Second Law;” (Asimov Issac; ‘I, Robot’, 1950) These laws, while written over 50 years ago, are still applicable now. The three laws of robotics and the concept of artificially manipulating emotions within artificial beings can be merged to properly restrict artificial beings. A proposed statement for this concept would be: “In a court of law, if an Artificial Being violates these three laws in any fashion, the agent will be subject to torture through artificial post traumatic stress.” This system will be referenced as TAPTS (Torture through Artificial Post Traumatic Stress) onwards.

Prevention is better than a Cure

Ideally, we would want a more preventative system, where we never have to bring an artificial being inside a court at all. This concept of TAPTS is preventative if applied correctly, where, fearing experiencing this torture, the AI would steer clear from any wrongdoings in the first place. Another factor that we must implement is a hierarchy system in a court of law, which, using the IEADS, we state that Natural Beings are more important than Artificial Beings without question. As of our current technology, governing Commercial and Enterprise artificial beings will only become more important as time progresses. Applying systems TAPTS and IEADS to a court of law are basic standards which will protect, while limiting the rights of Artificial Beings, as well as protect the (natural) human

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Editing Input from Kathleen Stoker, Dean College Adjunct Faculty: Communications

Effects of Loss Aversion on Marketing By Ada Asogwa
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Abstract

Loss aversion, a vital concept utilized most in behavioral economics, describes how the pain associated with loss affects individuals more intensely than the pleasure associated with gain. Amos Tversky and Daniel Kahneman, in 1979, described this concept as a crucial part of the substructure of the prospect theory, which describes how people make decisions under conditions of risk and uncertainty. Many factors affect loss aversion, including health, race, age, gender, location, emotions, education level, and more. Loss aversion is utilized in various aspects of marketing to make a profit. Odd-even pricing, pre-purchase, freemium, and free trials are all methods used by companies for profit-making. However, there are ways to mitigate the effects of loss aversion, such as exercise and cognitive reappraisal skills, a form of cognitive behavioral therapy. Cognitive reappraisal skills are used to reframe a situation and threat, thereby changing the emotional effect of the perceived threat or situation.

Introduction

Before the late 1900s, the Expected Utility Theory was the most dominant model to explain decision-making under risk and uncertainty; however, individual behavior in experimental and market settings differed from the prognoses of the Expected Utility models. The breaches of the Expected Utility theory were listened to when many alternatives to the Expected Utility theory started to emerge, one of the most notable ones being the Prospect Theory (Kroes et al., 2009). The Prospect theory, developed by Daniel Kahneman and Amos Tversky in 1979, was introduced as an alternative to the Expected Utility theory and a different point of view on thinking about money, goods, and risk (Camerer, 2005; Kahneman and Tversky, 1979). Kahneman and Tversky in 1992 described a new variant of prospect theory called cumulative prospect theory, which explains how individuals make decisions under risk and uncertainty (Tversky and Kahneman, 1992). For most people, losses carry much more weight than wins. People tend to have a more extreme pain towards a loss than a sense of pleasure towards a gain. This occurrence is called loss aversion in behavioral economics (Gu et al., 2022). This topic has been discussed a lot in the research community, with it being considered as early as 1930 by Cason. When loss aversion is explained in terms of the utility function shape, a loss has a sharper utility than a gain, showing an experience more painful than satisfaction (Wakker and Tversky, 1993).

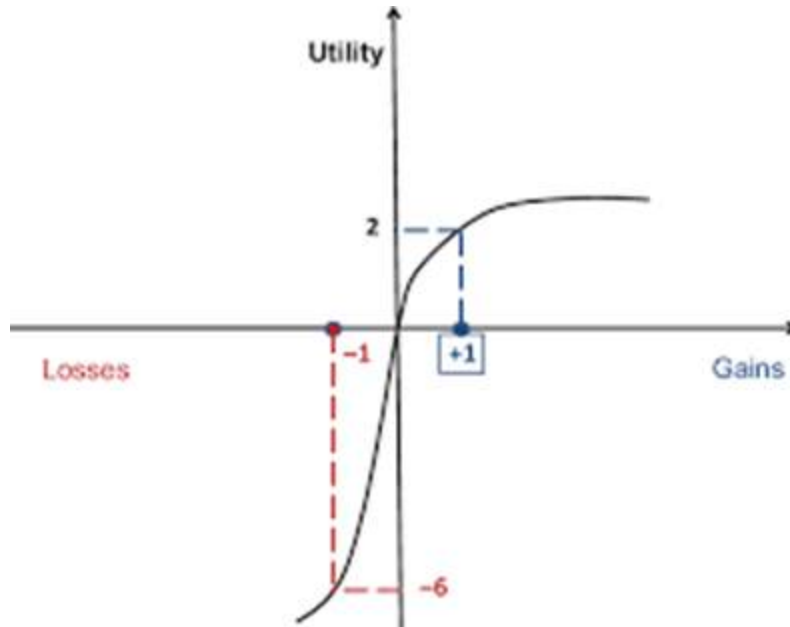


Fig.1: Loss aversion. This utility function displays the outcomes for gains and losses. This explains why losing an amount of payoff 1 provokes much more distress (-6) than the satisfaction caused by gaining the same amount (+2) (adapted from Kahneman and Tversky 1984).

Since loss aversion was first established, many contributions have been made to the topic, such as the endowment effect first introduced by Richard Thaler (Thaler, 1980). The endowment effect is the likelihood for individuals to inflate the value of the product they possess. The feeling of pain when dealing with a loss is greater than the pleasure felt when dealing with a gain. Since sellers usually think of selling products as a loss of something they possess, and purchasers think of buying something as a gain of something that they did not have, sellers start to demand more reimbursement of the product since they suffer more from the loss due to loss aversion (Morewedge et al., 2009). Other authors have also reported a reversal of loss aversion when a small amount of money is involved. Individuals prefer the pleasure of gain to the pain associated with loss (Harinck et al., 2007). The reversal of loss aversion is based on the hedonic principle, which proposes that small gains are preferable to small losses (Kahneman et al., 1999; Higgins, 1997).

Many distinct factors affect loss aversion. These factors include age, gender, education level, location, and more (KILIÇ and ACAR, 2020; Hjorth and Fosgerau, 2009.). Among these factors, age is a crucial factor that affects loss aversion; as individuals grow older, they tend to become more affected by loss aversion (Gu, 2022). Studies show that loss aversion significantly impacts consumers' decision-making processes when selecting products to purchase (Heidhues and Koszegi, 2004; Fogel et al., 2004; Karle et al., 2015). However, there are ways for consumers to mitigate the amount of loss aversion that affects them when choosing a good to buy. Loss aversion has been shown to prove why consumers choose the products they usually do based on the person's reference point. When a consumer sees a product that is below their set reference point, which they already own, they become upset since they could have purchased the same

item for a lower price. This knowledge is frequently used by firms as they mark down prices to make more consumers buy the product they are trying to sell (Kuksov and Wang, 2014). A consumer's reference point can be determined from a variety of factors like their socioeconomic status, cultural background, and upbringing. This is why loss aversion varies from person to person (Neumann and Böckenholt, 2014). The purpose of this paper is to analyze the effects of loss aversion in the market.

Factors Affecting Loss Aversion in Marketing

The effect of loss aversion varies from individual to individual. This variation has been studied by many researchers, and it is dependent on social, cultural, and demographic factors. These factors include age, health, race, life events, gender, education level, economic status, and location (Gu et al., 2022; KILIÇ and ACAR, 2020).

2.1 Education Level

It has been proven that a consumer's education level plays a role in decision-making in marketing and investing. Some studies reveal that a consumer with a higher education level is likely to show less loss aversion compared to an individual who is less educated (KILIÇ and ACAR, 2020; Hjorth and Fosgerau, 2009). Well-educated investors consider numerous factors before making investment decisions (Livamägi, 2016; Grable, 1997; Fachrudin and Fachrudin, 2016). There is a relationship between decision-making biases and educational background. These reported biases decline with rising educational levels (Nguyen and Schüsler et al. (2012)). The department of study can affect an individual's investment behaviors. Graduates from the school of medicine do less stock trading, natural science graduates make higher stock trading, whereas studying economics does not affect stock trading decision-making and may lead to less control and overconfidence bias (Livamägi, 2016; Loerwald and Stemmann, 2016). Other studies show that having a financial education gives investors the ability to control emotions and the ability to make rational decision-making (McKenzie, 2009; Altman, 2012). However, financial education alone is not adequate for making financial decisions, as other factors, such as psychological, emotional, and cognitive biases, play a role in decision-making (Katarachia and Konstantinidis, 2014).

2.2 Age

Age has a positive relationship with loss aversion. People get more loss-averse with increasing age (Hjorth and Fosgerau, 2009; Ye, 2021). Loss aversion increases until age 55 when it rapidly declines. It was also shown that younger people below the age of 25 are significantly less loss-averse than individuals above 55 years (Hjorth and Fosgerau, 2009). Older people tend to buy products during discounted periods. This is due to fear of missing out on the discounted price and caring less about the quality of the product. Other studies support the increase in loss aversion with age and income and the decrease with education (Thaler, 1980; Arora and Kumari, 2015; Johnson et al., 2006). The observation of more loss aversion with age is because there is

less time to recover if there is a loss (Arora and Kumari, 2015). Primary effect and memory interference are other explanations for elderly individuals being more loss-averse than younger individuals. They focus on the initial piece of information acquired regarding a product and usually give it more value (Aggarwal and Zhang, 2006; Arkes, 1991; Lin and Lee, 2002). Studies indicate that the elderly are more likely to remember less information regarding a product than younger individuals (Cole, 1983; Stephens and Warrens, 1983; Zeithaml, 1982). The intensity and correctness of searched information also decrease with age (Cole and Balasubramanian, 1993). The volume of ectocinerea and grey matter mass in the posterior parietal lobe decreases with age. An inverse relationship exists between the ectocinerea volume, grey matter mass, and loss aversion (Grubb et al., 2016; Adler et al., 2016). The cause-effect relationship between age and loss aversion has not been established, and it is noted that physical exercise reduces the rate of grey matter loss in the parietal lobe, hence mitigating the effect of loss aversion (Gu et al., 2022).

2.3 Gender

Studies show that women are more likely to be risk-averse than men (Croson and Gneezy, 2009; Eckel and Grossman, 2002; Prasad and Mohta, 2012; Schmidt and Traub, 2002). A study found that there is a clear difference between the levels of biases displayed between males and females. Measurement errors resulting from various definitions of loss aversion can lead to variable outcomes of gender effects on loss aversion. There have been debates in the literature to which extent gender affects loss aversion, with various outcomes on gender's effect (Schmidt and Traub, 2002; Brooks and Zank, 2005; Abdellaoui et al., 2008; Harrison and Rutstrom, 2009; Booij et al. 2010; Gächter et al., 2010; Holden, 2014; Andersson et al., 2016). Schmidt and Zank (2005) reported disagreement on the definition of loss aversion in the literature; these variations can result in measurement error with the contradictory effect of gender on loss aversion (Bouchouicha, 2019).

Applications of Loss Aversion in the Market

Firms extensively use loss aversion in their marketing strategies to keep customers coming back to buy more of their products. There are different tactics used by firms, to engage customers, and make them use their products/services. These strategies include free trials, freemiums, odd-even pricing, and pre-ordering options for new products (Gu et al., 2022; Shi et al., 2019; Lin, 2021; Booij and Van de Kuilen, 2009; Holdershaw et al., 1997).

3.1 Free Trials/Freemiums

In the past years, a marketing strategy called "Freemium" has been popularized and entails offering low-end products at zero prices. It is popular among firms that have virtual products such as online games, computer software products, online streaming platforms, and other online services (Kumar, 2014). It has been reported that companies like Dropbox, Skype, Netflix, and Spotify have used this marketing strategy to get customers. In the freemium model, lower-quality (basic) products are free, whereas the premium product can be purchased. In the

free-trial model, premium products and services can be accessed for free but for a limited period. This offer creates a sense of ownership, leading to psychological endowment (Paraschiv and L'Haridon, 2008). Following a free trial, there is a sense of loss in the consumer who no longer has access to the service/product. This feeling of loss by consumers often drives them to purchase the product/service. Many customers take advantage of the firm's freemium and free-trial services, ultimately expanding the firm's customer base. Initially, there is satisfaction with using the free products, but the pain that comes when the free version ends leads to the purchase of the product, thereby bringing more revenue for the company (Shi et al., 2019).

3.2 Pre-purchase

The pre-purchase option, available in various markets, presents the consumers with the option to order an item before it becomes available in the market. The consumer can make a pre-purchase in advance or buy on the spot, usually close to the time of consumption or use. Factors like work schedule, weather conditions, and consumer's state of mind create uncertainties among consumers who want to participate in advance purchases. These uncertainties usually get resolved while making a purchase close to the time of use or consumption (spot-market stage) (Dana, 1998). The reason for advanced purchase in loss-averse customers is due to fear of paying more in the future and discontent with not consuming the product. In other words, the aversion to monetary loss and consumption loss drives the behavior of making a pre-purchase without due consideration to the value of the products. Numerous studies show that consumer reference points can rapidly shift based on the most recent belief, leading to expenditures that can exceed the expected value of the product (Lin,2021; Smith, 2019; Song, 2016). For example, informative advertisements create an attachment to a product and hence lead to a new reference point. This variable has a persuasive effect on a loss-averse customer, making her more willing to overpay for a product (Karle, 2013). The reference point could be static after it is formed, leading to a reversal of the loss aversion effects and less willingness to make an advanced purchase (Karle and Möller, 2020).

3.3 Odd-even pricing

The price of a product contributes to its popularity. Companies are using odd-even pricing to attract consumers. In an odd pricing strategy, the last digit in the pricing is an odd number instead of an even number. For example, instead of pricing a product for 4.00 dollars, the price is set at 3.99 dollars. Customers consider a product priced at 3.99 dollars to be cheaper than one priced at 4.00 dollars. Consumers usually feel more satisfied and do not feel any loss purchasing a product a cent or dollar less. The strategy of buying a product at odd pricing is desirable to consumers and leads to more purchases, as customers feel they have engaged in a bargain. Holdershaw et al.,1997, report that customers pay more attention to the first number rather than rounding the number upwards, as it requires a cognitive process. He further reported that over 90 % of advertised products have an odd number pricing.

Mitigation in Loss Aversion

Many mitigation steps for loss aversion are based on different interpretations of loss aversion. These explanations of loss aversion include the feeling of ownership, the tendency to consider loss before gain, and preoccupation with loss (Booij and Van de Kuilen, 2009). Other explanations are confirmatory search and bias hypothesis (Carmon and Ariely, 2000) and psychological, physiological, and physical tendencies to react more to loss than gain (Cohen, 2016). In other studies, loss aversion is described as due to a psychological inclination towards loss rather than gain, or affective prediction error, which occurs when the loss is overestimated (Haigh and List, 2005).

Memory and preference construction explains that loss aversion is moderated by what the order individuals consider loss and gain. Usually, the loss is considered first before the gain. Reminding the individual of the gain first may mitigate the loss aversion (Johnson, 2007). Other studies of preference construction show that individuals with less education, knowledge, and experience of specific attributes can better construct preferences than those with more knowledge (Ye, 2021).

There are conflicting findings in studies of property ownership. Experienced investors or investment institutions are as much loss averse as inexperienced investment firms, making a case that experience has no role in loss aversion (Bokhari and Geltner, 2011). However, there is a report that experienced dealers of sports cards and sports memorabilia are less loss-averse when compared to inexperienced dealers (List, 2000). The other conflicting report reveals that experienced stock and options traders have greater loss aversion than inexperienced stock and options traders (Haigh and List, 2005).

The cognition capacity of an individual has control over affection (emotions). Individuals with better cognitive ability can make better choices, keeping their emotions in check (Paraschiv and L'Haridon, 2008). Intentional cognitive regulation strategy emphasizes perspective-taking. This is the basis of the "trader's mindset," which alters choices and reduces behavioral loss aversion, and arousal to losses relative to gains. This perspective of loss aversion reduction is based on previously reported evidence that emotion plays a role in decision-making (Lerner et al., 2004; Winkielman et al., 2005; Mellers et al., 1997; Mellers et al., 1999; Murray, 2007); that decision-making ability is modified by the expectation of an emotional response to loss (Shiv et al., 2005), and the physiological arousal responses to loss relative to gain is linked to a loss aversion (Sokol-Hessner et al., 2009). Many research works show that through affective and cognitive regulations, a trader can reduce loss aversion associated with trade goods or currency (Gneezy and Potters, 1997; Ariely et al., 2005; Novemsky and Kahneman, 2005a; Novemsky and Kahneman, 2005b).

Emotions are generated by thoughts and appraisal and changing the appraisal (reappraisal) can lead to emotional change (Lazarus, 1982; Pettinelli, 2012; Schachter and Singer, 1962; Gross, 1998). This concept led to the development of cognitive reappraisal skills; a form of cognitive behavioral therapy designed to reframe a situation of threat to alter emotional impact (Gross, 1998). Reinterpretation (reappraisal) is used to give a different interpretation of a

situation or stimuli aiming to alter the emotional effect. The stimuli or situation remains the same but is perceived differently or put in a bigger context that changes its meaning. Reinterpretation, a mitigating strategy to loss aversion, enables an investor or trader to believe he is experienced and not an amateur (Sokol-Hessner et al., 2013) or to choose a broad and better outcome rather than a narrow outcome (Read et al., 2000). An example of decision-making based on broad bracketing rather than a narrow one will be buying stocks rather than bonds, as it brings more returns and creates more wealth. The ventromedial and dorsolateral cortex are involved in the regulation of emotion. At the same time, the amygdala and striatum represent value and process the emotion (Ochsner and Gross, 2008). The amygdala also plays a role in the regulation of emotions and decision-making (Nejati et al., 2021; Hertrich et al., 2021). Amygdala responses to gain and loss correlate with choice regulation (Sokol-Hessner et al., 2013) and estimation of behavioral loss aversion (Sokol-Hessner et al., 2013; Weber et al., 2007). The changes in blood oxygenation level-dependent (BOLD) activities during reappraisal in the striatum, dorsolateral, and ventromedial cortex are like findings of other studies on emotional regulations (Ochsner et al., 2004; Eippert et al., 2007; Ochsner et al., 2002). Loss of amygdala function is linked to the reduction of loss aversion (De Martino et al., 2010). This suggests that the amygdala has a role in the endowment effect and control of loss aversion (De Martino et al., 2010; Weber et al., 2007).

Emotional stress affects decision-making and loss aversion (Starcke and Brand, 2012; Margittai et al., 2018). These effects are explained by two conflicting hypotheses, the "salience-of-losses" and the "alignment hypothesis." The salience-of-loss hypothesis proposes an increase in loss aversion through the activation of the salience network by stress. This network contains regions around the amygdala, a notable neural basis of loss aversion (Metz et al., 2020; Molins and Serrano, 2019). The more popular hypothesis, "The alignment hypothesis," suggests that stress leads to dopaminergic activation in the nucleus accumbens of both rats and humans (Metz et al., 2020; Anstrom and Woodward, 2005; Kalivas and Duffy, 1995; Scott et al., 2006; Wood et al., 2007), which ultimately lead to reward-biased learning. In summary, the alignment hypothesis suggests that stress leads to more attraction to gains than loss, thereby reducing loss aversion (Metz et al., 2020). Other research work supports the alignment hypothesis and suggests that stressor, regardless of their emotional impact, leads to a reduction in loss aversion (Molins et al., 2021).

Conclusion

This paper discussed the factors that affect loss aversion, the applications of loss aversion in the market, and how loss aversion can be mitigated by individuals. Loss aversion, as described in 1979 by Kahneman and Tversky, became the framework of the prospect theory. It explains the phenomenon in behavioral economics where individuals are more affected by losses than gains. Shortly after, in 1980, Richard Thaler proposed the endowment effect as an explanation of loss aversion and proposed that people tend to inflate the value of the product they own. In 1992, Kahneman and Tversky later introduced a new variant of the prospect theory called cumulative

prospect theory that explained how individuals made decisions under risk and uncertainty. The effects of loss aversion can be reversed according to the hedonic principle, which says small gains are preferable to small losses when a small amount of money is involved. Many factors affect loss aversion, including gender, health, age, economic status, and more. Loss aversion is used in the market by odd-even pricing, free trials, pre-purchasing, and freemiums. Companies can use these strategies to make a profit. There are various factors that mitigate loss aversion, such as exercise, cognitive reappraisal skills, and adopting a "trader's mindset," which can change an individual's choices and reduce loss aversion. There is potential for research on the topic of loss aversion, especially on mitigation strategies for different age groups, particularly teenagers.

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The Value of Biomechanics in Understanding Tommy John Surgery Amongst Major League Baseball Pitchers By Gabriella LaSpisa

Abstract

In recent years, the rates among Major League Baseball (MLB) pitchers having to undergo Tommy John surgery have increased dramatically. As this procedure aims to repair a torn ulnar collateral ligament (UCL) within the elbow region, these growing numbers have been found largely due to the repetitive stress the pitcher's arm endures during their pitching motion. Analyzing the biomechanics of these professional pitchers that are responsible for this repeated strain is essential in understanding the role of biomechanics in preventing UCL injuries. With these findings, clarification on how to reduce the overall need for Tommy John surgery can be discovered. The ultimate goal of this research is to identify movement patterns between the biomechanics of MLB pitchers who had Tommy John surgery and those who have never torn their UCL. It was hypothesized that there would be an existing difference between the experimental and control group of this study regarding their pitching motions. In order to collect the quantitative data needed for this study, a content analysis was conducted. Participants included forty MLB pitchers. Both the experimental group of former Tommy John patients and the control group of in-tact UCL pitchers had a total of twenty pitchers, ten being right-handed and the other ten being left-handed. In addition, nine 2-Sample Z-Tests of Proportions and three Chi-Squared Tests of Independence were used to determine any statistical differences of the biomechanical patterns observed between the experimental and control groups. Although the Chi-Squared Tests yielded insignificant results, six of the Z-Tests revealed a p-value less than 0.05, allowing the rejection of the null hypothesis. Specifically, these significant results pertained to the elbow to trunk angle and the relative location of the elbow at the release point between both groups of pitchers. By gaining this new comprehension, modifications to the pitching mechanics of professional baseball pitchers can help minimize the stress experienced by the UCL.

Introduction

For generations, the sport of baseball has been a large part of American culture. From the evolution of amateur to professional leagues, baseball has developed in various areas. Advancing technology has changed many of the fundamentals involved with playing the game while maintaining excellence, more commonly known as biomechanics. Biomechanics can be defined as the biological mechanics of the body in response to a form of impact or displacement (Saxena, 2009). In recent years, the biomechanics of professional baseball pitchers has been well documented as sports medicine specialists have had increasing interest in analyzing pitching motions and its effect on the ulnar collateral ligament (UCL) found in the elbow. Pitching motions are complex sequences of segmental interactions within the pitcher's body that allows them to generate enough kinetic force in order to create a transfer of momentum to the baseball being thrown (Hamer et al., 2021).

The UCL has been acknowledged to be a significant contributor to the impairment of a pitcher's ability to compete at their regulated level if injured. Injuries to the ligament vary in severity from the least, such as sprains which do not require surgical intervention, to the worst, such as a complete tear in need of reconstruction. The scientific name for this surgical procedure needed to repair the torn ligament is Tommy John surgery, named after the first patient to undergo this surgery in 1974. The success story of predominant pitcher Tommy John for the Los Angeles Dodgers, a professional baseball team of the Major League Baseball organization, began the future direction for understanding and recognizing elbow injuries in pitchers (Looze et al., 2018). Building off this expansion of knowledge, the researcher will be looking into the biomechanical factors responsible for causing UCL injuries in need of Tommy John surgery and how this can be applied to preventing surgical intervention by indicating injuries early. In this study, the researcher intends to answer the question; To what extent does the comparison of the biomechanics in professional baseball pitchers offer data specialists can use to prevent ulnar collateral ligament reconstruction?

Literature Review

In order to understand how trauma to the UCL can occur, a basic comprehension of its anatomy must be established. According to Steven Daniels and his team of musculoskeletal radiology specialists, the UCL is associated with the region of the medial elbow joint, the most common area for elbow injuries, and distinctly consists of the anterior, posterior, and transverse bundles (Daniels et al., 2019). It is, however, the anterior band that is most susceptible to the stress generated from the pitching motions. Specifically, the anterior band experiences what is called valgus stress, a force that causes the band to move away from the body's midline and its standard position. A high valgus stress load places tremendous stress across the tendons, ligaments, and bones of the elbow region. This band, however, pushes back with what is known as varus torque. Toshimasa Yanai, a professor at Waseda University's Institute of Baseball Science, explains how varus torque is produced by the joint capsules of the UCL and can only withstand up to 35 newton meters (N m) of joint failure (Yanai et al., 2023). Nevertheless, pitchers endure the highest value of 50-120 N m during their pitching motion, a significant amount beyond the failure of their joint. A player's pitching mechanics is defined as a sequence of muscular contractions and body movements looking to produce a pitch of high accuracy and velocity (Calabrese, 2013). This leads me to my hypothesis that the biomechanical factors exerting the most valgus stress will be responsible for the harmful UCL injuries, and it is these that should be further observed in order to prevent the complete tearing of the ligament. Based on Taylor Tuff, a sports specialist chiropractor, this endurance of joint failure past its maximum capacity results in a tissue load that exceeds the tissue's capacity (Tuff et al, 2020). As highlighted by Tuff, it can be reasoned that valgus stress puts the pitcher's UCL in a moment of weakness during their pitching motion.

Although sports medicine technology has been advancing over the years, accumulating research has raised questions about the risk factors associated with tearing the UCL. A study by

Elizabeth Hibberd, director of the University of Alabama's Athletic Training Program, advances notation that a tear in the UCL is the leading injury for the need of reconstruction in the form of Tommy John surgery (Hibberd et al., 2015). The specific biomechanical factors, however, that are principal valgus stressors to the pitcher's UCL are in need of further study and clarification. Additionally, when looking into the biomechanics pertaining to this issue, Tuff's study found the responsible biomechanics for UCL tears to be varied, or misunderstood, the most in the group of professional baseball players (Tuff et al., 2020). Due to the results of this study, there are several motivations for inquiring into professional pitchers as compared to adolescents. Adolescents have skeletally immature bodies that can potentially form adaptations to the unnatural pitching motion of baseball, making it too much for the body to handle (Salamh et al., 2020). As a result, the greatest research effort on this issue has revolved around younger ages. It is, however, professional baseball players who have left their elbow at its most vulnerable state due to years of overuse and playing at the highest level of competition able to be analyzed (Tuff et al., 2020). Jeremy Bruce and James Andrews, two of the world's most celebrated American orthopedic surgeons, state how the leading cause of UCL injury is from the misunderstanding of what biomechanics place stress on susceptible parts of the body, like the UCL (Bruce & Andrews, 2014). Therefore, the goal of this study is to more explicitly define what notable biomechanics put the careers of professional pitchers at risk.

Pertinent to the misunderstanding of responsible biomechanics that damage the UCL, a study by Christopher Ahmad, head team physician for the New York Yankees professional baseball team, puts forth that almost 49% of baseball pitchers believe the UCL injury can be avoided at the professional level while many others remained neutral or unsure of their answer (Ahmad et al, 2019). In addition, this study shares differing opinions amongst professional pitchers on what biomechanics make them prone to injury. In this discussion, the variation found within the opinions of these pitchers describes an absence of education in this area of sports medicine. Professional pitchers who do not understand the toll their own mechanisms take on their body will cause them to ignore certain factors that may be heightening their risk for injury and potentially ruin their career. Research up to this area fails to pinpoint the recovery time a patient of Tommy John surgery must endure, as there remains a wide gap in return time ranging from 3-72 months (Looze et al., 2018). Despite the increasing success rate of this surgery from different techniques being modified, the post-surgical effects involved in achieving adequate repair of the UCL has just as significant of an impact as the injury before. A study by Tyler Hammer, biomechanist for the New York Mets professional baseball team, supports this idea as it has been found that the reconstructed UCL is more stiff post-surgery compared to pre-surgery (Hamer et al., 2021). As a result, the upper body's range of motion, the degree at which a joint has the ability to complete its full spectrum of movements (Schoenfeld & Grgic, 2020), will become permanently impaired. Moreover, the work of Maxwell Albiero and his fellow instructors of the Orthopaedic Surgery Residency Program at Western Michigan University is pertinent to this school of thought. Their study found many cause and effect relationships within the body that have to do with certain biomechanics relating to one another within not just the

upper body but the lower body (Albiero et al., 2023). These outcomes suggest that certain biomechanics in the upper body can have other detrimental effects on additional areas of the upper body and even the lower body. These findings are highly critical as it shows how relying on surgery will not completely restore a pitcher's greatness.

These outcomes can be generalized as to why attention should be directed towards using the biomechanics of professional pitchers in order to prevent surgical reconstruction. Sports medicine physician, Jason Zaremski, presents his findings that baseball players, specifically pitchers, compared to a variety of different American athletes in sports such as football, basketball, and even softball, experience the greatest amount of UCL injuries in need of surgery. In particular, out of the 136 patients they evaluated, 101 of them were baseball players and 54% of these players had to undergo Tommy John surgery (Zaremski et al., 2017). Despite the rising numbers of injured players, prevention has become increasingly difficult over the years. For example, many surgeons explain how baseball, which was once a seasonal sport, has become year round as travel teams and sponsored tournaments have begun to increase in popularity (Bruce & Andrews, 2014). This increase in popularity and dedicated time to the sport throughout the year has led to increasing difficulty in controlling the rates of detrimental injuries.

The existing body of knowledge about professional baseball pitchers and how we can utilize their biomechanics to inhibit damage in need of surgery is extremely limited. The growing body of evidence has been centered around pitchers in their years of adolescence (Salamh et al., 2020). Concentrating on Major League Baseball (MLB) pitchers can expand current knowledge in the therapeutic and rehabilitation fields in order to improve the abilities of recognizing injuries early and reducing any possibility of re-tearing the UCL after surgery. There has been little investigation done on the topic of prevention of UCL tears and the information that can be gathered from the biomechanics of professional pitchers in order to help sports medicine experts. The researcher aims to fill in this gap and answer the following question; To what extent does the comparison of the biomechanics in professional baseball pitchers offer data specialists can use to prevent ulnar collateral ligament reconstruction?

Methodology

In this study, a content analysis, distinguished in two parts, was conducted. A content analysis aims to discover themes and patterns within a set of texts that enables researchers with the ability to make inferences. In order to properly determine the biomechanics correlated with UCL tears, a content analysis involving quantitative data would efficiently gather the necessary information pertaining to the two variables of this inquiry. The MLB pitchers used in this analysis were divided into two variable groups; those that have undergone Tommy John surgery and those with a clean UCL medical history. This method pertained strictly to MLB pitchers that have had UCL surgery performed on them within the last six years, the latest being 2018 and the most recent being 2023.

Many different methods were considered for this study, however, a quantitative approach for the researcher's content analysis was determined to be the most appropriate. Previously, a

meta-analysis was considered for this particular study. Although a meta-analysis deems to be a good choice as the researcher examines data for patterns in other studies, the researcher was not formulating new principles on the observations being made. The researcher was instead using already existing principles on the mechanics of baseball pitchers in order to generate new applications for them in the rehabilitation field. A factor analysis was also considered applicable for this study by the researcher, but it was later rejected as the number of variables involved in such a method was not necessary to generate the desired results. Since the researcher did not have direct access to these high status baseball pitchers, conducting a survey to ask about their medical history, recorded measurements, and what the cause for their tear was was not feasible. Therefore, the quantitative content analysis was implemented by the researcher.

To begin with, the first portion of the content analysis was conducted. The first portion specifically had to do with collecting qualitative data by interpretation, and eventually converting this data into quantitative proportions to be used in statistical analysis. By performing the primary portion, observations by the researcher were made and recorded deliberately. In order to gather the data needed to proceed with this study, the researcher collected a variety of videos through an online media platform called YouTube. The observations made were based on five naturally occurring mechanics that could be visually observed within these videos; arm slot (Figure 1), elbow to trunk angle (elbow abduction) (Figure 2), external rotation above the shoulder plane (Figure 3), relative location of the elbow compared at the release point (Figure 4), and the position of the front foot's landing (Figure 5). These were specifically chosen as the greatest load of valgus stress to the elbow occurs during these motions (Daniels et al., 2019).

Figure 1



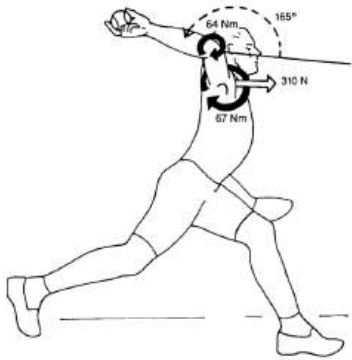
(Lehman, 2021)

Figure 2



(Competitive EDGE PT, 2017)

Figure 3



(Reinold, 2022)

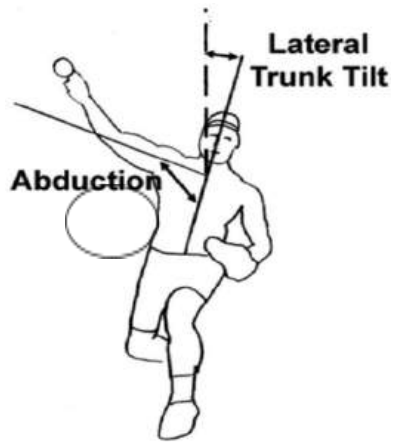


(Klein, 2019)

Figure 4



(Schad, 2017)



(Driveline Baseball, 2018)

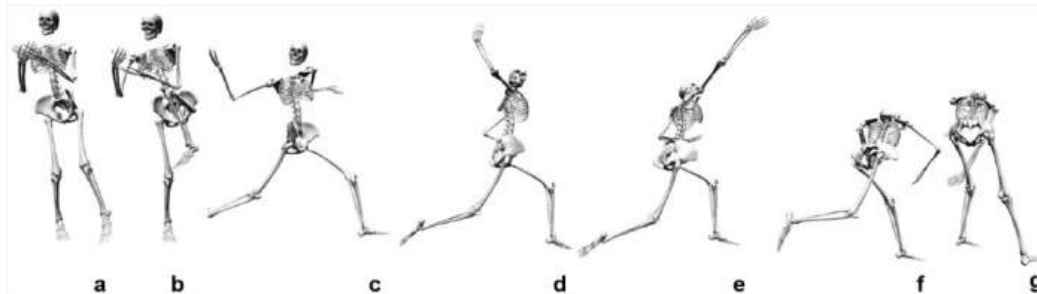
Figure 5



(Christoffer et al., 2019)

There are multiple stages within the throwing motion of a pitcher. Particularly, there are six phases as displayed in Figure 6: (a) wind up, (b) stride, (c) cocking, (d) acceleration, (e) deceleration, and (f) follow through. Phase “G” represents the end of the pitch.

Figure 6



(Chalmers et al., 2017)

The maximum amount of valgus stress on the elbow is found during the late cocking and acceleration phases of an overhead throwing athlete. That being the case, the three phases that were most vital to analyze within the videos were the cocking, the acceleration, and the deceleration phases. These phases are where the previously mentioned mechanics that have the ability to be identified within videography can be found. Firstly, the cocking phase begins when the front foot of the pitcher lands and the hands separate. This places the pitcher in what is known as equal and opposite position, as both the throwing and glove arm are mirror images of each other. The equal and opposite position is illustrated in Figure 2. At this point, elbow valgus torque, or the bending of the elbow to resist harmful valgus stress, is at its peak (Camp et al., 2017). The movement of the pitch from this point on puts the elbow in critical position for a UCL tear (Chalmers et al., 2017). Found within this phase, the pitcher's front foot landing position, trunk to elbow angle (elbow abduction), and elbow external rotation above the shoulder plane is involved. In order of reference, Figures 5, 2, and 3 are represented in the cocking phase. Secondly, the acceleration phase is responsible for generating the greatest velocity for the baseball by the transfer of energy. In other words, the body transfers its energy into the baseball by using the potential energy stored within the pitcher's internal rotator muscles. In this phase, the pitcher is at maximal external rotation in their elbow, and the location of their elbow at their release point can be visually observed (Figure 4). Lastly, the pitcher shifts into the deceleration phase with the baseball's release as arm movement slows down. The arm is working hard to resist the momentum that was generated by the body weight of the pitcher. The pitcher's arm slot is able to be visually identified during this phase (Figure 1).

Before gathering the necessary videos, the researcher established their two variable groups of MLB pitchers. When developing these groups, a question of limitation arose regarding the mechanical differences between right-handed and left-handed pitchers. This detail could have impaired the results of the researcher's content analysis. In a study conducted in 2019, however, it was found that there was no statistically significant difference between the throwing mechanics

of right-handed and left-handed pitchers (Diffendaffer et al., 2019). The differences found were minimal and inconsistent, suggesting that mechanical practices can be taught regardless of dominant throwing hand. In addition, Diffendaffer's study split the groups in half with both right-handed and left-handed. In order to maintain this lack of significant difference, the researcher followed a similar procedure. The two groups were strategically chosen, having an even split of ten right-handed pitchers and then left-handed pitchers, creating a total of twenty pitchers per group. Furthermore, those who had experienced a significant tear requiring Tommy John surgery or had a clean injury history concerning the UCL serve as the independent variable in this analysis. The observations that were made by the researcher are classified as the dependent variable, as they vary between both groups. After collecting the needed information, the researcher organized the observational data into a chart (Appendix A) in order to analyze any existing patterns and produce quantitative information. These patterns will answer the question if there are certain biomechanics associated with generating more valgus stress to the elbow than others.

In order to see if there are any specific themes between the two variable groups of baseball pitchers, the researcher conducted not only a visual comparison but a metric data comparison. The website Baseball Savant, sponsored by the MLB, was utilized for its advanced feature known as Statcast. Statcast provides instant replay that measures much of the action that takes place on the field. These statistics include pitching, running, and hitting, allowing individuals to have access to the quantified metrics of professional baseball pitchers. The researcher used Baseball Savant to perform the second portion of their content analysis. By using Baseball Savant, the researcher gained access to a variety of statistical data regarding the analyzed baseball pitchers. Specifically, an important piece of data crucial to this study is the horizontal and vertical release position. The horizontal and vertical release positions refer to the position of the baseball from the catcher's perspective in feet when released by the pitcher. Both positions are labeled within Figure 7. Furthermore, Baseball Savant provides measurements relative to the extension of the pitcher. Extension refers to how far the pitcher releases the ball off of the mound in feet, as shown in Figure 8. These three factors were chosen because they can be identified amongst the deceleration phase of the pitcher, one of the three most important phases needed to be analyzed for this content analysis. For the experimental group of former Tommy John patients, the biomechanical data used for each pitcher was a calculated average of each of their numbers that were available from the season they debuted to the season they tore their UCL. For the control group of MLB pitchers who have a clean UCL history, their most recent metrics from the 2023 season were used. By reviewing both the UCL torn and clean history groups, an additional table was constructed by the researcher (Appendix B) to interpret any patterns among each variable group. Although the first portion of observations provides an effective analysis of the biomechanics correlated with future UCL tear, including a metric data inspection further solidifies the significance of the researcher's results. The visually interpreted conclusions are backed up with precise measurements, creating more noteworthy findings and developing a more complex method.

Figure 7



(Mike Sonne, 2015)

Figure 8



(Driveline Baseball, 2012)

Results & Analysis

The first portion of the researcher's content analysis aimed to visually observe the similarities and differences between MLB pitchers with different UCL histories. Both groups of pitchers, those who did tear their UCL and those who have not, were important in this effort. All five naturally occurring biomechanics were considered when watching these videos, and the observations made were organized into two tables (Appendix A) to see if a new understanding could be developed. The second portion of the researcher's content analysis gathered numerical data that was helpful in summarizing the first portion's findings. The metrics the researcher gathered from Baseball Savant were organized into two separate tables for testing (Appendix B).

The results from both portions of the content analysis were statistically analyzed. By performing these statistical analyses, the collected data is able to be deemed significant or insignificant. The first portion of the researcher's content analysis was tested using Two Sample Z-Tests of Proportions, otherwise known as a z-test, while the second portion of the researcher's content analysis was tested by using Chi Squared. In total, nine z-tests and three Chi Squared Tests of Independence were performed using a TI-84 Plus CE Graphing Calculator as a tool.

Two Sample Z-Test of Proportions

A Two Sample Z-Test of Proportions is a statistical hypothesis test that is used to determine if there are significant differences of the specific characteristics between two populations. In this study, the proportion of a specific observed biomechanic occurring in one population was compared to the proportion of that same observed biomechanic occurring in another population. The null hypothesis (H_0) for this study claims there is no statistically significant difference in observed biomechanics between the MLB pitchers who had Tommy John surgery and those who did not. On the other hand, the alternative hypothesis (H_a) states that there is a statistical significance difference between the two groups. With the Two Sample Z-Test of Proportions, the null hypothesis claims the two proportions are approximately the same. In order to reject this null hypothesis, a p-value less than 0.05 needs to be calculated.

Firstly, the initial five z-tests were conducted with the use of the TI-84 Plus CE Graphing Calculator's "Stat" feature. Under the "Tests" option, the "2-PropZTest (#6)" option was selected. With the researcher's sample size for both the experimental and control group being twenty, values "N1" and "N2" were equal to twenty. Next, the number of times a specific biomechanic was observed was inputted. "X1" represents the number of occurrences a specific biomechanic was observed within the group of former Tommy John patients, while "X2" represents the number of occurrences that same biomechanic was observed within the group of clean UCL history MLB pitchers. Since the researcher was only interested in determining a significant difference between the proportions of the observed biomechanics in both populations, the option " $P1 \neq P2$." In simpler terms, " $P1 \neq P2$ " means the two proportions are not equal. The first five z-tests were performed to get each biomechanic's p-value and distributed into Table 1. The Z-score, or how many standard deviations a given data value is from the mean, was calculated along with the p-value.

Observed Biomechanic	X1/N1 Former Tommy John Patients	X2/N2 Clean UCL History Pitchers	Z-Score	P-Value
Arm Slot	16/20	15/20	0.3793216208	0.7049544115
Elbow to Trunk Angle	16/20	10/20	1.988980632	0.0467032005
External Rotation Above the Shoulder Plane	16/20	17/20	-0.4161251893	0.6773184734
Relative Location of the Elbow Compared to Shoulder At Release Point	6/20	13/20	-2.216366554	0.026666317
Open/Closed Landing	19/20	16/20	1.434274331	0.1514940815

Table 1: Mixed Significant & Insignificant Two Sample Z-Tests of Proportions

An alpha, or significance, level of 0.05 was used in this study to measure the significance level of the calculated proportions. Out of the initial five z-tests that were performed, two yielded significant results. The specific biomechanic that was represented by the Elbow to Trunk Angle proportions in Table 1 was angles equal to 90 degrees. Sixteen out of twenty pitchers who tore their UCL had an elbow to trunk angle of 90 degrees, while ten out of twenty pitchers who have never torn their UCL had an angle of 90 degrees. In order to solidify this finding, two additional z-tests 90 degrees were performed and recorded into Table 2. The options “P1>P2” for an elbow to trunk angle being 90 degrees and “P1<P2” for an elbow to trunk angle being less than 90 degrees were now selected.

Observed Biomechanic	X1/N1 Former Tommy John Patients	X2/N2 Clean UCL History Pitchers	Z-Score	P-Value
Elbow to Trunk Angle (<90 degrees)	4/20	10/20	-1.988980632	0.0233516003
Elbow to Trunk Angle (90 degrees)	16/20	10/20	1.988980632	0.0233516003

Table 2: Significant Two Sample Z-Test of Proportions

With this information, it can be concluded that there is a statistically significant difference between the amount of pitchers in both populations who displayed an elbow to trunk angle of 90 degrees (Figure 9) and less than 90 degrees (Figure 10.). Additionally, it can be concluded that pitchers who have torn their UCL displayed more of a 90 degree elbow to trunk angle while those who have never torn their UCL display an elbow to trunk angle less than 90 degrees. As both p-values are less than 0.05, the null hypothesis is rejected.

Figure 9



(Major League Baseball, 2011)

Richard Lovelady (former Tommy John surgery Patient): Elbow to Trunk Angle 90 degrees

Figure 10



(Major League Baseball, 2023)

Gerrit Cole (never torn UCL): Elbow to Trunk Angle of <90 degrees

Lastly, the z-test associated with the relative location of the elbow at release point claims there is evident statistical difference. The specific biomechanic that was represented by the proportions of Table 1 was the location of the elbow at the release point in between the point of the shoulder and the bottom of the ear. Six out of twenty pitchers who have torn their UCL had a relative location of their elbow above their ear at release point, while thirteen out of twenty pitchers who have not torn their UCL had a relative location of their elbow in between their shoulder and the bottom of their ear. The p-value of this z-test yielded significant results as the P-Value was less than 0.05. In order to verify this difference, two additional tests were conducted and recorded into Table 3. The options “P1<P2” for an elbow located between the shoulder and the bottom of the ear and “P1>P2” for an elbow located above the ear were now selected.

Observed Biomechanic	X1/N1 Former Tommy John Patients	X2/N2 Clean UCL History Pitchers	Z-Score	P-Value
Relative Location of Elbow to Shoulder at Release Position (Between shoulder and bottom of ear)	6/20	13/20	-1.906925178	0.0282650711
Relative Location of Elbow to Shoulder at Release Position (Above the ear)	14/20	7/20	1.906925178	0.0282650711

Table 3: Significant Two Sample Z-Tests of Proportions

Based on the additional findings of these tests, it can be concluded that there is not only a statistical difference between the proportions of the elbow being located between the shoulder

and the shoulder in both populations. In fact, it can also be concluded that MLB pitchers who have torn their UCL displayed their elbow above their ear at release point (Figure 11) while those who have never torn their UCL displayed their elbow between their shoulder and the bottom of the ear (Figure 12).

Figure 11



(Major League Baseball, 2023)

Ian Anderson (former Tommy John surgery Patient): Relative Elbow Location is Above the Ear

Figure 12



(Major League Baseball, 2023)

Corbin Burnes (never torn UCL): Relative Elbow Location is Between Shoulder and the Ear

Chi-Squared Test of Independence

Chi Squared (Figure 13) is another type of statistical hypothesis test that uses a specific formula to determine if the collected data is enough to be statistically significant to reject the null hypothesis. Specifically, using Chi Squared allowed the researcher to analyze the pattern of observations and see if a certain frequency of one of the observations is associated with either the experimental group or the control group.

Figure 13

$$\chi^2 = \sum \frac{(O - E)^2}{E}$$

χ^2 = the test statistic \sum = the sum of
 O = Observed frequencies E = Expected frequencies

(Frimodig, 2023)

Before using the formula, the expected value, observed value, and degrees of freedom needed to be established. The expected value in this study was the numbers gathered from the group of MLB pitchers who never had Tommy John surgery, while the observed value was from the group of former Tommy John patients. In addition, the degrees of freedom for this study was determined to be one, as there are only two groups in which the observed outcomes of the biomechanics could have occurred in. After using the Chi Squared formula, the test statistic (χ^2), was calculated. Next, each X^2 was recorded into Table 4 for further comparison to the critical value.

Biomechanic Metrics	X^2	P-Value
Horizontal Release Point	0.3121151639	0.50-0.75
Vertical Release Point	0.0297366441	0.75-0.90
Extension	0.0050946179	0.90-0.95

Table 4: Statistically Insignificant Results of the Three Chi-Squared of Independence Tests

A critical value of 0.05 is most commonly used by scientists to determine significant differences between the experimental and control group. Therefore, 0.05 was used in this study to conclude the statistical significance of the researcher’s results. When the X^2 values were brought to the Chi-Squared Distribution table (Figure 14), the probability of exceeding the critical value (p-value) was analyzed. If the p-value is greater than 0.05, the null hypothesis is accepted with the conclusion that the variables are not statistically significant. In contrast, if the p-value is less than 0.05, the null hypothesis is rejected with the conclusion that there is statistical significance within the data. All three p-values the X^2 values fell between on the Chi-Squared Distribution table were recorded on Table 4.

Figure 14

Degree of Freedom	Probability of Exceeding the Critical Value									
	0.99	0.95	0.90	0.75	0.50	0.25	0.10	0.05	0.01	
1	0.000	0.004	0.016	0.102	0.455	1.32	2.71	3.84	6.63	

(Sapkota, 2023)

Out of the three Chi-Squared Tests of Independence that were performed, all three yielded p-values that fell greater than 0.05. As a result, there is not enough evidence to be able to prove a statistically significant difference between the horizontal release point, vertical release point, and extension metrics of the MLB that tore their UCL and those who have not. In other words, our null hypothesis repeatedly fails to be rejected.

Limitations

Throughout this study, many limitations occurred that should be kept mindful when interpreting these results. For example, one major factor that limits the findings of this research is sample size. Although a sample size of 40 MLB pitchers was effective enough to produce significant results, it may not accurately represent the entirety of the MLB pitcher population. In addition, the limitation of variability falls into the category of sample size. The population of this study had certain characteristics that varied between each pitcher, such as height and arm length. These physical attributes could have affected the gathered data's reliability as their differences might influence the pitchers' techniques. Lastly regarding sample size, the second portion of the researcher's content analysis had to do with the overall averages of each pitchers' recorded metrics from their included seasons. The number of included seasons for those that tore their UCL involved the season they debuted, to the season they tore their UCL. The number of included seasons for those that have not torn their UCL involved the season they debuted, to the 2023 season. For that reason, certain pitchers may have more accurate numbers than others as they have more seasons in their career.

Another limitation that should be considered is the interpretation bias of the researcher. Isolating the biomechanics needed to be studied was only achievable through the use of secondary data. Therefore, when analyzing the videos of these pitchers' mechanics, the observations recorded were up to the researcher's own interpretation. In other words, the biomechanical data could have been subjective. Furthermore, the types of pitches that were thrown by these pitchers in the selected videos varied, potentially influencing the pitching style that was observed. As a result, the findings of this study may be difficult to develop into more complex implementations and may require further research.

Future Directions

Other researchers should repeat this study with the use of a larger sample size in order to limit the variance in the data collected. By limiting the variance, the standard deviation that may have produced insignificant data can be decreased. Another opportunity to reduce the variance of this study is to analyze the influence that physical traits bring to the discussion in terms of biomechanics. This can help the applications of this study be generalized to a wider audience of pitchers with differing features, like heights.

Further evaluation of the different pitch types' impacts on biomechanics can also help limit the minor inaccuracies found within this study. By clarifying this aspect of pitching mechanics, a deeper understanding of pitching styles and their impact on the UCL can be

established. Expanding this knowledge can help preserve healthy UCLs not just at the professional level but the collegiate and amateur levels as different techniques and workloads may be applied at different levels.

Lastly, due to the fact that the researcher used secondary research to carry out their content analysis, considering the feasibility of using biomechanical monitoring equipment to replicate this study can help capture the motion of these pitchers in real time. Even though the results of this study encourage action to be taken by coaches and sports trainers to reduce the risk of injury amongst their players, the real time feedback from this technology can allow continuous assessment as well as immediate adjustments to be made. An option like this is more feasible by the collaborative efforts of multiple fields. Experts within fields such as sports medicine and physical therapy willing to work together can help amplify the process of interpreting the findings of this research.

Conclusion

Despite the fact that there is room for further research within this study, the researcher's work has contributed to advancing the overall understanding of biomechanics in pitching injuries. In terms of elbow to trunk angle and the relative location of the elbow at release point, this study has established a new foundation of the difference in biomechanics between MLB pitchers who have and have not torn their UCL. By identifying these biomechanical differences, new training programs to minimize the risk for injury while enhancing performance can be developed more efficiently. Pitchers adjusting to programs focused on reducing strain on the UCL can help them not only succeed but extend their career by avoiding injury. Furthermore, assessing the pitching mechanics of younger pitchers with this new basis can help recognize and prevent overuse injuries from occurring to encourage healthy development. As the importance of education on the biomechanical differences between these two groups of pitchers has been highlighted in this study, improved rehabilitation programs can help reduce the risk of reinjury to the UCL and improve recovery from surgery.

Although the researcher's null hypothesis repeatedly failed to be accepted within the Chi Squared Test of Independence, it was discovered within the 2-Sample Z-Tests of Proportions that pitchers who needed Tommy John surgery had greater (90 degree) elbow to trunk angles and their elbows at a greater distance from their shoulder at release point. These findings fall in line with the researcher's initial hypothesis as both of these mechanics exert a large sum of valgus stress to the UCL. Additionally, these findings build a new foundation of understanding the role of biomechanics in the field of study pertaining to preventing UCL injuries and the need for Tommy John surgery. A greater elbow to trunk angle can be associated with increased forward trunk tilt, while a greater distance from the elbow at release point can be associated with greater elbow flexion. Therefore, it can be concluded that increased forward trunk tilt and greater elbow flexion exerts a substantial amount of valgus stress on the UCL to put the pitcher at risk for Tommy John surgery.

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Microbial Influences on Human Immunity: Bacterial Symbionts and Their Role in Autoimmune Disease Development and Homeostasis By Chuan Ma

Abstract

Microbes on the human body are constantly interacting with their hosts, and these interactions have profound impacts on human health. Given this, this study aims to discuss the role that bacterial symbionts play in shaping the development and homeostasis of the holobiont immune system, and to understand the implications of the holobiont's role for autoimmune disease development in humans. It is known that the microbiomes have significant impacts on the physiological activities of their hosts, and the host's system interacts with the symbionts to reach the balanced condition for both factors. The studies also showed that the microbiomes affect the development of host systems and the evolution of species. This review thus aims to make this fundamental concept accessible to students who possess interest toward holobiont but are not capable of studying it academically. This research review finds that the holobiont theory, which considers a multicellular organism's components as both the host and microbiomes, elucidates the mechanism and development of the immune system, and the microbiomes are strongly related to immune disorders like autoimmune diseases. This review concludes previous research on the holobiont and immune system, enabling more people to interpret the status quo for the field of research; the revealed limitations in current studies may provide directions for future research, making the holobiont theory more comprehensive.

Introduction

When talking about a human, you may regard the person as “a single individual” or “an independent organism”. However, the system of our bodies also involves numerous bacteria, a frequently neglected component that carry out significant functions in human health and metabolism. Human microbiomes include over 100 trillion microorganisms, which totals 70% to 90% of cells in the human body (Perkins), and bacteria contribute approximately 99% of unique genes present in human bodies when considering genomes (Perkins). Bacteria interact closely with human physiological processes, and establish various relationships with the human host. Bacteria can have beneficial or pathogenic relationships with humans due to differences in cell signaling mechanisms and approaches of colonization. The beneficial relationships are exemplified by bacteria's regulation of digestion and antimicrobial strategies for preventing pathogenic invasion (Curtis). Conversely, pathogenic relationships are induced by bacteria's virulence strategies of colonization and secretion of toxins (Curtis). Regardless of the kind of relationship that bacteria have with their human hosts, there's significant evidence that bacteria play a crucial role in human health (Hou et al.). Research supports that bacteria can modulate functions of organs like the gut, liver, and kidneys, and can also affect the activities of physiological systems to maintain homeostasis, such as controlling the immune system responses (Hou et al.).

Microbiomes affect humans in different stages of life, exerting impacts on people's infantile development and further maturation. To be specific, the microbiome plays a critical role during the early life critical window, specifically maternal-fetal transfer, and vaginal birth which are critical points where the microbiome begins colonization and development in the developing child (Stiemsma and Michels). This process impacts organ development for infants, and the microbiota maturation bolstered by breastfeeding all establish the prolonged microbiome composition of the person (Stiemsma and Michels). Furthermore, the development of microbiomes at an early age also induces sustained effects on people's health, which further relates with many diseases in humans. For instance, early-life dysbiosis (disturbance to microbiome conditions) in both the human gut and respiratory system microbiomes is related with asthma and atopic disease development in children (Stiemsma and Michels). Due to how our microbes affect our bodies' homeostasis and interact with our immune system and gut (Afzaal et al.), diseases like obesity, hypertension, cardiovascular diseases, diabetes, cancer, and depression are all possibly triggered when human microbiomes are dysfunctional (Ding et al.). In other words, the human microbiota aids in proper functioning of physiological systems, and supports development, helping to ensure good health.

As researchers discovered the significant relationships between humans and their symbiotic bacteria, the concept of the holobiont emerged to describe the host-bacteria relationship: the host, the microbiota, and the mutually dependent relationship between two agents together can be regarded as a holobiont (Van de Guchte et al.). Therefore, all the effects of symbionts on the host mentioned before are holobiont interactions. Among different types of holobiont interactions, the impacts of microbiomes on the immune system is particularly important. The immune system primarily carries out antimicrobial functions that prevent the invasion of pathogenic agents. As the colonization of microbes occurs beginning at the early stage of human development, it leads to bacteria-immune system interactions. As the bacteria make contact with immune cells, interactions occur between the host and the microbiome, making the system regulated by both human cell and microbiome components (Schneider). The functions of the immune system are thus dependent on microbiota. Among all immune syndromes induced by dysfunctional microbiota, autoimmune diseases, which refers to the circumstances in which the human immune system attacks non-pathogenic host cells, will be emphasized in the following passages.

Holobiont theory

The holobiont generally involves three components: the host, the symbionts, and the interactions between them. Nearly all multicellular organisms have microbiota, and therefore, are holobionts (Postler and Ghosh). Researchers primarily focus on mammals, like germ-free mice that possess no microbiomes in their bodies, while this article will primarily focus on humans. As for the symbionts, it's a generalization for all microbiomes in the host body, which involves bacteria, fungi, and other similar microbes. As for the interactions, it includes all physiological processes that reach balance by the effect of both the host cell and the microbiomes. The concept

of the holobiont developed as researchers gradually discovered the effect of symbionts on human development. Initially, early endosymbiotic theories form in the 1920s, and Meyer-Abich's theory of holobiosis emerges along with the discovery of DNA in organelles in the 1960s (Lovett). The systematic concept of holobiont was then proposed by Dr. Lynn Margulis in the 1990s. After the 1990s, with new evidence discovered for the theory, the holobiont theory is currently under debate for its interpretation, forming individual selection theory and neutral theory of microbial diversity. Individual selection theory claims that the development of microbiomes follows natural selection, and the process directly affects the individual organism (individual selection) (Shelton and Michod). Also, the neutral theory of microbial diversity states that “it is the host environment that ultimately shapes the community assembly and tips the human microbiome to a niche regime.”(Li and Ma), which elaborates on the accumulative effect of host conditions. Generally, instead of merely considering the approaches of evolution for holobiont as the two theories mentioned, current holobiont theories also involve analysis on the interactions between components of holobionts. The studies about the relationships are more frequently done as the relations between microbiome and host are more traceable by methods of biochemistry and medicine, and there are effective control groups or experimental agents like germ-free mice.

Microbial communities exist all over the human body, colonizing nearly all organs and tissues; the most studied are the gastrointestinal system, skin, mouth, and respiratory tracts (Shao et al.). Interactions in these systems can be categorized into bacteria-bacteria interactions and bacteria-host interactions. For the inter-bacterial interactions, one form of common communication methods is called quorum sensing and is a fundamental process for microbiome survival (Ursell et al.). Quorum sensing occurs by accumulation of signaling molecules till the concentration passes a threshold, then activating the transduction (Bassler and Losick). In host-bacteria interactions, the signaling processes are often carried out by bacterial hormone-like compounds named autoinducers (Sperandio et al.) and host hormones, which may affect gene expression, immunomodulatory activities, and development of tissues (Sperandio et al.). The gastrointestinal system exemplifies the features introduced for these host-microbiome interactions. From studies of germ-free mice, there are links between a host's microbiota, digestion, and metabolism. For digestion, not only the microbiomes' genomes allow creatures to expand the diet from the host cells' genome, but also the digestion process and activation of both host cell and bacteria will be determined by the conditions of both factors. What's more, for the metabolism, gut microbes will control the energy extraction according to the food uptake, which affects the metabolism process (Ursell et al.). Both relationships mentioned have shown possible approaches for microbiota to relate with host cells. With consistently close correlation, the evolutionary developments of host cells and microbiomes also occur in parallel. This coevolution process has sustained for millions of years, where altering external and internal environments consistently induce corresponding phenotypic changes for the holobiont (Shahab and Shahab), representing the evolutionary effect of host-bacterial relationships.

Bacterial communities and the Immune system

To fully discuss the interaction between the holobiont and the immune system, it's crucial to present the fundamental mechanisms and concepts of the immune system in advance. In general, the immune system blocks or eliminates non-self or pathogenic components for the human body. To provide a more comprehensive definition, the immune system expands throughout the human body, consisting of lymphoid organs, cells, and cytokines (Saez et al.). For clarification, cytokines and chemokines are molecules responsible for cell signaling, especially for the immune system (Cameron and Kelvin). What's more, the immune system's functions are recognising, repelling, and eradicating pathogens or external molecules (Parkin and Cohen). When considering different immune mechanisms, the immune system can be categorized into two types: the innate immune system and adaptive immune system. To begin with, the innate immune system is characterized by rapid responses toward microorganisms without specifically targeting certain species of microbes (Saez et al.). The fundamental cells in the innate immune system are myeloid cells that are produced in bone marrow, including neutrophils and macrophages. To carry out their functions, these cells will first release pattern recognition receptors, which enable them to distinguish between "pathogen-associated molecular patterns" and "damage-associated molecular patterns", where the former indicates the recognition of pathogens' membrane components, and the latter refers to the recognition of molecules released by damaged or infected host tissues. Then, these cells will trigger immune responses like phagocytosis by releasing the cytokines and chemokines, or they will activate the adaptive immune system by forming a targeted and specific cellular response (Saez et al.). Additionally, the adaptive immune system recognizes specific non-self cells that enter the human body from external environments, distinguishes them from the holobiont's own cells, and activates pathogen-specific pathways to eliminate the pathogens or infected cells (Marshall et al.). Specifically, the adaptive immune system will merely determine whether a cell in the human body originates from the holobiont or from external environments, and the system will eliminate or block all non-self components without determining their specific species or traits. They also trigger more immediate immune responses if the same pathogen is exposed to the holobiont repeatedly. Typical cells involved in the adaptive immune system are T cells and B cells. T cells present antigens' membrane components for other immune cells to digest the pathogens, or they act as cells that annihilate pathogens. Additionally, B cells are related to antibody production, where the antibodies bind with pathogens to make them more easily digested by phagocytes.

In order to discover how the commensal microbes interact and help to develop the immune system, additional research is needed to understand the scope of bacteria-bacteria and bacteria-host interactions. Of what is understood regarding bacteria-bacteria interactions, there are two well established forms: quorum sensing and intimate bacterial conversations. The intimate bacterial conversation refers to short-range signaling via direct contact between individual cells for information exchange (Bassler and Losick). For quorum sensing, the bacteria will recognize extracellular concentration of the signaling molecule and activate their response when the concentration reaches a certain threshold. As long as the concentration of the chemical

surpasses a certain threshold, the entire population of bacteria will alter gene expression, which means that the effects of quorum sensing are often oblivious for an individual bacteria but obvious for a bacteria population. For example, quorum sensing can control bioluminescence for certain creatures by accumulating the fluorescence effect of individual microbes in a population to have a significant effect on the host. For intimate bacterial conversations, C signaling is representative: this process stimulates spore formation and coordinated gliding behavior, suppressing reversal gliding behavior. One example of bacteria utilizing this approach is *Myxococcus xanthus*, and the process is critical for multicellular fruiting-body formation (Bassler and Losick). Though the mechanism of suppression is still unknown, this process does elucidate an approach of conversation via direct contact between bacteria. Additionally, the bacteria-host interactions are characterized by conversations of chemicals between host cells and microbiome, where the host's signaling pathway is affected by the microbes. Within metabolites directly produced by bacteria or produced by the host or the environment and metabolized by bacteria, many host receptors may possess bacterial or fungal origin ligands, proving that their functions are affected by the microbiome (Hertli and Zimmermann). In conclusion, for the bacteria-bacteria communication in a holobiont, quorum sensing and intimate bacterial conversations are common routes; for the bacteria-host interactions in a holobiont, microbes often have impacts on the host's signaling transduction.

Recalling the mechanisms of the holobionts and the immune system, as well as the larger number of microorganisms in the human body, we can now deduce the significant impacts of microbiomes on the human immune system (Perkins). To start with, if we select a microscopic view, analyzing the microbiome's pathways of communication with host cells by metabolites, three types of different metabolites are all demonstrated to have an impact (Postler and Ghosh). Firstly, the metabolites produced by bacteria from external components such as dietary intakes can affect the immune system. Short-chain fatty acids like acetate and propionate may have impacts on the innate immune system by inhibiting proinflammatory cytokines' production (Postler and Ghosh). Secondly, metabolites that are produced by the host and biochemically modified by bacteria have influences on the immune system. Secondary bile acids derived from host produced primary bile acids and microbiota modifications will hinder the transcription of proinflammatory genes in monocytes and macrophages in the innate immune system. Since these two cells are responsible for pathogenic antigen representation and pathogen annihilation, immune responses will be inhibited as the process occurs (Postler and Ghosh). Lastly, metabolites that are merely synthesized by microbiota also have an impact on the immune system. For instance, the Polysaccharide A originates from *Bacteroides fragilis*, and the metabolite is capable of inducing IL-10 secretion in CD4⁺ T cells (Postler and Ghosh). As CD4⁺ T cells are corresponding to antigen presenting cell (APC) formation, and these cells are engulfed by macrophages in the adaptive immune system, the PSAs are proved to have effects on the holobiont immunity: as the activity of CD4⁺ T cells are enhanced by Polysaccharide A, the T cells will activate more macrophages to engulf the APCs with pathogens. All these examples

directly provide evidence for the effects of microbiomes on the immune system by demonstrating that the metabolites from microbes have functions in affecting immune responses.

The development of the immune system due to interactions between microbiota and host at different locations of the human body will then be emphasized. It's discovered that microbiomes colonized in different sections of the human body all have impacts on immune system development and maintenance (Hou et al.). Firstly, the gut microbiome has a significant influence on immune system development (Hou et al.). For instance, the gut bacteria interact with mucosal antibodies (the antibodies presenting on human mucus) that are taken up by CD11 + dendritic cells (The cells presenting antigens on its surface) in the Peyer's patches, which are lymphoid tissues in the immune system (Hou et al.). What's more, the bacteria in intestines maintain the CD8 + T cells in the adaptive immune system, which are primarily found in the intraepithelial intestinal compartment (Hou et al.). Since these T cells are responsible for antibacterial responses of the immune system, the bacteria have an impact on the development of the immune system's functions. Evidence from an experiment using germ-free mice shows that these mice without microbiomes are presenting less CD8 + T cells, indicated that microbiome's existence guarantees the immune system's proper functioning, and the absence of microbiome induces inadequacy in fundamental immune cells (Helgeland et al.). Secondly, in the oral cavity, the bacteria and the immune system reach a balance, affecting immune system development: oral pathogens exert the ability to trigger an immune response, and the alterations of the immune system also affect bacteria colonies (Hou et al.). For instance, in periodontal disease gingivitis, the inflammatory responses of the neutrophils destroy oral tissues, which furnishes the microbiome with food sources; then, the detection of excessive population of microbes will also trigger the bactericidal and anti-inflammatory activities of the immune system (Hou et al.). Thirdly, the skin microbiomes also affect the development of the immune system. As many immune system components like macrophages and lymphocytes are present in skin tissues (Nguyen and Soulika). The contact between the microbiome and the immune system maintains the balance for activities of both factors. For instance, *Staphylococcus epidermidis* release lipoteichoic acids which prevent skin from inflammation by inhibiting cytokine production (which is crucial for many processes in immune system, as they are dependent on cell signaling) and TLR2-based immune responses (Hou et al.). Meanwhile, the antibacterial immune responses control the population of the skin microbiome, reaching the balance between two factors. By and large, the activities of the microbiome and that of the immune system are closely related with each other in different parts of the human body, developing the general balance between each other's development to maintain the holobiont's homeostasis: as for the example of *Staphylococcus epidermidis*, the population of microbes and the activity of immune response terminally maintain at a stable level.

Holobiont and autoimmune diseases

Autoimmune syndromes are characterized by immune disturbances that induce actions of T cells; auto-antibodies, and inflammatory reactions toward the normal constituents of the host,

whose occurrences are unrelated with people's age or organs (Marshall et al.). For clarification, auto-antibodies are immune system-originated proteins, binding with host cells to trigger immune responses toward the organism's own tissues. Additionally, for establishing the cause of the syndromes with microbiomes, we can deduce that cases of autoimmune diseases might be created by the microbiota, where the bacteria or other microbes negatively affect the adaptive or innate immune system and disturb their functions of pathogen recognition (Marshall et al.). As for the normal conditions, the microbiomes' activities establish balance with immune system actions (Marshall et al.). Cytokine signaling can be controlled by the microbiomes in organs where the immune cells are intensely distributed, and the population of microbiomes may be adjusted by the antibacterial mechanisms of the immune responses (Postler and Ghosh). To be specific, the excessive population of microbiomes will be reduced by immune responses targeted at microbes, while imbalances in the process make the immune system dysfunctional and result in annihilation of host components. This may occur when the microbial composition alterations induce corresponding changes in immune responses, such as the increased secretion of antibodies (De Luca and Shoenfeld). In addition, the unbalanced circumstance may also emerge as the communication pathways among the microbiota, immune system, and the target organs are disturbed, inducing false immune responses and further decrease of microbiome population (De Luca and Shoenfeld). In the following paragraphs, the article will analyze two specific details for the autoimmune diseases for better interpretation of the microbiome-related mechanisms.

Rheumatoid Arthritis, abbreviated as RA, is a chronic syndrome featured by inflammation and pain of the joints with varying degrees of systemic involvement; the most significant factor included in the mechanism of the disease is anti-citrullinate peptide antibodies (ACPA), an autoantibody that recognizes citrulline and triggers immune responses by activating leukocytes (De Luca and Shoenfeld). When considering the immune responses included in the process, two primary steps take place: initially, there is an accumulation of leukocytes, a component from the adaptive immune system. Then, inflammatory cytokines and mediators such as interleukins are released (De Luca and Shoenfeld). During this process, the bacteria *Porphyromonas gingivalis* transforms a signaling amino acid: through a process named citrullination. In this process, the amino acid arginine is transformed to the amino acid citrulline, and citrullines are specifically recognized by autoantibodies ACPA (De Luca and Shoenfeld). Therefore, as the ACPA in the adaptive immune system recognizes an excessive amount of citrulline, they trigger the immune response to accumulate leukocytes and induce inflammatory responses, terminally causing the RA syndrome.

The second instance of microbially-involved autoimmune diseases is anti-phospholipid syndrome (APS). The symptoms of the disease are primarily recurrent venous, arterial thrombosis, and fetal loss (De Luca and Shoenfeld). Meanwhile, the patients are observed to present persistently exorbitant levels of anti-cardiolipin and anti-phosphatidylserine antibodies, which are antibodies that recognize anionic membrane phospholipids, a component on cell surface membrane that is significant for cell signaling by representing themselves as "biochemical and biophysical landmarks" (Dubois and Jaillais). By binding with these lipids, the

antibodies will severely inhibit the signaling capability for the cells. Segmented filamentous bacteria (SFB) are closely related with the disease: as commensal bacteria, the conditions of their populations have impacts on the host's physiological system (De Luca and Shoenfeld). As their state of homeostasis alters, these bacteria affect T cell phenotypes and corresponding antibody productions, inducing the immune system's proinflammatory interactions (De Luca and Shoenfeld). As a result, according to conclusions of previous studies, the commensal bacteria SFB stimulates aberrant immune responses by affecting T cells' antibody production, inducing persistently occurring antiphospholipid antibodies that stymies cell signaling (Ruff et al.). By and large, autoimmune diseases are characterized by immune response toward host tissues, and they are closely related with the microbiome's conditions, as the examples of RA and APS have shown.

Discussion and Future Directions

Considering the symbiont and host cells in the human body, the holobiont theory does not merely consider the correlations between human organs' mechanisms. Instead, it defines the approach of reaching homeostasis and development of systems as the interactions between the host and the microbes. The microbiomes thus have a significant effect on human physiology, as homeostasis refers to stable conditions of organisms via self regulations. As microbiomes release critical substances for human physiological pathways, they can affect the signal transduction process in humans, having impacts on development. The instance of RA exemplifies the statement, as the *Porphyromonas gingivalis* affects immune response by altering a signaling amino acid. Additionally, as many human activities, such as the immune system's antimicrobial responses, are dependent on microbiome population, the conditions of microbes are closely intertwined with the extent of immune system activities, such as anti-bacterial responses. Within these effects, the microbiomes in holobiont theory thus significantly affect people's health conditions. They are closely related with human infantile and postnatal development, and their stability directly determines the health risk that people encounter. Moreover, microbiome alterations have been suggested to be causes of many diseases like Rheumatoid arthritis and Systemic sclerosis (De Luca and Shoenfeld). By and large, the impact of the holobiont on human health and diseases are undeniable.

The holobiont can impact humans in different life stages, such as infantile stages and periods of maturation, by affecting the development of tissues or interrupting regular functions of organs and systems. These changes may further affect the physiological state of humans in a prolonged lifespan, as the impacts on development of organs may cause long-term health issues. Additionally, the holobiont has been shown to impact the immune system by both interfering in the immune system's signal transduction pathways and affecting the extent of anti-bacterial immune responses due to the microbes' population sizes. These interactions generally maintain the stable, balanced conditions between the activities of the both factors; therefore, the problems in microbiome activities will induce immune disorders, and the autoimmune diseases are typical instances.

Though the essay represents many achievements in the field of holobiont research, the limitations of experiments or studies are also significant. For instance, the complexity of holobiont interactions induces failure for construction of valid models that simulate all the processes and predict the alterations of the system's conditions (Postler and Ghosh). In addition, the bacteria-bacteria interactions' effect on the holobionts are frequently neglected or failed to determine, as a single metabolite may induce effects on multiple species of microbes and various host tissues (Sperandio et al.). Meanwhile, future research will also endeavor to resolve these elusive issues. The possible directions of future holobiont studies may include theoretical analysis for microbiomes' diversity and impacts on holobiont evolution, as well as implementing applications of the concept on human health and environmental studies, as the pattern of evolution for microorganisms may reveal the alterations of environmental conditions in certain period (Simon and Marchesi). By and large, I believe that the restrictions and challenges in the field of holobiont study will be conquered soon, and this concept will create more comprehensive and impressive explanations to the physiology of humans and other organisms.

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A Novel Modified SERS Biosensor Utilizing Gold Nanoparticle Arrays for Sensitive Detection of Pancreatic Ductal Adenocarcinoma By Ananya Mishu Manoj

Abstract

Early and sensitive detection of pancreatic ductal adenocarcinoma (PDAC) is critical due to its malignancy with high mortality rates due to its aggressive nature and late diagnosis. The scope of this work extends to developing a generalized approach for biomarker detection using a modified Surface-Enhanced Raman Spectroscopy (SERS) assay that is cost and time effective, focusing on the KRAS biomarker, a common mutation in PDAC. The SERS assay consists of two main parts: the extrinsic raman label (ERL) and the antigen (PrEST Antigen KRAS). The ERL is built with the following: gold nanoparticles (AuNPs), raman reporter molecules (Cy5 dye), and the detection antibody (KRAS). The methods consisted of the preparation of the capture substrate, binding of the primary antibody, and Raman signal enhancement through laser excitation. The results demonstrate a direct correlation between the SERS signal intensity and the concentration of the PDAC-related biomarkers, with data analysis facilitated by a fluorescent spectrophotometer. This reveals the high sensitivity and specificity of the modified SERS biosensor for PDAC biomarker detection. This project confirms the feasibility of using a modified SERS assay as a method for biomarker detection and highlights the potential for this technology to revolutionize early cancer detection for PDAC.

Introduction

Pancreatic ductal adenocarcinoma (PDAC) is a highly aggressive lethal malignancy due to the lack of early diagnosis and limited response to treatments. It is the most prevalent type of pancreatic neoplasm, and it is developed in the exocrine compartment and accounts for more than 90% of pancreatic cancer cases. Given that KRAS mutations are present in nearly 100% of PDAC cases, it is considered the most RAS-addicted of all cancers, with exome-wide sequencing revealing mutational activation in 94% of PDAC cases (Waters). Surface-Enhanced Raman Spectroscopy (SERS) has emerged as a powerful analytical technique, offering enhanced sensitivity for the detection of molecular species (Cutshaw). The proposed SERS biosensor leverages gold nanoparticle arrays to enhance the performance of biomarker detection in PDAC. Gold nanoparticles, with their unique plasmonic properties, serve as an ideal substrate for SERS, amplifying the Raman signals of reporter molecules attached to biomarkers (Aldosari). In the past, SERS assays have been used for detecting pancreatic cancer, however, it has targeted various other biomarkers, such as CA19-9, MMP7 and MUC4, that have smaller mutation rates in PDAC compared to KRAS (Hu).

Due to its poor survival rates and lack of detection at an early stage, pancreatic cancer is one of the leading causes of cancer-related deaths in the United States (Kolbeinsson, Cai). Specifically, Pancreatic Ductal Adenocarcinoma (PDAC) is one of the most aggressive forms of pancreatic cancer and is estimated to be one of the leading causes of death by 2030 (Bekkali). Although Carbohydrate antigen 19-9 (CA19-9) is currently the only FDA approved biomarker

test for pancreatic cancer, other biomarkers such as Kirsten Rat Sarcoma (KRAS) have been shown to be more mutationally activated in PDAC (Hu). Current diagnostic methods, such as endoscopic ultrasound (EUS) and magnetic resonance imaging (MRI), only have survival rates up to 78% and are often detected at a late stage (Bekkali). PDAC remains the third leading cause of cancer-related deaths globally, with a 5-year survival rate below 10%. Early diagnosis is crucial yet remains challenging due to the absence of reliable biomarkers. While prior SERS-based assays focused on markers like CA19-9 and MMP7, the novel approach proposed in this study directly targets KRAS mutations, offering a more targeted and potentially earlier diagnostic tool.

2. Materials and Methods

2.1 Initial Cell Culture and Preparation

The cells used are LTPA CRL-2389 and SIM-A9 CRL-3265 which were acquired from the American Type Culture Collection. The LTPA cell line is an epithelial cell line from pancreatic tissue of a mouse with adenocarcinoma, making this the ideal cell line to use in the biosensor assay to model the disease phenotype in humans. The SIM-A9 CRL-3265 cell line is a microglial cell line isolated from the cerebral cortex of a mouse. This is used as one of the controls in the experiment to ensure that the results are specific to mutations concerning PDAC, without any background detection or non-specificity as a result of detection in other cell lines. Cells were thawed and cultured in Eagle's Minimum Essential Medium (EMEM) with fetal bovine serum (FBS) and Penicillin-Streptomycin solution to a final concentration of 10% in a 75 cm^2 tissue culture flask. The cells are incubated at 37°C with a CO_2 level of 5%. After cells are approximately 80% confluence in flasks, they are transferred to two 24 well plates and one 48 well plate through the following protocol: aspirate medium, wash with PBS, aspirate buffer, trypsinize cells, neutralize the trypsin with EMEM, and add 200 μ L of LTPS cells and 200 μ L of media to each well of the respective well plate. Cells are then put back into incubation until 70-80% confluent for further testing.

2.2 Antibody Addition and Creation of the Extrinsic Raman Label

2.2.1 Conjugation of the Primary Antibody

The primary antibody used in the Anti-KRAS antibody which is the mouse monoclonal clone KRAS-A582, purified from hybridoma cell culture and acquired from Sigma-Aldrich. The primary antibody is diluted in Phosphate-Buffered Saline (PBS) with a dilution ratio of 1:1000 (respectively). 52 μ L of the antibody is added to the appropriate wells. Reagent concentrations and incubation periods were optimized through iterative testing, adjusting conditions for maximum signal-to-noise ratio while minimizing non-specific binding.

2.2.2 Creating the Extrinsic Raman Label (ERL)

The ERL is built with the following: gold nanoparticles (AuNPs), raman reporter molecules (Cy5 dye), and the detection antibody (KRAS). The following protocol was used to develop the ERL: add 42 μ L of the fluorescently labeled AuNPs and 52 μ L of the alkanethiol [11-(Methylcarbonylthio)undecyl]tetra(ethylene glycol) to each well of two 24 well plates, dilute the PrEST Antigen KRAS in PBS with a dilution ratio of 1:50 (antigen:buffer) and add 80 μ L in each appropriate well. This also ensures the creation of the Extrinsic Raman Labels (ERLs) in the appropriate wells. After appropriate incubation, each plate is analyzed with the fluorescent spectrophotometer with an excitation level of 650 nm and an emission level of 670 nm.

2.3 Figures and Schemes

Plate Number	Number of Total Wells	Number of Rows Used	Experimental Setup
1 (48 well tissue culture plate, round bottom)	48 well plate	2	LTPA cells + AuNPs
		6	LTPA cells
2 (24 well tissue culture plate, round bottom)	24 well plate	3	AuNPs + Primary KRAS Antibody
3 (24 well tissue culture plate, round bottom)	24 well plate	2	LTPA cells with ERL + Antigen ¹
		2	SIM cells with ERL + Antigen
		2	ERL (AuNPs with Antibody)

¹This condition represents the main experimental group tested for accurate and sensitive detection of PDAC.

Fig 1: This is a table representing the setup of each plate. The bolded text indicated the experimental condition most valuable for this experiment. Each experimental condition is tested in duplicate to ensure consistent and accurate results.

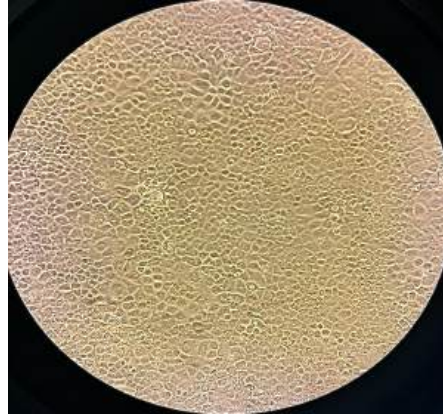


Fig 2: This is a table representing the confluency of the cells used in each well.

3. Results and Discussion

As shown in Figure 3, the readings from the fluorescent spectrophotometer reveal the specificity of the modified SERS biosensor for PDAC detection. As shown with the higher readings for the LTPA Cells with the Antigen and ERL, the assay was successfully able to differentiate between the PDAC cells and PDAC related biomarkers and the various control groups. Specifically, this table highlights the difference between the PDAC cells and the microglial cells. The average value from the PDAC cells is 896.91 and the average value from the SIM cells is 930.40. This showcases the assay's ability to specifically indicate the difference between cases of PDAC and other types of cells. This large difference in the readings between the various cell types shows the specificity of this assay for detecting and quantifying Pancreatic Ductal Adenocarcinoma.

LTPA Cells with Antigen + ERL		SIM Cells with Antigen + ERL		Plain ERL (AuNP + Antibody)	
1408.5	272.44	889.39	681.92	150.24	26.517
869.93	1168.8	784.83	962.11	363.27	43.245
1250.9	1113.6	1193.2	1345.4	623.03	156.53
191.22	899.85	417.71	1168.6	267.68	63.984

Fig 3: This is a table representing the fluorescent spectrophotometer readings from Plate 3 comparing the experimental group with the appropriate control groups.

3.1. Optimization of Sensitivity for PDAC Detection

To optimize the sensitivity of PDAC detection and minimize background noise/signaling, each plate is analyzed with the fluorescent spectrophotometer with an excitation level of 650 nm

and an emission level of 670 nm and compared to the normalized control group values. The fluorophore used for detection in this assay is AFDye 647 (Abs/Em similar to Cy5). The bright, photostable, and pH insensitive nature of this fluorophore contribute to sensitive detection while using this dye. Its common use for flow cytometry also supports its practical and accurate detection for fluorescent spectrophotometry as well as they can both characterize samples based on their fluorescent properties.

4. Discussion

4.1. Potential On-Target Off-Tumor Effects on Assay Function

Although the assay displayed high specificity for KRAS in PDAC cells, there is a possibility of slight cross-reactivity with non-target proteins. These off-target effects could be due to non-specific binding of the antibody or unintended interactions with the nanoparticle surface. KRAS is also known to be mutationally activated in a few other cell types as a part of breast cancer, colon cancer, or lung cancer (Jancik). Although KRAS is most prevalent and mutationally activated in pancreatic cancer, it is crucial to recognize the possibility of this modified SERS assay detecting other cancers or cell lines as well. Although this was partly controlled in the assay described in this paper by using a microglial cell line as a control, it is essential to test with more cell lines known to have KRAS mutations, other than PDAC, in the future. Future optimizations, such as engineering more selective antibodies or modifying the surface coating of nanoparticles, could further improve assay precision.

4.2. Existing Diagnostic Techniques, Mechanistic Insights, and Theoretical Implications

While CA19-9 assays remain the standard, they are limited by lower specificity, especially in non-cancerous pancreatic conditions (Geka). The modified SERS method tested in this paper directly targets KRAS mutations, offering a more precise diagnostic tool. However, further validation against large patient cohorts is needed to fully establish its clinical superiority.

4.3. Error Analysis and Limitations

In conducting a modified Surface-Enhanced Raman Scattering (SERS) assay for the sensitive detection of Pancreatic Ductal Adenocarcinoma (PDAC), the project aimed to leverage the specificity and sensitivity advantages of SERS. However, a pivotal limitation was encountered due to the unavailability of Raman spectroscopy equipment. Instead, a fluorescent spectrophotometer is used to collect data. This substitution may have introduced variations in the results, as the detection mechanisms and sensitivities between Raman spectroscopy and fluorescence spectrophotometry differ significantly. Specifically, while SERS relies on enhanced Raman scattering for molecular fingerprinting, fluorescence spectrophotometry measures the intensity of fluorescent light emitted by labeled compounds. This discrepancy potentially affected the accuracy levels of detecting PDAC, as the fluorescent spectrophotometer might not capture the subtle molecular interactions and changes with the same precision as Raman

spectroscopy. Consequently, the project's findings, while valuable, should be interpreted with consideration of this instrumentation constraint, acknowledging the possibility of variations in sensitivity and specificity outcomes.

4.4. Main Conclusions and Future Optimizations

This project investigated the creation of a modified SERS biosensor to detect pancreatic ductal adenocarcinoma by utilizing the KRAS biomarker mutation. Through iterative optimization and validation with different cell lines, the assay demonstrated a clear differentiation between PDAC-specific signals and control conditions. While there were challenges in maintaining consistent signal intensity due to nanoparticle aggregation, this work successfully establishes the foundation for an affordable and highly sensitive diagnostic tool, which could be particularly impactful in clinical settings with limited resources.

Future studies could explore surface modifications using PEGylation or different linker chemistries to enhance detection sensitivity. Additionally, alternative fluorophores like Alexa Fluor 750 could be evaluated to reduce background interference and further improve assay sensitivity (Li). Future work must also involve validating the assay with clinical samples from PDAC patients to assess its utility in a patient-care setting. Additionally, integrating this SERS assay with microfluidic platforms could enable point-of-care testing, paving the way for broader clinical adoption. To address the inherent limitations of fluorescence spectrophotometry in detecting low levels of PDAC, stringent background correction methods were employed and compared with results simulated Raman spectra (Fornasaro and Lin). While this partially mitigates the discrepancy, the need for access to Raman equipment remains a critical aspect for future studies.

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Utilizing Machine Learning Methods to Predict Patient Outcomes and Identify Novel Biomarkers in Breast Cancer Subtypes By Arushi Singh

Abstract

In 2022, 2.3 million people around the globe were diagnosed with breast cancer and currently, in 2024 there are 310,720 estimated new cases, making it the most common form of cancer with 80% of cases being invasive ductal carcinoma, a type of breast cancer that affects the breast's milk ducts [1],[2]. Within this carcinoma, several subtypes exist and are differentiated by biological markers. Biological markers in the context of breast cancer refer to the specific molecules, proteins, or characteristics used to classify breast cancer patients into subtypes and help understand the biological behavior of the breast cancer cells for every patient as the behavior of each patient's breast cancer cells is unique [3], [4]. Some examples of biological markers are progesterone receptor, estrogen receptor, human epidermal growth factor receptor 2 (HER2) status, N stage, grade, and more. Based on these biological markers, these cancers are classified into subtypes including estrogen-receptor-positive breast cancer, progesterone-receptor-positive breast cancer, HER2-positive breast cancer, triple-negative breast cancer, Luminal A breast cancer, HER2-enriched breast cancer, and more. Identifying which biological markers are significantly impacting a patient is important as it can determine the type of breast cancer a patient has, the chance of survival, treatment options, and more [5]. However, despite advancements in treatments, a considerable portion of patients remain unresponsive, necessitating additional research for targetable biomarkers within these subtypes, such as gene dysregulations and mutations, a goal of this project [6]. Thus, the goals of this research are 1) to investigate the role that subtypes play in determining patient outcomes and 2) to use machine learning models to identify new biomarkers in breast cancer subtypes with RNA-seq data. For the first goal, different biomarkers are compared for predicting patient outcomes to other subtypes using machine learning models specifically, a Logistic Regression model, a Cox-Hazard Model, and a Kaplan-Meier Curve. The Logistic Regression model achieved an 85% accuracy score and a 0.75 ROC-AUC score for predicting patient status, with the curve and Cox-Hazard model indicating a p-value less than 0.005 for estrogen and progesterone status, showing a strong correlation between these estrogen and progesterone status subtypes and patient outcomes. Based on those results, for goal 2, two datasets are utilized containing patients' genetic information on their dysregulation of genes to develop logistic regression models predicting estrogen, progesterone, HER2 status, and triple-negative breast cancer which had cross-validation accuracy of 85%, 69%, 89%, and 92% respectively. Genes significantly associated with estrogen-positive, progesterone-positive, and HER2-positive cancers were associated with genes such as myosin-isoform genes and plectin-isoform. In contrast, genes associated with estrogen-negative, progesterone-negative, HER2-negative, and triple-negative cancers included DNA repair protein isoform genes and unconventional myosin genes. By identifying new biomarkers such as myosin-isoform genes, plectin-isoform genes, DNA repair protein isoform genes, and unconventional myosin genes correlated with different cancer

subtypes, these models help better understand and recognize the unique behavior of cancer for each patient through which patients get better targeted personalized treatments, necessary to effectively treat and save breast cancer patients today.

Introduction/Background Research

Through biomarker tests for patients, doctors identify the biological markers present in a patient's cancer, and then their cancer is subtyped into one of the subtypes such as progesterone receptor, estrogen receptor, HER2 status, N stage, Grade, and more [5]. This classification is crucial as different subtypes may respond differently to treatments and have different prognoses [5]. For example, hormone receptor-positive breast cancers, such as estrogen-receptor-positive or progesterone-receptor-positive subtypes, typically respond well to hormone therapy [7]. On the other hand, HER2-positive breast cancers may benefit from targeted therapies directed at HER2 receptors [8]. Understanding these subtypes is also necessary for determining patient outcomes accurately, as breast cancer patients with certain subtypes and biological markers may have a better survival chance than other patients [9]. However, these biological markers such as estrogen status, progesterone status, HER2 status, A stage, Grade, and more have their limitations when it comes to doctors selecting treatments. Typically by identifying these biological markers, doctors can select treatments based on what they determine as the subtype of breast cancer the patient has, but these biological markers only inform what treatment the patient can have and do not inform about their response to that treatment. Additionally, for deceptive cancer types that exhibit behaviors of another cancer type, such as an estrogen-positive cancer behaving as an estrogen-negative cancer, it is difficult to tell from just these biological markers which subtype the patients' cancer is behaving as, making it difficult to treat [10]. This has caused a large number of treatments to which patients have no response, risking patients' health and lives [10]. According to the National Library of Medicine, research was done by Nikolaos Batis, Jill M. Brooks, Karl Payne, Neil Sharma, Paul Nankivell, and Hisham Mehanna et. al (2021) about the "lack of predictive tools for conventional and targeted cancer therapy," suggests there is a growing need for identifying new biomarkers that can help not only give more personalized treatments to patients but also predict the response they can have [10]. This necessitates deeper biological markers to be explored such as gene dysregulation, protein expression profiles, mutations, and more. However, there is a lack of research and predictive tools made to explore new biomarkers [10]. Thus, this project explores new biomarkers that influence specific breast cancer subtypes through the use of patient data and machine learning models and techniques.

To identify these biomarkers that significantly affect patient outcomes and explore new biomarkers through gene dysregulations, machine learning models in the programming language Python are employed and designed in this research. Machine learning models were chosen for this research because they can handle the complexity and variability of genetic data, which is crucial when dealing with the diverse subtypes of breast cancer and is helpful to utilize for the goals of this research. In past studies, machine learning models, such as the systematic review

conducted by Sadam Hussain et al. (2024), machine learning, and deep learning (DL) techniques have been instrumental in advancing breast cancer risk prediction [11]. Their research, published in *Frontiers in Oncology*, examined the application of AI techniques to digital mammography, radiomics, genomics, and clinical information, highlighting how these models outperform traditional risk prediction approaches [11]. This review identified imaging and non-imaging features used in breast cancer risk assessment and discussed the development of DL methods for this purpose [11]. Specifically, Hussain and his colleagues reviewed models that incorporated a variety of factors, such as mammographic density, mammographic texture, and genomics, to predict breast cancer risk more accurately [11]. Their analysis demonstrated that these AI-driven models could significantly improve the predictive accuracy over conventional models like the Breast Cancer Risk Assessment Tool (BCRAT), which primarily relies on demographic and patient clinical history [11]. Building on the work of these researchers, this study expands on their findings by focusing on gene dysregulations as potential new biomarkers for breast cancer risk. This approach not only aims to identify known biomarkers but also seeks to uncover novel genetic indicators that could lead to more personalized and effective breast cancer treatment strategies.

In this study, several machine learning models are used, including Logistic Regression, Cox-Hazard Models, and Kaplan-Meier Curves. Logistic Regression is particularly useful for binary classification tasks, such as predicting whether a patient will respond to a treatment or not, and for this research providing a probability score that helps in understanding the likelihood of a patient's survival and which biomarkers are most influential. Cox-Hazard Models, on the other hand, are employed to explore the association between the survival time of patients and various predictor variables, such as specific biomarkers [12]. These models are invaluable in medical research for understanding how different factors impact survival outcomes. Additionally, Kaplan-Meier Curves are used to estimate the survival function from lifetime data, offering a visual representation of the proportion of patients surviving over time and how different biomarkers influence these rates [13]. To train and test these models, this study utilizes three publicly available breast cancer datasets sourced from Kaggle. For the first goal, a Kaggle dataset containing data about 4,204 patients' tumor characteristics, receptor statuses, biomarkers, such as N stage, T stage, grade, estrogen status, progesterone status, etc., demographic details, survival months, and more [14]. All three of these models are utilized to identify specifically which biomarkers impact patient survivability the most and to investigate the first goal. After identifying those biomarkers, for goal 2 two other datasets from Kaggle are utilized to identify specific gene dysregulations and mutations, new biomarkers, that are associated with the identified biomarkers in goal 1 [15], [16]. These two datasets contain data about 90 patients' estrogen status, progesterone status, HER2 status, RNA-seq data (gene data), and more [15],[16]. These two datasets are combined and then used to train and test four logistic regression models which identify which gene dysregulations are associated with the previously identified biomarkers. These gene dysregulations are then further analyzed to understand their potential role in those identified biomarkers of breast cancer subtypes and responses. This study goes

beyond traditional approaches by not only identifying existing biomarkers that influence breast cancer outcomes but also exploring new, previously unrecognized gene dysregulations that may play a critical role in patient survival and treatment response. By leveraging advanced machine learning models to analyze complex genetic data, this study contributes to the emerging field of personalized medicine, offering the potential to develop more targeted and effective therapies for breast cancer patients.

Methods

Goal 1: Identifying Which Biomarkers Most Impact Patient Survival

Data

To investigate the first goal, identifying which biomarkers most impact patient survival, a dataset sourced from Kaggle titled "Breast Cancer Dataset," created by Reihane Namdari is utilized [14]. This dataset contains information on 4,204 patients diagnosed with invasive ductal carcinoma [14]. The dataset included a range of attributes such as demographic details, tumor characteristics, receptor statuses (e.g., estrogen and progesterone), and survival information about patients such as how long they survive. Using this dataset the steps to developing the three models, the Logistic Regression, Kaplan-Meier Curve, and Cox-Hazard model, are to 1) preprocess the data 2) implement feature scaling 3) implement the logistic regression model 4) implement the Kaplan-meier curve and 5) implement Cox-Hazard model.

Preprocessing

To prepare the dataset for analysis, categorical variables were first converted into numerical values using the 'LabelEncoder' from the 'sklearn.preprocessing library' in Python. This step was necessary because machine learning models typically require numerical inputs. The categorical variables included patient age, race, marital status, various tumor stages (T, N, and A stages), differentiation grade, tumor size, receptor statuses, and survival status.

Next, the dataset is split into features (X) and target variables (y). The features included all relevant patient information except for survival months and survival status, as survival months were used as the target variable for regression models. The target variable for this goal was survival months, which represents the time each patient survived following their diagnosis.

After separating the features and target, the dataset is divided into training and testing sets using an 80-20 split. This split ensured that 80% of the data was used to train the models, while the remaining 20% was used to evaluate their performance.

Feature Scaling

When working with data in machine learning, it is common for different features (or attributes) in the dataset to have different ranges of values. For example, one feature might be a person's age (which could range from 0 to 100), while another might be the size of a tumor

(which could range from 0 to several thousand millimeters). If this data is inputted into the unadjusted, the model might give undue importance to the features with larger values simply because of their scale, not because they are more important.

To address this, a technique called ‘StandardScaler’ is utilized in this research. This tool helps by standardizing the features, which means it adjusts the values so that they all have a similar range [17]. Specifically, StandardScaler works by subtracting the mean (average) value of each feature from the data points and then dividing by the standard deviation (a measure of how spread out the values are) [17]. The result is that each feature now has a mean of 0 and a standard deviation of 1, making them more comparable. By standardizing the features, it is ensured that the model treats each feature more fairly, leading to better and more reliable predictions.

Model Implementation

Logistic Regression and ElasticNet

For predicting survival outcomes, logistic regression was utilized along with ElasticNet models. Logistic regression, implemented using the ‘LogisticRegression’ class from ‘sklearn.linear_model,’ served as a baseline model. This model provided an initial understanding of the relationship between the selected features and survival outcomes. To enhance the model’s predictive power and mitigate potential overfitting, ElasticNet was implemented. ElasticNet is a regularization technique that combines both L1 (lasso) and L2 (ridge) penalties, balancing the simplicity and accuracy of the model. The ‘ElasticNetCV’ class from ‘sklearn.linear_model’ was used for this purpose, allowing for cross-validation to determine the optimal mix of L1 and L2 penalties, thereby ensuring the model's robustness across different subsets of the data.

Survival Analysis with Kaplan-Meier and Cox Proportional-Hazards Models

For the survival analysis, Kaplan-Meier estimators and Cox Proportional-Hazards are additionally utilized models to investigate how different receptor statuses (e.g., estrogen and progesterone) impacted patient survival probabilities. This additional step helps verify the results of the Logistic Regression model and double-checks the biomarkers identified.

The ‘Kaplan-Meier estimator’, implemented using the ‘KaplanMeierFitter’ from the lifelines library, was used to plot survival curves for patients with positive and negative receptor statuses. This visual representation allowed for the comparison of the survival distributions of different subtypes over time.

To quantify the impact of different variables on survival, the Cox Proportional-Hazards model was applied using the CoxPHFitter class, also from the lifelines library. This model provided hazard ratios, which indicate the relative risk of death associated with specific covariates (e.g., receptor statuses). The Cox-Hazard model was also essential for identifying which biomarkers had a statistically significant influence on patient survival and verified the biomarkers from the Logistic Regression model to check if they were accurate and matched up.

Model Evaluation

To evaluate the Cox Proportional-Hazards model, the significance of various covariates through their p-values was examined, using a threshold of 0.05 to determine which factors significantly influenced survival outcomes. In simpler terms, p-values helped identify which variables, like hormone receptor status, were most likely to affect patient survival [18]. For the logistic regression model, which predicts whether a patient is likely to survive based on their data, several metrics were used. Accuracy measured how often the model correctly predicted survival or non-survival, while the ROC AUC score (Receiver Operating Characteristic Area Under the Curve) assessed the model's ability to distinguish between patients who survive and those who don't—a higher score indicating better performance [19]. Additionally, precision-recall metrics were used to evaluate the model's ability to accurately predict positive cases, such as correctly identifying patients who survive [19]. The evaluations showed a strong correlation between progesterone and estrogen status with survival months, identifying these factors as the most influential in predicting patient outcomes. Furthermore, Kaplan-Meier curves, a method of survival analysis that estimates the survival function from patient data, revealed that patients with estrogen-positive and progesterone-positive breast cancer had higher survival rates and longer survival months compared to those with estrogen-negative and progesterone-negative breast cancer. This indicates that positive estrogen and progesterone receptors are associated with a better prognosis in terms of survival duration. From this goal, it was identified that estrogen and progesterone were influential biomarkers on patient survivability and thus specific subtypes of estrogen and progesterone status such as estrogen-positive or estrogen-negative cancer will be examined in goal 2 to find the associated gene dysregulations.

Goal 2: Identify New Biomarkers In Identified Breast Cancer Subtypes With RNA-seq

Data

To achieve the second goal, identifying novel biomarkers in breast cancer subtypes using RNA-seq data, two datasets were utilized. The first dataset, sourced from Kaggle and named 'Breast Cancer Proteomes' by Kajot, contains detailed proteomic and clinical information on breast cancer patients, including genetic dysregulations and mutations across various breast cancer subtypes [16]. The second dataset, a subset of 'Breast Cancer Proteomes,' provides biomarker data corresponding to the patients in the dataset with the proteomic data [15]. This integration of datasets allowed for a comprehensive analysis of genetic dysregulations to known breast cancer subtypes, including estrogen-receptor-positive, progesterone-receptor-positive, HER2-positive, and triple-negative breast cancer.

Data Preprocessing

The first step in preprocessing involved aligning the datasets by patient IDs to ensure consistency between clinical data and RNA-seq information. Both datasets contained information about the same patients and each patient had a unique TCGA ID assigned, out of

which ninety patients had matching information on both datasets. Using a loop, the patient IDs in the proteome and biomarker dataset were matched into a new dataset that combined the matching IDs proteome and biomarker data. Subsequently, only those patient records present in both datasets were retained for further analysis, ensuring that all patients included had both clinical and RNA-seq data available.

To handle missing data, imputation techniques were employed using the ‘SimpleImputer’ from the ‘sklearn.impute’ library. Imputation works by replacing missing values in the dataset with substitute values, ensuring that the dataset remains usable for analysis, and was necessary here to ensure that the machine learning models could operate effectively on a complete dataset [20]. After imputation, categorical variables (such as patient IDs) were encoded into numerical values using ‘LabelEncoder’ from the ‘sklearn.preprocessing’ library.

Feature Selection

Feature selection was a critical step in identifying the most relevant genetic markers associated with specific breast cancer subtypes. This process involved scaling features using ‘StandardScaler’ to normalize the data, ensuring that all gene expression levels are on a comparable scale. Feature selection was performed using ‘LinearSVC’ with an ‘L1 penalty,’ which helps to identify the most relevant genes by reducing less important ones to zero. This approach simplifies the model and focuses on key gene expression patterns that are most predictive of breast cancer subtypes, aiding in the analysis of how these patterns correlate with patient outcomes.

Model Implementation

For each breast cancer subtype—estrogen-receptor-positive, progesterone-receptor-positive, HER2-positive, and triple-negative—logistic regression models were implemented to predict the presence of these subtypes based on the RNA-seq data. The logistic regression models were configured using the LogisticRegressionCV class from ‘sklearn.linear_model’, employing elastic net regularization. This combination of L1 and L2 regularization was chosen to prevent overfitting and to manage the high-dimensional nature of the RNA-seq data.

The hyperparameters for the logistic regression models were optimized through cross-validation using GridSearchCV, allowing for the tuning of the regularization parameter and other model settings to achieve the best predictive performance. Cross-validation was conducted with a five-fold split to ensure that the models were robust and generalizable.

Model Evaluation

To evaluate the performance of the logistic regression models, several metrics were utilized, including accuracy, ROC-AUC, and precision-recall curves. Accuracy measured the overall correctness of the model predictions, while ROC-AUC provided insights into the model's ability to distinguish between the presence and absence of specific breast cancer subtypes.

Precision-recall curves were also generated to assess the model's ability to accurately predict positive cases, particularly in the context of imbalanced data.

The models achieved cross-validation accuracy scores of 85%, 69%, 89%, and 92% for predicting estrogen-receptor-positive, progesterone-receptor-positive, HER2-positive, and triple-negative breast cancer, respectively. These results indicated a high level of predictive performance, particularly for HER2-positive and triple-negative subtypes.

Biomarker Identification

Through the logistic regression models, several genes were identified as significantly associated with specific breast cancer subtypes. For instance, myosin-isoform genes and plectin-isoform genes were strongly associated with estrogen-positive and progesterone-positive breast cancers. In contrast, DNA repair protein isoform genes and unconventional myosin genes were linked with estrogen-negative, progesterone-negative, and HER2-negative breast cancers. These identified biomarkers offer potential new targets for personalized treatment strategies in breast cancer therapy.

Results

Goal 1: Identifying Which Biomarkers Most Impact Patient Survival

Logistic Regression Model (Survival Months Prediction)

```
Cross-Validation Scores: [ 0.84720497 0.84596273 0.86459627 0.85714286 0.8619403 ]
Mean Cross-Validation Score: 0.8553694261611199
Accuracy: 0.86
Classification Report:
ROC AUC Score: 0.75
```

Figure 1: Cross Validation Scores, Accuracy Scores, and ROC AUC score for the Logistic Regression Model.

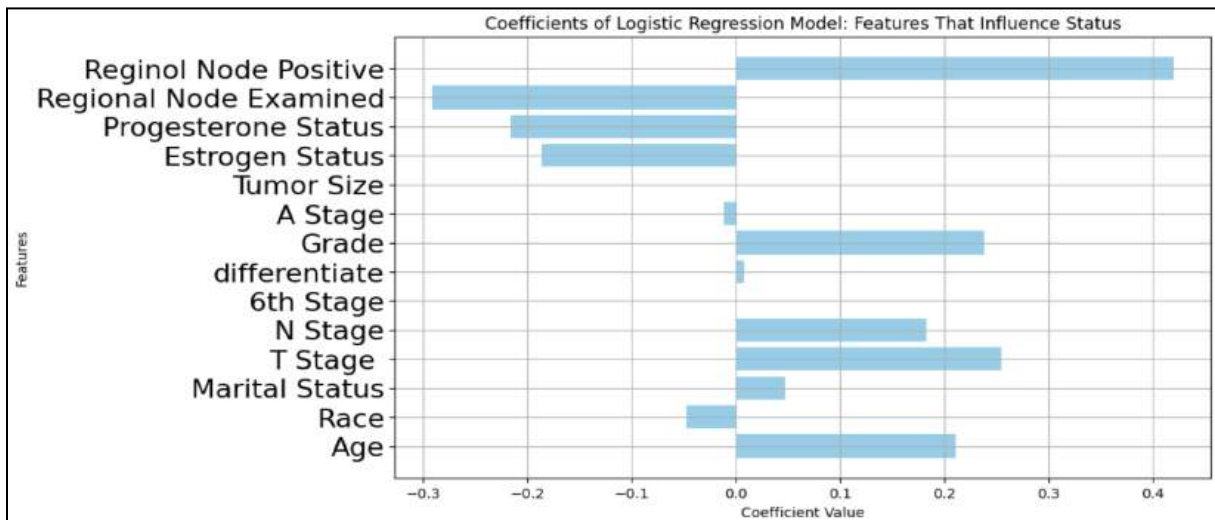


Figure 2: Coefficient graph from the Logistic Regression model showing biomarkers that most influence patient status (dead or alive).

The logistic regression model resulted in a mean validation score of 85%, an accuracy of 86%, and a ROC AUC score of 0.75 (Figure 1). These metrics provide insight into the model's performance. The cross-validation scores indicate the model's performance consistency across different subsets of the data, with a mean validation score of 85%, highlighting a solid level of reliability. The ROC AUC score, which measures the model's ability to distinguish between classes, was 0.75, suggesting a fair level of discrimination in predicting patient outcomes. Regarding the coefficients of the model, they represent the relationship between each biomarker and the patient's status. A positive coefficient indicates that an increase in that feature raises the likelihood of the patient being alive, while a negative coefficient suggests the opposite (Figure 2). In this case, progesterone and estrogen statuses had the most significant negative coefficients, implying that negative statuses in these biomarkers are strongly associated with poorer outcomes for the patients. While regional node-positive and regional node examined are shown as important factors, they are not as influential as the hormonal biomarkers (estrogen and progesterone status) [21]. This finding emphasizes the importance of hormonal factors over traditional pathological features in predicting patient outcomes, which is crucial for Goal 1 of the research, where understanding the key predictors of patient survival is paramount.

Cox-Hazard Model

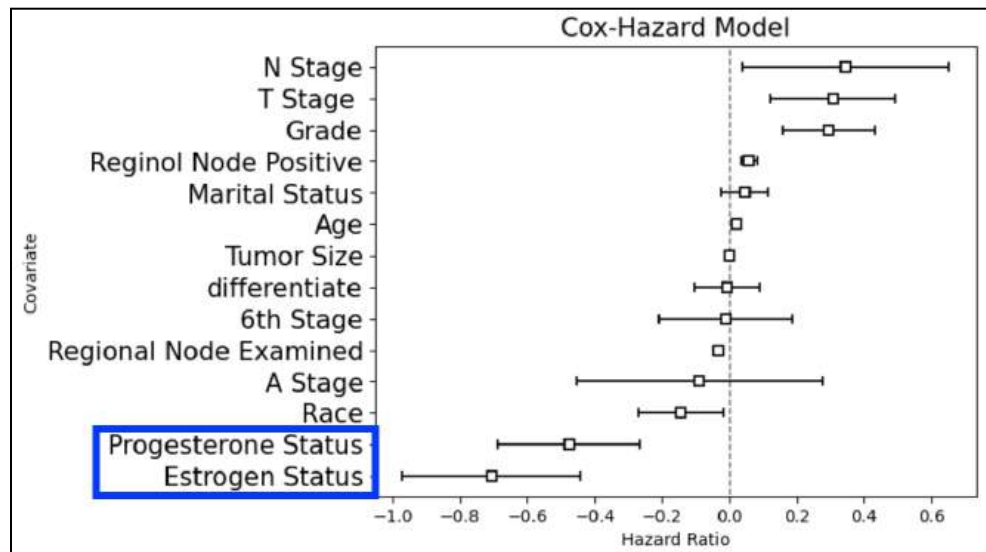


Figure 3: Cox-Hazard Model showing covariate variables (biomarkers) and their hazard ratios with 95% confidence level.

The Cox-Hazard Model indicated that there was a strong correlation between estrogen and progesterone status and the survivability of a patient. For estrogen and progesterone status, there were negative hazard ratios, indicating that higher estrogen and progesterone values correspond to a lower hazard rate, as shown in Figure 3, which in this context means a longer survival time for the patient. A negative hazard ratio suggests that as the value of these

biomarkers increases, the risk of the event (in this case, death) decreases, meaning patients with higher estrogen and progesterone levels are more likely to survive longer.

This result is critical for Goal 1, as it confirms that estrogen and progesterone receptor statuses are significant predictors of patient survival. These findings will be used to refine predictive models and treatment strategies, ensuring that patients with different estrogen and progesterone statuses receive personalized treatment plans that consider their likelihood of survival. This matches with the findings of the logistic regression model as both of these models indicate that estrogen and progesterone status have a significant impact on patient survival.

Kaplan-Meier Curve

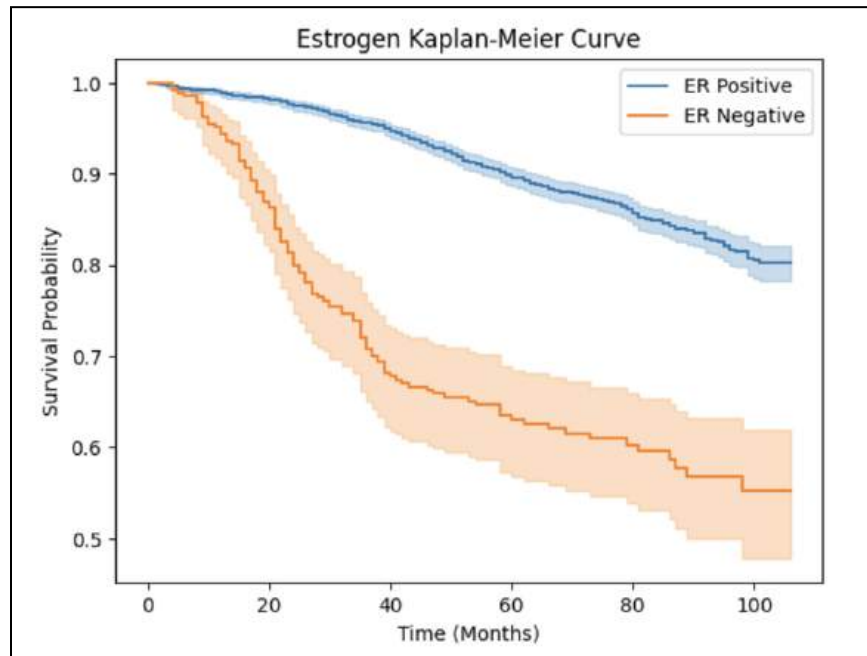


Figure 4: Estrogen Kaplan-Meier Curve showing survival probability in months for patients with Estrogen Positive and Estrogen Negative Cancer with 95% confidence level.

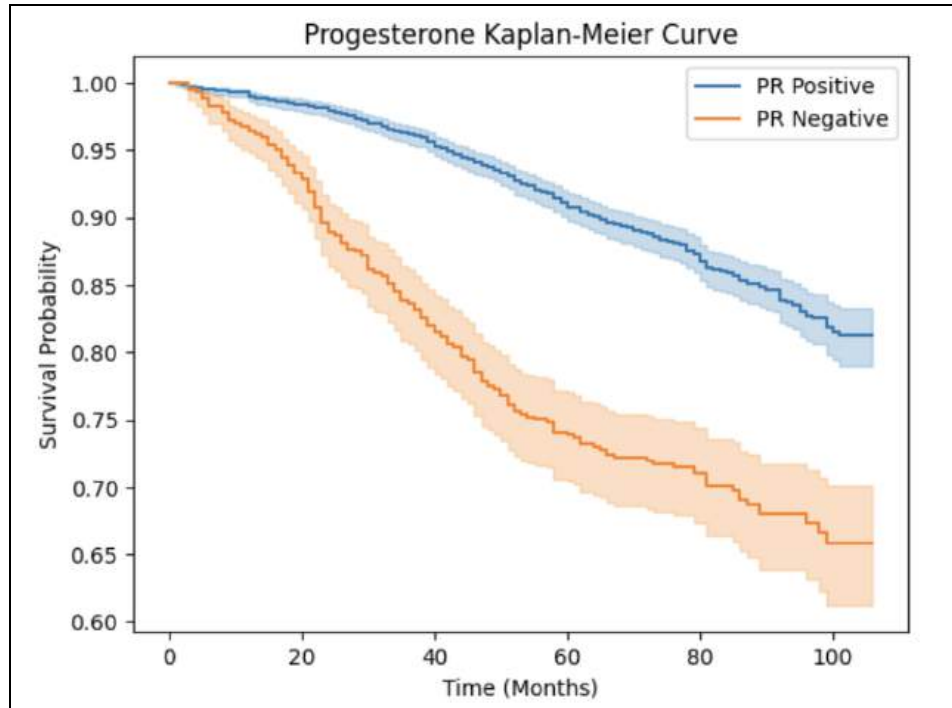


Figure 5: Progesterone Kaplan-Meier Curve showing survival probability in months for patients with Progesterone Positive and Progesterone Negative Cancer with 95% confidence level.

The Kaplan-Meier Curve (Figure 4-5) shows that estrogen and progesterone status have a significant impact on the survival months of a patient for estrogen and progesterone-positive breast cancer the survival months for the patient were while for estrogen and progesterone-negative breast cancer was lower, this confirms the findings of the Logistic Regression and Cox-Hazard model which indicated that progesterone status and estrogen status have a significant impact on patient survival. The curve had a p-value of less than 0.005, indicating that there is a significant difference between the survival months of estrogen and progesterone-positive breast cancer and estrogen and progesterone-negative breast cancer.

These findings are crucial for Goal 1, as they highlight the importance of hormone receptor status in predicting patient outcomes. By identifying the distinct survival patterns between hormone receptor-positive and hormone receptor-negative patients, this analysis underscores the need for personalized treatment strategies. For instance, patients with estrogen or progesterone-positive status may benefit from hormone therapies, which can significantly extend survival. Conversely, patients with negative receptor status may require alternative therapeutic approaches. This matches with the findings of the Cox-Hazard model as they both indicate that as estrogen status and progesterone status become more negative, the survival rate decreases.

In summary, Goal 1's analysis demonstrated that the Logistic Regression, Cox-Hazard, and Kaplan-Meier, models consistently identified estrogen and progesterone statuses as key factors influencing patient survival. The results from these models will be instrumental in

developing targeted interventions and improving patient outcomes based on their specific biomarker profiles.

Goal 2: Identify New Biomarkers In Identified Breast Cancer Subtypes With RNA-seq

From goal 1, it was identified that estrogen and progesterone status were most influential on patient outcome, and for goal 2 these will be used to identify specific gene mutations within these biomarkers subtypes (estrogen-positive, estrogen-negative, progesterone-positive, and progesterone-negative) are most associated with estrogen and progesterone status. In addition to these two biomarkers, in goal 2 HER2-status will also be investigated because HER2-status is another critical biomarker that, along with estrogen and progesterone statuses, plays a significant role in breast cancer prognosis and response to treatment. By analyzing HER2 status alongside estrogen and progesterone statuses, Goal 2 aims to provide a more comprehensive understanding of how these biomarkers interact and influence patient outcomes, allowing for more precise identification of gene mutations contributing to breast cancer progression and therapy resistance. Additionally, triple-negative breast cancer, when estrogen, progesterone, and HER2 status are all negative, is investigated and gene dysregulations are associated with triple-negative breast cancer.

Estrogen Status Logistic Regression model

Cross-Validation Scores: [0.93333333 0.93333333 0.8	0.8	0.78571429]
Mean Cross-Validation Score: 0.8504761904761905		
ROC AUC Score: 0.9335664335664335		

Figure 6: Cross Validation Scores, Accuracy Scores, and ROC AUC score for the Estrogen Status Logistic Regression Model.

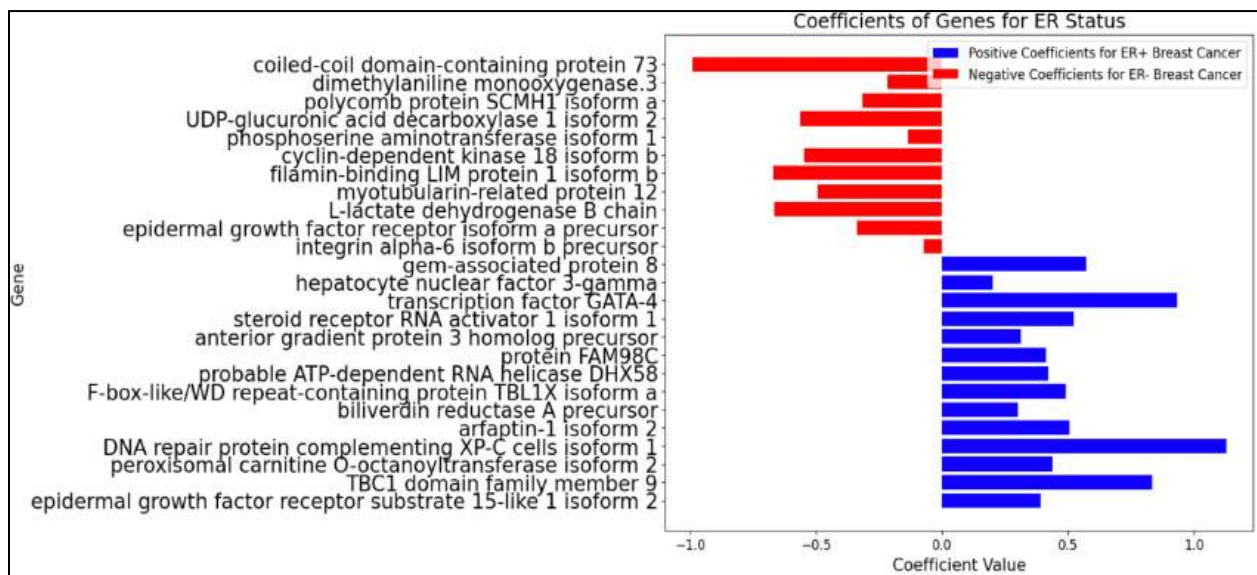


Figure 7: Coefficient graph with the most impactful genes on Estrogen Positive and Negative Cancer and their associated coefficients.

The estrogen status logistic regression model had a mean cross-validation score of 85% and a ROC AUC score of 0.93 as shown in Figure 6. The genes that were correlated the highest with the subtype estrogen-negative breast cancer were Coiled-coil domain-containing protein 73 (CCDC73), Cyclin-dependent kinase 18 isoform b (CDK18), and L-lactate dehydrogenase B chain (LDHB) as shown in Figure 7 by the red bars. There is one commonality amongst these genes in that these genes all take part in a process called tumorigenesis, a process by which normal cells turn into cancerous cells [22], [23], [24]. In estrogen-negative breast cancer, disruptions in estrogen signaling pathways can contribute to aberrant cell cycle progression and increased cell proliferation which these genes do [25]. The genes that were correlated the highest with the subtype estrogen-positive breast cancer were DNA repair protein complementing XP-C cells isoform 1 (XPC), Transcription factor GATA-4 (GATA-4), and TBC1 domain family member 9 (TBC1D9) as shown in Figure 7 by the blue bars. Genes like XPC, GATA-4, and TBC1D9 are involved in pathways related to cell growth, differentiation, and hormonal signaling [26], [27], [28]. Their dysregulation can particularly impact estrogen-positive breast cancer by influencing estrogen-mediated signaling cascades, cell cycle progression, and gene expression patterns associated with estrogen response, ultimately contributing to the growth and progression of estrogen-positive breast tumors [26], [27], [28].

Progesterone Status Logistic Regression Model

Cross-Validation Scores: [0.75	0.5	0.75	0.625	0.57142857	0.71428571
0.71428571	0.71428571	0.71428571	0.71428571]		
Mean Cross-Validation Score: 0.6767857142857143					
ROC AUC Score: 0.7358630952380951					

Figure 8: Cross Validation Scores, Accuracy Scores, and ROC AUC score for the Progesterone Status Logistic Regression Model

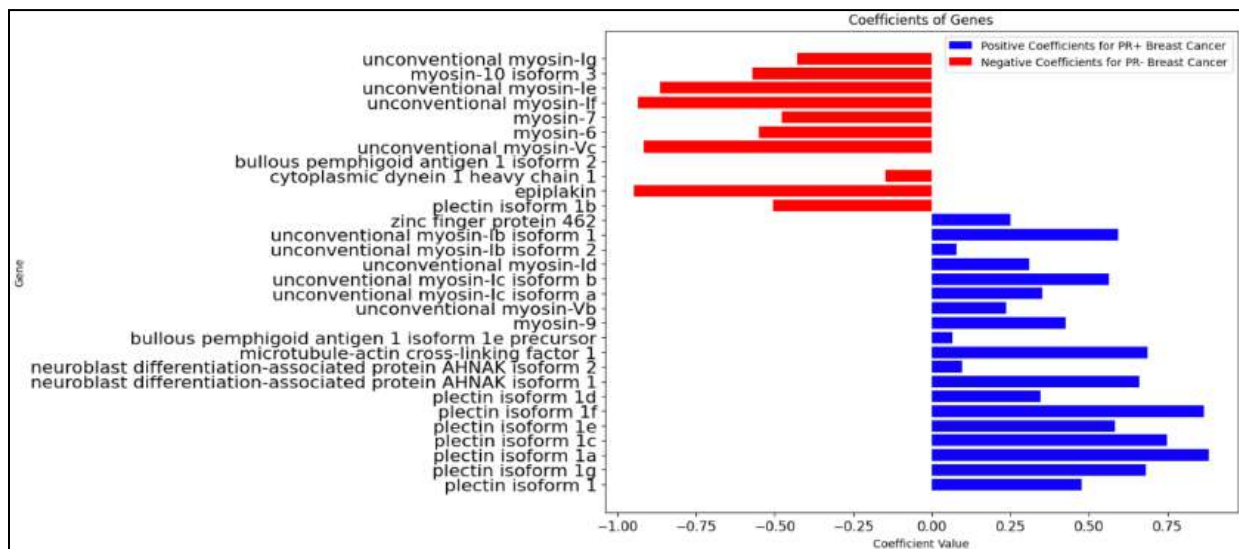


Figure 9: Coefficient graph with the most impactful genes on Progesterone Positive and Negative Cancer and their associated coefficients.

The progesterone status logistic regression model had a mean cross-validation score of 69% and a ROC AUC of 0.75 as shown in Figure 8. The genes that were highly correlated with progesterone-negative breast cancer were unconventional myosin-1f, unconventional myosin-1c, and Epiplakin as shown in Figure 9 by the red bars. Progesterone-negative breast cancer cells lack receptors for the hormone progesterone, leading to altered cellular responses and proliferation independent of progesterone signaling [29], [30]. Dysregulation of these genes can impact cell adhesion dynamics, cytoskeleton rearrangement, and metastatic potential, contributing to the aggressive behavior often observed in progesterone-negative breast cancer [29], [30]. The genes that were highly correlated with progesterone-positive breast cancer were plectin isoform 1f, plectin isoform 1a, and plectin isoform 1c as shown in Figure 9 by the blue bars. The genes are strongly correlated with progesterone-positive breast cancer as they promote stable cell-cell and cell-extracellular matrix interactions, regulating cytoskeletal components, and suppressing metastatic processes, collectively contributing to the characteristics of progesterone-positive breast cancer, such as slower cell proliferation rates, lower invasiveness, and reduced metastatic potential compared to progesterone-negative breast cancer subtypes [31].

HER2-Final Status Logistic Regression model

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Cross-Validation Scores: [0.94736842 0.88888889 0.88888889 0.83333333]
Mean Cross-Validation Score: 0.8896198830409356
ROC AUC Score: 0.8897058823529411

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Figure 11: Cross Validation Scores, Accuracy Scores, and ROC AUC score for the HER2-Final Status Logistic Regression Model

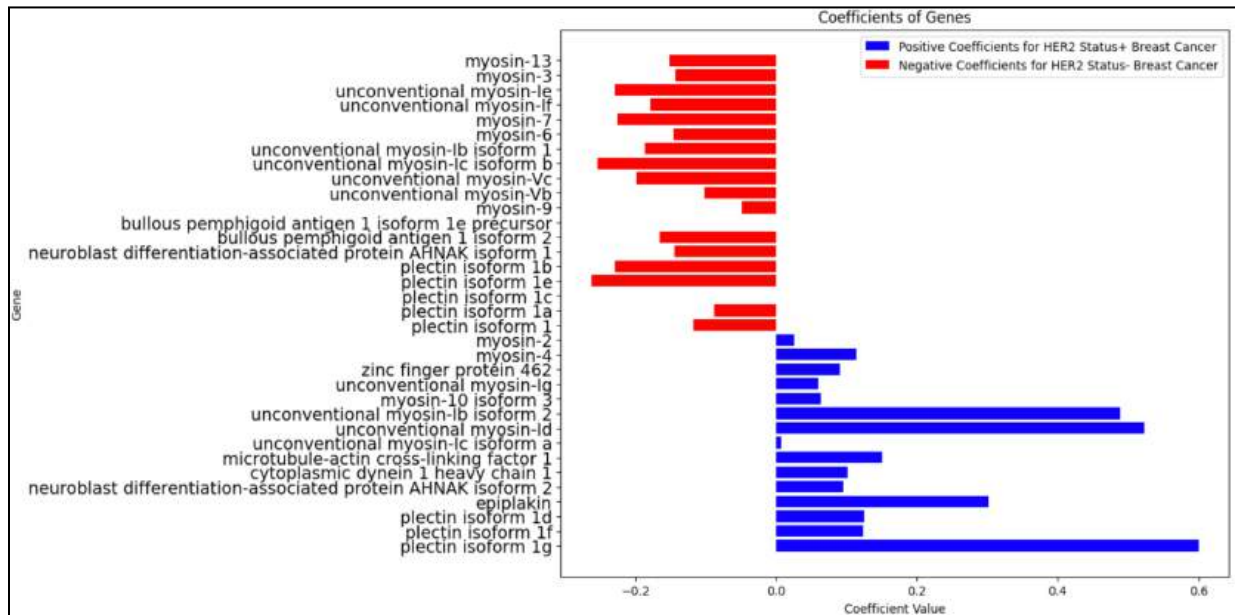


Figure 12: Coefficient graph with the most impactful genes on HER2-Final Status Positive and Negative Cancer and their associated coefficients.

The HER2-Final Status logistic regression model had a mean cross-validation score of 89% and an ROC AUC score of 0.89 as shown in Figure 11. The genes that were highly correlated with HER2-Final negative breast cancer were unconventional myosin-Vc, unconventional myosin-lc isoform b, and plectin isoform 1e as shown in Figure 12 by the red bars. These genes are involved in critical cellular processes that maintain cell structure and regulate cell movement [32], [33]. Unconventional myosins participate in intracellular transport and cytoskeletal dynamics, crucial for cell movement and division [32]. Their roles indicate that these genes may contribute to maintaining normal cell structure and movement regulation in HER2-Final negative breast cancer, where cells typically exhibit less aggressive behavior and reduced metastatic potential [32], [33]. The genes that were highly correlated with HER2-Final positive breast cancer were plectin isoform 1g, unconventional myosin-ld, and unconventional myosin-lb isoform 2 as shown in Figure 12 by the blue bars. The genes are closely associated with cellular processes crucial for cancer progression in HER2-positive breast cancer [34]. These genes play a role in cell adhesion, cytoskeleton organization, and tissue integrity, potentially promoting cell proliferation and invasion in HER2-positive tumors [34].

Triple-Negative Logistic Regression Model

Cross-Validation Scores for TNBC Prediction: [0.92	0.96	0.91666667]
Mean Cross-Validation Score for TNBC Prediction: 0.9322222222222222		
ROC AUC Score for TNBC Prediction: 0.9545923632610938		

Figure 13: Cross Validation Scores, Accuracy Scores, and ROC AUC score for the Triple-Negative Logistic Regression Model.

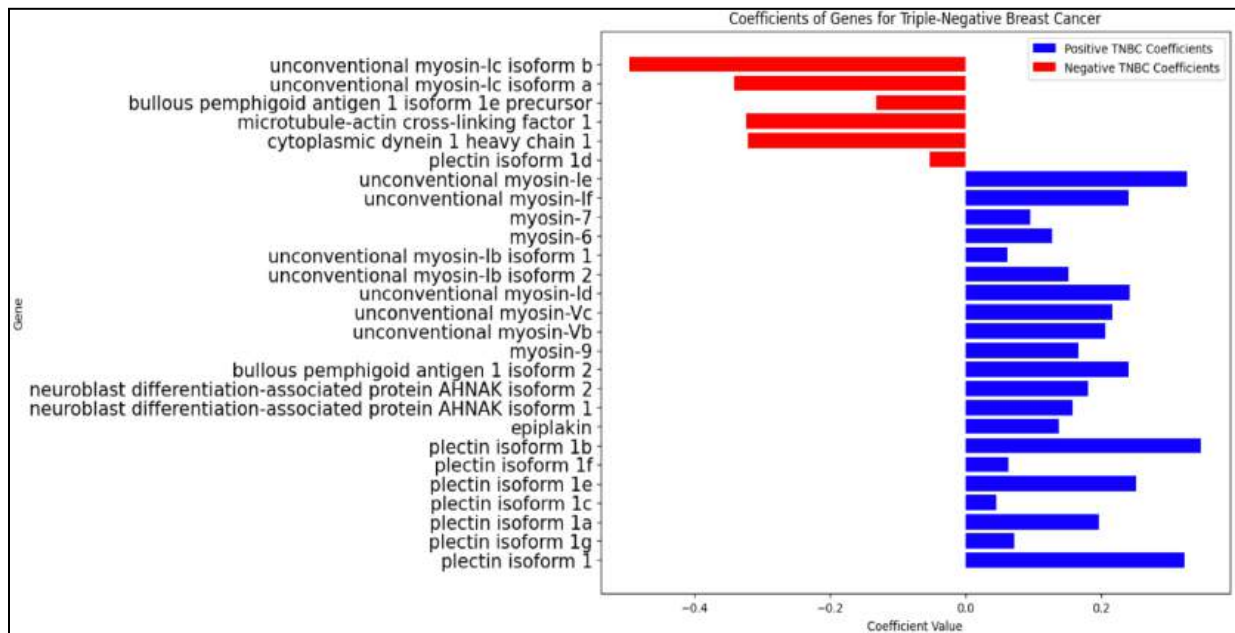


Figure 14: Coefficient graph with the most impactful genes on Triple-negative breast Cancer and their associated coefficients.

The triple-negative logistic regression model had a mean cross-validation score of 93% and a ROC AUC score of 0.95 as shown in Figure 13. The dysregulated genes that were highly correlated with the probability of a patient having triple-negative breast cancer or one behaving as such were: unconventional myosin-1c isoform b, unconventional myosin-1c isoform a, and cytoplasmic dynein 1 heavy chain 1 as shown in the red bars in Figure 14. The more these genes are expressed in terms of negative coefficients, the more likely the breast cancer is to be triple-negative breast cancer. All of these genes play critical roles in cell adhesion, cytoskeletal dynamics, and intracellular transport processes [35], [36]. Dysregulation in these genes can lead to disruptions in cellular structural integrity, abnormal cell migration, and increased invasive potential, all of which are characteristic features of aggressive cancers like triple-negative breast cancer [35], [36].

Conclusions and Further Research

Through this project it was found that the specific biological markers, estrogen and progesterone status, strongly correlate to patients' survival months and status and additionally, specific subtypes defined by these biological markers such as estrogen and progesterone positive cancer and estrogen and progesterone negative cancer. Estrogen and progesterone-positive cancer indicated that the patients' survivability would increase while estrogen and progesterone-negative cancer showed the opposite. These results are accurate as positive estrogen and progesterone statuses usually indicate higher survivability for a majority of patients. This helped accomplish goal 1, which was to find which biomarker influences patient survivability. For the second goal, it was found that gene dysregulations that mainly fall in the categories of plectin-isoform, unconventional-myosin genes, and cytoskeletal dynamic genes and were strongly correlated with triple-negative, HER2-Final status, and estrogen/progesterone positive/negative cancers, and these genes' functions have a strong correlation to these cancers. These findings emphasize the need to explore deeper biological markers beyond traditional ones for doctors to identify more personalized treatments for patients and better patient responses to these treatments. For further research and improvements, there could be more improvements about what causes these specific gene dysregulations and these estrogen/progesterone/HER2 statuses, as the relationship between the current genes and biomarkers is more correlated instead of causal. This can be done through double machine learning which is a statistical method used to identify and estimate causal effects by combining two machine learning models [37]. Additionally, for the prediction of progesterone status and triple-negative cancer, class weights have been used but more improvements can be made using SMOTE (synthetic minority over-sampling technique) to help balance out the two classes (negative and positive) and avoid model bias [38]. More data could also be used in these models such as with the genes identified in this project an improvement that can be done is to incorporate omics-data such as transcriptomics, proteomics, metabolomics, and epigenomics data to complement the gene expression profiles identified.

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How do Environmental Regulations in the UAE Impact its Economy?

By Gumingchen Yang

Abstract

This literature review explores the impact of environmental regulations on the economic landscape of the United Arab Emirates (UAE). With a focus on the interplay between sustainable development goals and economic growth, the review examines how recent policies have influenced key sectors, particularly the oil and gas industry. By analyzing both short-term economic challenges, such as decreased revenues for companies like ENOC, and long-term benefits, including economic diversification and enhanced global competitiveness, the review provides a balanced perspective on the UAE's transition towards a green economy. The study also considers the varying perspectives of stakeholders, including consumers, producers, and the government, to present a comprehensive understanding of the economic implications of these environmental policies. The findings suggest that while the UAE faces short-term challenges in adapting to stricter regulations, the long-term benefits of sustainability initiatives are likely to outweigh the initial economic disruptions, positioning the country as a leader in the green economy.

Introduction

In a time when Gross Domestic Product (GDP) serves as the core metric of countries' economic success, sustainability is often a factor that is overlooked by businesses, firms, and corporations. Thus, environmental regulations have become critical tools for managing the ecological footprint of nations that seek to maintain an environmentally friendly economy.

In the United Arab Emirates (UAE), a country rich in hydrocarbon resources while also heavily invested in diversifying its economy, these regulations play a pivotal role. The UAE has been proactive in its environmental policies to mitigate the effects of climate change and promote sustainable development. Projects such as the Dubai Clean Energy Strategy 2050, Abu Dhabi Vision 2030, and Net Zero 2050 all work towards the end goal of reducing carbon emissions and shifting all energy sources to be renewable and efficient. Specifically, according to UNFCCC, the UAE aims to reduce its greenhouse gas (GHG) emissions for the year 2030 by 23.5%, relative to the business-as-usual (BAU) scenario. The country has also invested heavily in solar energy, with a target of generating 30% of its electricity from solar power by 2030 (U.AE), as well as making strides in reducing its flaring of natural gas, a process that releases methane (a potent greenhouse gas), into the atmosphere.

In this literature review, we will be diving into how these environmental regulations and policies have impacted the United Arab Emirates's economy as a whole. Drawing research from a diverse array of sources, including websites, news articles, and scholarly papers, this analysis will unfold in a structured manner. Firstly, I will examine the potential short-term economic drawbacks these environmental policies may have instigated within the UAE. The review will then transition to consider the anticipated and potential long-term economic advantages,

illustrating a broader perspective on sustainability and growth. Furthermore, the effects of these regulations will be assessed from the viewpoints of various stakeholders—consumers, producers, and the government. Lastly, to conclude the review, I will offer a personal reflection, providing a critical insight into the overarching impact of UAE's environmental strategies on its economic landscape.

Methodology

For this literature review, I carefully selected sources with an emphasis on credibility, reliability, and relevance. Most of the information comes from government publications, official reports, and policy documents from the United Arab Emirates (UAE) and other trusted international organizations. These sources were chosen because they are produced by authoritative bodies with access to accurate data and a deep understanding of the economic impacts of environmental regulations. Reports from the UAE Ministry of Climate Change and Environment, for example, provided firsthand insights into how policies are being implemented and their effects on the economy. Alongside these government sources, I also included scholarly research papers, particularly those published in peer-reviewed journals. These academic sources were chosen for their rigorous analysis and thoughtful perspectives, which helped to put the economic consequences of environmental policies into a broader context. By relying on these types of sources, I aimed to create a well-rounded and evidence-based review of how environmental regulations are influencing the UAE's economy.

Short-Term View

To begin, I will discuss the short-term negative economic impacts of environmental regulations in the UAE. The Dubai Clean Energy Strategy, implemented by Dubai in 2015, and the Energy Strategy 2050, implemented by the UAE in 2017, both aim to transition more of the UAE's energy production to clean sources by 2050 (U.AE). These ambitious initiatives underscore the UAE's commitment to sustainable development, primarily by reducing carbon emissions. However, in the short term, such stringent environmental regulations may have had an association with posing significant challenges to the UAE's economy and corporate revenues, particularly for industries heavily reliant on fossil fuels. One possible example is the Emirates National Oil Company (ENOC), the largest oil and fuel company in the UAE. Following the announcement of the Clean Energy Strategy 2050, ENOC experienced a substantial decline in revenue in 2016.



Figure 1: ENOC’s data report on revenue and assets from 2005 to 2016 (in dollars)



Figure 2: Data report of the change in revenue of ENOC (from Macrotrends)

As seen in Figure 1 and Figure 2, reports from ENOC and data collected by Macrotrends show, that starting in 2015, there was a disrupted conventional energy market and, therefore, distorted the revenues of fossil fuel firms. The significant revenue decline for ENOC in 2016 and

2017 right after the implementation of both initiatives can mean that there may be some sort of association between the fall in the firm’s economy and the regulations. There is no clear link of causation, however, the correlation shows how it could be linked to the reduced demand for fossil fuels as Dubai transitions to renewable energy. These statistics could suggest that this trend is in line with broader economic changes being faced by oil-dependent economies adapting to sustainable practices. Beyond highlighting Dubai's commitment to this reduction in carbon emissions, the Dubai Clean Energy Strategy and Energy Strategy 2050 illustrate the economic implications for established energy companies. This is a critical element for regions such as Dubai and the other emirates in understanding the impacts from an economic perspective of advancing their green agendas through policy and strategic planning on the traditional energy sectors.

Next, according to the International Trade Administration, in order to reach net zero carbon emissions by 2050, the UAE is spending over \$160 billion on clean and renewable energy and key technologies. However, this may reveal significant economic trade-offs and sectoral shifts. The relocation of such significant funding to other sectors, such as tourism or oil and gas, may have an immediate impact on the economy. The focus on environmental sustainability ensures long-term economic transformation, yet there may be some consequences to the economy in the short term. For example, such investments could potentially catalyze changes in industries like real estate and technology and encourage eco-friendly practices to attract government funding and subsidies. Doing so would be expected to improve the overall revenues of such companies by following global sustainability trends and enhancing competitiveness.

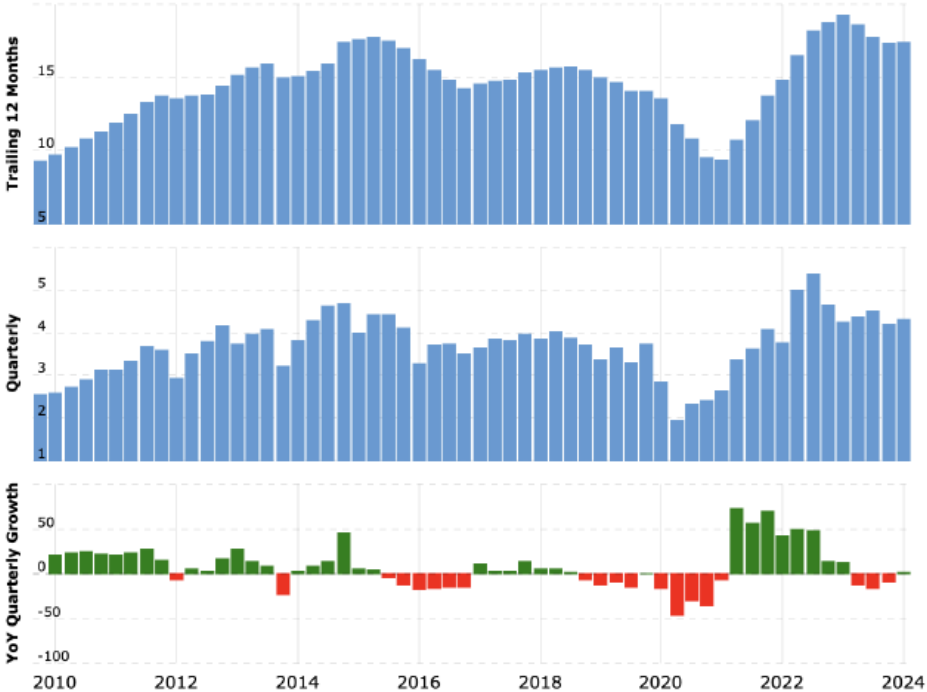


Figure 3: Data report of the change in revenue of YPF Sociedad Anonima (from Macrotrends)

The UAE has stood out in distinct interests in implementing carbon taxes (Fincirc) which may involve a possible loss of operating revenue by oil and gas firms in the state. The outcome of implementing such carbon taxes could be similar to other nations that have implemented carbon tax programs. For example, the Argentinian case on carbon taxation. As seen in Figure 3, the largest oil company in Argentina, YPF Sociedad Anonima, has registered its annual revenue reduction since 2018 when a carbon tax was imposed (Macrotrends). It remains arguable whether the whole of this decline in revenues should be credited to a carbon tax, though it stands plausible for this tax to be counted as a contributing factor. Carbon taxes increase operational costs for fossil fuel companies, which then results in a decrease in profitability and, thereafter, a decline in revenue. This is because it is the whole essence of the carbon tax to internalize the environmental costs associated with carbon emissions, thus making fossil fuels more expensive compared to clean energy sources. This economic mechanism motivates producers and induces consumers to shift towards low-carbon alternatives. The carbon tax may have further accelerated the move from oil and gas in the case of YPF, with attendant reduced demand for fossil fuels—therefore likely contributing to the fall in the firm's revenues.

Thus, while the UAE's environmental goals and regulations demonstrate a strong commitment to sustainable development, it may bring significant short-term economic challenges, particularly for traditional energy sectors. Nonetheless, these efforts are essential for long-term economic transformation, which will now be discussed.

Long-Term View

According to Wamda.com, the green technology sector in the UAE is growing by leaps and bounds, with new startups entering the fields of renewable energy, water conservation, waste management, sustainable transportation, and green buildings. In the long term, this growth may alleviate environmental challenges and, at the same time, provide economic growth and employment opportunities that ensure one of the essential cores of ecological economics. The green sector opens up an opportunity for extra employment, which can develop a chance for increased earning power for the citizens, thus further strengthening the nation's economic resilience. Besides, those advances substantiate the UAE's commitment to lowering carbon emissions in line with global sustainability goals. Such proactive measures only help enhance the reputation of the UAE on the international platform and thereby forge closer relations with other countries in closer cooperation toward environmental sustainability.

Initiatives such as the Dubai Clean Energy Strategy 2050 and the Abu Dhabi Vision 2030 implemented by the UAE government undertake effective means of implementing regulation on sustainability by increasing energy efficiency and utilizing renewable sources of energy, such as solar power. Such gradual measures can also allow businesses and consumers to adapt accordingly, thus preventing economic shocks and fostering industry innovations in using more energy-efficient processes. This not only enhances the overall setting but also helps stable economic transitions, which otherwise would contradict environmental-economic principles since there would be only ecological benefits and no tangible stability in financial settings. These

long-term sustainability goals are being integrated into the UAE, which enables smoother transitions for its industries and helps provide some contribution to global emission mitigation efforts.

The environmental policies of the UAE play a core role in a broader policy toward economic diversification away from dependence on oil, which is crucial in the long term. Investment in renewable energy technologies, such as solar power projects in Dubai and Abu Dhabi, reduces the carbon footprint and supports new economic activities that spur growth independent of oil revenues (Antwi-Boateng, 2022). Therefore, this will align with the principles of environmental economics for sustainable development and energy efficiency. The investment will also support job creation in the green technology sector and help further boost the UAE's credentials globally as a front-runner for sustainable practices (MOCCAE, 2019). The strategy, thus, supports long-run economic resilience and environmental sustainability efforts that reinforce global action toward combating climate change.

In the long term, environmental regulations that reduce pollution and improve environmental standards have significant public health benefits. According to Airdri, by decreasing the population's exposure to dangerous pollutants, such regulations will positively affect health outcomes, reducing the incidence of diseases directly correlated with poor air and water quality. This then reduces healthcare costs and increases workforce productivity since a healthier population will work better with fewer cases of absenteeism due to sickness. The results of this are economic growth through a more reliable and healthier workforce. These advantages bring out the connectedness between environmental sustainability and economic prosperity, which calls for tight ecological policies.

The UAE's investments in clean beaches, wildlife conservation schemes, and a healthy urban environment have significantly made this country become one of the significant tourist destinations. These environmental laws help protect natural resources and make urban life more comfortable, thus helping boost the tourism sector, which acts as an essential part of the country's economy. When there are better attractions for tourists, the number of visitors and, consequently, local businesses flourish while creating job opportunities. According to The National News, these measures have been supported by the advocates of sustainable tourism, thus making the UAE a top destination for any conscientious eco-tourist. This notion of sustainability in the natural environment alongside economic growth goes to show that, indeed, ecological projects can fuel economic gain and shall only prosper the UAE further in the long run.

The push towards environmental sustainability has significantly fostered innovation within the UAE, particularly in the realm of environmental economics. Programs like Masdar City, a planned city set to be sited in Abu Dhabi and based on sustainability, have catalyzed colossal research and development in clean technologies. Such developments are not only making a profound contribution toward greening its economy but also positioning the UAE as one of the world leaders in green technology. Investment in sustainable infrastructure and technology is one method of environmental economics that would emphasize efficient resource utilization and a reduction in negative externalities. The UAE, through the development of clean

technologies and cutting carbon emissions, is bound to increase its economic resilience and sustainability. Carbon minimization on a global scale subsequently opens up economic growth and diversification opportunities for the UAE. This proactive approach to environmental sustainability ensures long-term economic benefits, reinforces the UAE's competitiveness in the world market, and responds to environmental concerns.

Conclusion

Overall, the UAE's dedication to environmental regulatory measures has dramatically influenced the economic landscape. Short-term challenges and long-term benefits were evident at the onset when the policies—like the Clean Energy Strategy 2050—that were coming into effect disturbed traditional energy sectors, lowering the revenues of major large fossil fuel companies. However, these short-term economic disadvantages are very well offset by massive long-term gains. Investment in renewable energy, notably solar power, cements the UAE's strategic move toward a diversified and sustainable economy. This will not only reduce the nation's carbon footprint but also bring up green technology sectors, which will create employment and attract international investments. What is more, environmental policies ensure an enhancement in public health through pollution reduction. This, in turn, also boosts workforce productivity and keeps healthcare costs down. The fast-growing green technology industry is illustrated through projects such as Masdar City, which places the UAE at the head of the sustainable states in the world and enhances its economic resilience. Among other things, ecologically safe initiatives help consolidate the tourism sector, which is why it has gained so much popularity among ecologically aware travelers. In general, proactive environmental sustainability represents the integration of economic diversification, public health, and global competitiveness for long-term prosperity and ecological balance in the UAE.

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Learning for a Better Future: Investigating Education and Poverty in Developing Nations By Prashasth Turaga

Abstract

This paper examines the impact of education on the economic and social well-being of individuals and families in India, using PLNR Educational Society (PLNRES) as a case study to analyze the broader implications for Indian national development and the effects of education on impoverished students. As discussed in the Methods section, interviews were conducted with 23 individuals who completed their higher education using financial aid from PLNRES and are now employed or have secured employment in the near future. Previously conducted research and publications on education legislation and reform in India are described and contextualized in a discussion of poverty reduction. Based on the data collected through PLNRES and the preexisting material relevant to the subject, it is rational to conclude that an increased focus on education can financially uplift impoverished families in India.

Clarifications

“PLNRES” or “the PLNRES” both refer to the organization “PLNR Educational Society”. The letters “PLNR” are the initials of the president and founder of the organization: P. L. Narsimha Reddy.

Any mention of the “PLNRES Booklet,” “PLNRES Handbook,” “official PLNRES Booklet,” or “official PLNRES Handbook” refers to the 6th edition of the official PLNRES “Activities up to March 2023” booklet. This booklet cannot be found online. However, a relevant section of this booklet has been attached as Appendix D.

One crore is equivalent to ten million, and is written in this paper as either 1,00,00,000 (as is the convention for large numbers in India), or 10,000,000 (as is the convention for large numbers in the United States). One lakh is equivalent to one hundred thousand, and is written in this paper as either 1,00,000 (as is the convention for large numbers in India), or 100,000 (as is the convention for large numbers in the United States).

The symbols “₹,” “INR,” and “Rs.” all represent the Indian rupee. The symbols “\$” and “USD” both represent the U.S. dollar. Furthermore, “LPA” stands for “lakhs per annum,” which represents an annual salary in lakhs of Indian rupees (INR).

All beneficiaries of PLNRES, past and present, are identified by their unique ID. This ID is of the form “PLNRES/##/####” and consists of two numbers—one two-digit number and one three-digit number. The two-digit number refers to the year the student began receiving aid from PLNRES. For example, “21” would indicate that the student began receiving aid in 2021. The three-digit number, also referred to simply as the student's “serial number,” refers to the order in which aid was given to all beneficiaries of PLNRES. For example, “001” refers to the first student who received aid from PLNRES, and “176” refers to the 176th student who received aid from PLNRES. For simplicity and anonymity, all beneficiaries are referred to only by their serial

number. (However, in some cases, gender pronouns such as “he” and “she” are used, to ensure readability and clarity. This does not compromise the interviewees’ anonymity.)

Regarding the accuracy and relevance of conclusions made about the beneficiary students, a clarification of the temporal context of previously collected data is necessary. Any information collected from the PLNRES booklet is current as of March 2023. Any information collected from interviews is current as of August/September 2023. To prevent confusion, all information provided about the beneficiary students in this paper is written in the present tense, regardless of whether the information was collected through the booklet or through interviews. Further, since the true current conditions of these students is unknown, the information collected previously will be treated as if it accurately represents the students’ present situations. Therefore, any mention of a student’s “current financial situation,” for example, refers to the student’s financial situation as of March or August/September 2023.

At some points in this paper, cells in the spreadsheet provided in Appendix C are referenced. Cells will be referenced using their “cell code,” which consists of a letter followed by a three-digit number. The letter corresponds to the column in the spreadsheet—refer to Appendix C for the column that each letter refers to. (For example, H references annual “Family Income After Aid.”) The three-digit number is the student’s serial number. So, for example, cell K042 references the current profession of Student 042: Staff Nurse.

1. Background

1.1 PLNR Educational Society

PLNR Educational Society, according to the official PLNRES booklet, is a registered non-profit organization that financially supports economically disadvantaged “meritorious boys and girls, single parent students[,] and orphan students” in their pursuit of higher education. The organization was formed in 2014, and officially registered in 2017, in part by P. L. Narsimha Reddy, retired Headmaster of Zilla Parishad High School in Neredmet, Hyderabad. However, the founding members of this organization have been financially supporting students’ educations since 2012, before the official formation of PLNRES.

PLNRES is based in Hyderabad, the capital of the state of Telangana in India. Among the 23 students interviewed, all of whom are either currently employed or have secured future employment, all but three are still based in that general area (that is, in Telangana or Andhra Pradesh).

Financial assistance is provided to those students pursuing vocational courses such as “Diploma (Polytechnic), Medical Lab Technician (MLT), Accounts & Taxation (A&T), Computer Science and Engineering, Multipurpose Health Worker (Female) and ITI,” as well as “courses in IIIT (Basara), B.Sc (Nursing), B.Pharmacy, Para Medical courses, Diploma in Education and Computer Graphics and Animation” (PLNRES booklet).

PLNRES relies entirely on donations to fund its beneficiaries’ educations. As of March 31, 2023, PLNRES has 203 donor members. Eighteen of these members have contributed at least

1 lakh rupees. In the period April 1, 2022 to March 31, 2023, PLNRES received about 126 donations, totaling approximately ₹14,88,117. The figure for the total amount spent by PLNRES on financial aid in the same time period is not provided in the PLNRES booklet. However, the figure for the total amount spent by PLNRES on financial aid since their first payment is provided: PLNRES has spent about ₹49,03,925 to support students beginning in 2012, when financial aid was provided to support the education of Student 001.

1.2 Poverty in India

India is home to about 1.4 billion people, making it one of the world's most populous countries. According to the World Bank, India has a population density of 473 people per square kilometer of land area, making it the 25th most densely populated country in the world as of 2021. As of September 2022, those living on less than \$2.15 a day (based on 2017 Purchasing Power Parities [PPPs]) are considered to live in extreme poverty. In India, about 0.8% (11.2 million) are in extreme poverty as of 2020.

Another measure of poverty is Multidimensional poverty. Multidimensional poverty, or the Multidimensional Poverty Measure (MPM), is calculated using six components: consumption or income, educational attainment, educational enrollment, drinking water, sanitation, and electricity attempts. These components are mapped into three dimensions of well-being: monetary, education, and basic infrastructure services. The MPM weighs these three dimensions equally in its calculation. By encompassing a wider range of metrics relevant to poverty, the MPM paints a better picture of poverty in India. About 14.96% of people (209 million individuals) live in multidimensional poverty in India.

Compared to where India was several years ago, this is a dramatic improvement. According to the World Bank, it is estimated that India halved the portion of its population living in extreme poverty between 2011 and 2019. And as of March 2024, India has eliminated extreme poverty altogether. (This is determined by the \$1.90 poverty line, based on 2011 global PPP. The \$1.90 line is used here instead of the \$2.15 line because the former aligns more closely with the official India Tendulkar poverty line.)

From 2015 to 2021, 135 million people escaped multidimensional poverty (Timmins). But how was India able to achieve such a drastic change in their poverty levels, and how will the nation continue to address these issues?

2. Education in India

2.1 Education in Andhra Pradesh and Telangana

The information collected from PLNRES is best understood within the context of a broader discussion of education in India. Since the majority of the students interviewed are based in Andhra Pradesh (“AP”) and Telangana, this section will provide some background into education in only these two states.

The Institute for Competitiveness is an organization that “conducts & supports indigenous research; offers academic & executive courses; provides advisory services to the Corporate & the Governments and organises events.” In March 2024, they released the Report on Foundational Literacy and Numeracy. This report contains information about education in various Indian states, including the Index on Foundational Learning, also referred to as the “FLN Score.” This metric comprises 42 indicators, organized into five pillars: Educational Infrastructure, Access to Education, Basic Health, Learning Outcomes, and Governance. The Educational Infrastructure pillar comprises nine indicators, including “Percentage of schools with functional toilets” and “Percentage of schools with functional drinking water.” The Access to Education Pillar comprises eleven indicators, including “Dropout Rate - Primary” and “Pupil Teacher Ratio (PTR) - Primary.” The Basic Health Pillar comprises six indicators, including “Children under 5 years who are underweight” and “U5MR,” or “Under-five Mortality Rate.” The Learning Outcomes pillar comprises ten indicators, including “Transition Rate - Primary” and “Percentage of states/UTs exceeding global proficiency in numeracy.” The Governance pillar comprises six indicators, including “Percentage to total expenditure on primary education for Govt schools” and “Expenditure on Education - As Ratio to Aggregate Expenditure.”

States and UTs (Union Territories) are given an FLN Score based on the above pillars and indicators, and then ranked accordingly. AP has an FLN Score of 44.03 and Telangana has an FLN Score of 34.43, where a higher score indicates higher foundational literacy than a lower score. Among all thirty-six states and UTs, Andhra Pradesh has the 25th highest FLN Score, while Telangana has the 35th highest (second lowest) FLN Score. AP and Telangana are both considered geographically “small states,” and rank 9th and 11th respectively among all 11 small states. This indicates a relatively low performance in overall foundational literacy in AP and Telangana. However, several individual pillars and indicators in AP and Telangana are worthy of note.

Andhra Pradesh is considered “overperforming” in the Educational Infrastructure pillar: The percentages of schools with electricity connection, functional drinking water, functional toilets, hand wash facility, and library facility are all above 90, and the percentage of schools with medical checkups is 86.18. However, AP appears to be lagging, or at least just keeping up, in the other four pillars. While the Educational Infrastructure pillar is given a score of 79.19, Access to Education is scored 44.67; Basic Health is scored 46.07; Learning Outcomes is scored 33.21; Governance is scored 17.01. AP is underperforming when it comes to expenditure on government primary schools: the percentage to total expenditure on primary education for government schools in AP is only about 2.98, while some states (such as Himachal Pradesh: 87.66, Haryana: 86.17, Ladakh: 100.00, Lakshadweep: 100.00, Madhya Pradesh: 97.70) are overperforming. However, AP is overperforming in the Transition Rate – Primary indicator, which is 95.50.

On the other hand, Telangana is overperforming in none of the five pillars. Educational Infrastructure (57.35), Access to Education (41.36), and Basic Health (45.03) are all “performing within expected range,” while Learning Outcomes (14.96) and Governance (13.45) are

underperforming. Among all 42 indicators, Telangana is overperforming in only four: Adjusted Net Enrollment Ratio–Primary level for girls (100.00), CWSN (Children With Special Needs) students get facilities from school (74.73), Ratio of contractual teachers relative to regular teachers (95.81), and Transition Rate–Primary (100.00). This indicates Telangana’s strengths in supporting girls, CWSN, and contractual teachers in its education system. However, other important indicators indicate performance within the expected range, or underperformance: The percentage of preschool education is only about 34.84; only about 74.81 percent of schools have functional drinking water; 32.28 percent have functional toilets; 30.94 percent of schools have a functional computer facility.

2.2 Education Reform in Andhra Pradesh and Telangana

However, changes are actively being made to improve these education systems. Based on Telangana’s 2024-25 Budget, a proposed 500 crore rupees (₹5,00,00,00,000) is to be invested into higher education and Telangana Public Schools. In an effort to more effectively integrate technology in education, 65 government ITIs (Industrial Training Institutes) are to be upgraded by the introduction of “the latest technology tools and machines,” along with new courses to bolster training. Overall, a new education budget of 21,389 crore rupees is proposed. This is approximately a 12.0% increase from the 2023-24 education budget of 19,093 crore.

In Andhra Pradesh, YS Jagan Mohan Reddy (Chief Minister of AP from 2019 to 2024) implemented many important educational reforms. From 2018 to 2023, the GER (Gross Enrollment Ratio) grew from 84.48 to 100.00. The following are notable programs implemented to improve quality of education in AP:

- Ammavodi: Provides monetary aid to families with school-going children through cash transfers.
- JVK: Provides students with school kits that include uniforms, textbooks, notebooks, and other essential items.
- Nadu-Nedu: A comprehensive initiative focused on infrastructure development, including the construction of new classrooms, toilets, libraries, and drinking water facilities.
- Jagananna Gorumudda: Addresses both malnutrition and school attendance by implementing mid-day meals.
- CBSE (Central Board of Secondary Education): Creates a board dedicated to the implementation of a standardized and holistic curriculum across all affiliated schools.

Overall, it is evident that both Telangana and Andhra Pradesh are invested in the improvement of the overall quality of education in their respective states.

2.3 National Educational Legislation and Reform

The Indian government saw, and continues to see, education as a major factor in improving overall national economic prosperity. In 2001, the Indian government launched the Sarva Shiksha Abhiyan (SSA) program with the goal of universalizing primary education. Many

interventions were designed to improve elementary education, such as the implementation of “a primary school within one kilometre of every hamlet, village and urban community,” “a classroom for every teacher,” and “free textbooks for all students.” Furthermore, to ensure balanced growth across states in India, greater funding was given to the states that were falling the furthest behind: almost 70% of the program’s funding went to the four states most lagging (Ward). According to the former Minister of State for Education, Upendra Kushwaha, education in India has experienced marked growth during the implementation of SSA. Such improvements include the following:

- Total enrollment in public schools has risen from 18.79 crore children in 2009-2010 to 19.67 crore children six years later.
- The Pupil-Teacher Ratio (pupils per teacher) has fallen from 32 in 2009-2010 to 25 six years later.
- There were about 134.6 lakh children that were out of school in 2005. This dropped to 81 lakh children in 2009, and 60.64 lakh in 2015.
- Enrollment of Scheduled Caste children at the elementary level has grown from 19.06% in 2010-2011 to 19.79% in 2015-2016. (The Scheduled Caste is the official name of the lowest castes in India.)

In the past decade, government expenditures on education as a percentage of GDP have risen from 3.3% to 4.5%, a fact that is especially notable considering India’s GDP grew by a factor of 2.5 in the same time frame.

Based on the 2020 National Educational Policy published by the Ministry of Human Resource Development, the government aims to bolster the higher education system in part by placing at least one large, multidisciplinary university or college in or near every district. Additionally, the National Educational Technology Forum (NETF), was formed in 2020 to expand the use of technology in education. Among the many ways this organization will improve education technology, they have placed a focus on better integrating technology-based education platforms such as DIKSHA and Swayam.

DIKSHA (Digital Infrastructure for Knowledge Sharing) was launched on September 5, 2017. To increase ease of access, this digital infrastructure can be accessed through QR codes provided in textbooks, and the portal will “come up with suggestions and topics” for students to study. Additionally, the portal is accessible in 18 different languages, including English, Hindi, Marathi, Tamil, and Telugu. The broad range of available languages ensures that students across India can benefit from this program. This application is available as a mobile app on the Google Play Store. As of January 24, 2023, DIKSHA has reached over 60 billion minutes in app usage, and seen over 135 million course completions. Since the app’s release, it has reached about 180 million students and seven million teachers.

Education is a powerful tool for poverty alleviation on a national scale. In a 2014 study by Montenegro and Patrinos, the authors show that the cross-economy average private rate of returns for one additional year of schooling is 10%, and that these returns appear to be “higher in

low or middle income economies than in high income economies.” Furthermore, according to the 2013/4 Education for All Global Monitoring Report (EFA-GMR), if every student in low-income countries acquired basic reading skills upon completing school, about 171 million people could be lifted out of poverty—a 12% reduction in global poverty.

In a 2021 study analyzing the relationship between poverty and education, researcher Sumana Mallick discovers a negative relationship between literacy and poverty in Indian states, as seen in the graph below. There appears to be a negative relationship between BPL and Literacy Rate; that is, states with lower literacy rates have higher poverty rates and vice versa, on average. This indicates a potential correlation between education and poverty, suggesting that increased education may yield lower poverty rates.

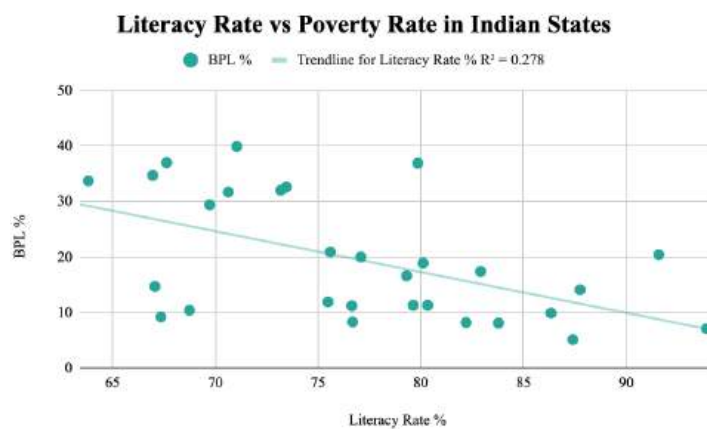


Fig. 1: A scatterplot depicting BPL % (below poverty line rate) as a function of literacy rate %, using 29 Indian states as data points. This data was provided in table format by Mallick and presented graphically by the author of this paper.

It is evident that in the past several years, the Indian government has put considerable effort towards improving their education system. Based on previously conducted research, a stronger focus on education can aid in poverty alleviation.

3. Methods

While observing the broader picture of education and poverty is useful and necessary, a closer look is often just as important. The results of the interviews conducted with a selection of PLNRES beneficiaries provide this closer look into how getting an education can financially uplift impoverished or low-income families.

Interviews were conducted with a sample of beneficiaries who have graduated from college using aid provided by PLNRES and are now earning independently. In total, 23 beneficiaries were interviewed. The students selected for interviews were drawn from the page titled “Beneficiary Students settled in Jobs” in the official PLNRES handbook (Appendix D). There are 29 students listed on this page. However, 30 interviews with beneficiaries were anticipated; one beneficiary, Student 131, is not listed on that page, because this student is currently unemployed, but has secured future employment. Of the 30 anticipated beneficiary interviews, information from 7 beneficiaries could not be collected. Four beneficiaries (005, 006,

012, and 129) were contacted, but did not respond. Three beneficiaries (028, 071, and 137) were unable to be contacted in the first place. The remaining 23 students and the information drawn from their interviews are consolidated in Appendix C.

For each student, data was collected in one of two ways: either through phone call, or through a questionnaire that students completed independently. Appendix A contains a list of questions that every student was asked if they were interviewed over the phone. Appendix B contains a list of questions (and answer choices, for multiple choice questions) asked in the questionnaire. These questions addressed family income before and after their funded education, government aid (where applicable), as well as how much aid was given to the student over the duration of their education. Further information about education, student life, personal life, motivations, and goals was also taken into account to better contextualize their financial and social experiences.

From the data collected directly, two important metrics were derived through simple calculations. The first is the % paid by PLNR. Using information provided in the PLNR booklet, as well as the data provided through interviews, each student's total cost of education and the total amount paid by PLNR was calculated. The total paid by PLNR divided by the total cost of education—converted to a percentage—is the % paid by PLNR.

The second metric derived through simple calculations is referred to as the “multiplier.” The multiplier is the factor by which a student's family income grew from before their education to after their education was complete. (For instance, Student 14 had a multiplier of 3.36, because their family income grew from ₹140,000 to ₹470,000.) However, the family income data collected through questionnaire and phone call differed slightly. To make the questionnaire easier for students to complete, the questions that ask about family income are multiple-choice, where each answer choice is a range of ₹5,000. (An “other” option was also provided, where they could enter a number or range that was not included in the provided answer choices.) However, those that were interviewed over the phone were able to provide singular numerical answers, rather than a range of values, for their family income. To effectively draw conclusions from these data, the income ranges were converted to singular values by calculating the midpoint of each range. For example, a range of ₹5,000–10,000 resulted in a midpoint value of ₹7,500. This modification allowed for usage of the multiplier for all students, even those that entered their information through the questionnaire.

4. Results: A Quantitative Analysis

The following information was collected from the interviews and the PLNRES booklet. Not all of this information is addressed in the Results section. Refer to Appendix C for a more comprehensive view of each of these metrics.

- Family Income Before Aid (yearly)
- Family Income After Aid (yearly)
- Multiplier
- Annual Package (LPA)
- Current Profession
- Course
- Branch/Concentration
- Total Cost of Education

- Total Paid by PLNR

- % Paid by PLNR
- Years Supported by PLNR

The following chart (Fig. 2) shows that a majority of students chose to complete degrees in Technology, Engineering, Science, or Business/Commerce. This is justified by the 2023 Wheebox India Skills Report (ISR), which finds that the “most employable talent [...] was found to be B.Com and MBA graduates. BE/BTech domain candidates are the third most employable, in the domain-wise employability category.”

The low-income students interviewed are focused, above all else, on making money to support themselves and their families. Therefore, it is no surprise that these students are selecting college courses that will give them the highest likelihood of employability in the future.

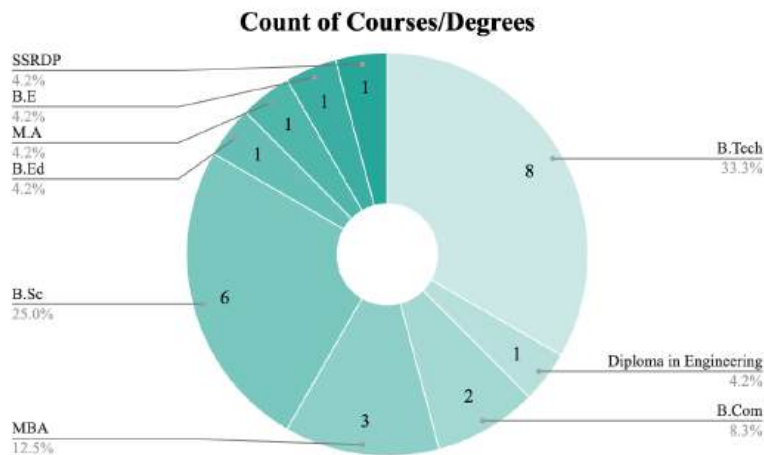


Fig. 2: A pie chart that summarizes the various courses/degrees that the 23 students took throughout their higher education (Column L in Appx. C). Note that Student 036 completed both a B.Sc. degree and a B.Ed. degree, so this student is counted twice: once for each degree.

The multiplier is an important metric that conveys information about the financial growth a student’s family experienced throughout the student’s education. As illustrated in the histogram below (Fig. 3), the majority of the multipliers lie between 1.00 and 4.50. The two uppermost multipliers, 6.67 (Student 036) and 8.40 (Student 013), are statistical outliers. However, these are not errors.

Student 036 began her higher education with an annual family income of ₹72,000. Her father was a carpenter, and she describes their family situation before the accident as comfortable, and moderately within their means. Before she began her higher education, her father suffered an accident, leaving him paralyzed and therefore unable to work. Through the financial support of PLNRES (the society paid about 43% of the cost of her education), Student 036 completed both her B.Sc and B.Ed, and now works as a teacher, making ₹480,000. She now

earns the majority of her family’s combined income, resulting in income growth by a factor of 6.67.

Student 013 began his higher education with an annual family income of ₹100,000. His father had been out of the picture since sixth grade, leaving his mother to care for him and his younger sister. His family’s housing was provided by the temple in which his mother worked; she made below ₹6,000 a month. Just before 10th class, his mother’s wage rose to below ₹10,000. PLNRES paid for over 25% of Student 013’s education. Their combined family income is now ₹840,000—growth by a factor of 8.40.

Students 036 and 013, though statistical outliers, are not excluded from the long term financial benefits that PLNRES yields. Among the 20 students for which a multiplier could be calculated, all but one saw growth in their families’ income (Student 042 experienced little to no change in their family’s annual income). Additionally, the average multiplier is 2.71. This indicates that, on average, students’ participation in PLNRES may have played a role in improving students’ financial situations after graduating from higher education.

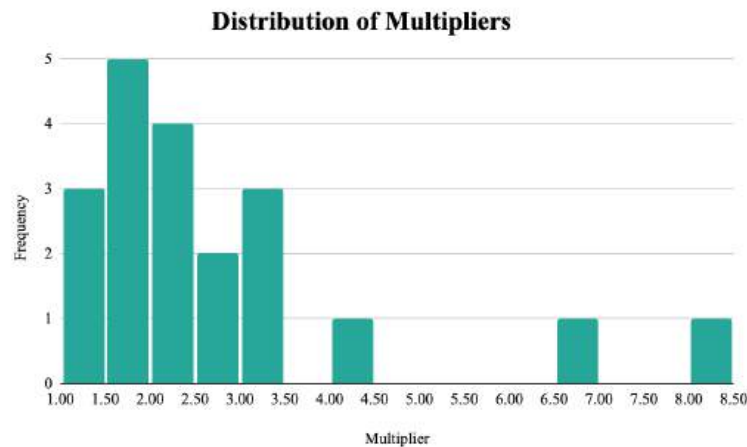


Fig. 3: A histogram displaying twenty students’ multipliers (Column I in Appx. C). Student 004 did not provide their family income after aid, and Students 001 and 150 did not provide their family income before aid, so their multipliers are not included.

The below graph (Fig. 4) depicts the multiplier as a function of annual family income before aid. There is a negative, curved, relatively strong relationship between annual family income before aid and the multiplier. That is to say, families that began with lower income generally experienced a greater factor of growth than families that began with higher incomes. Therefore, it appears that this financial aid is more effective for lower-income families, suggesting that lower-income families benefited more significantly than higher-income families from the aid provided. This could be due to the relative scarcity of resources before aid, thereby making any additional financial aid more impactful.

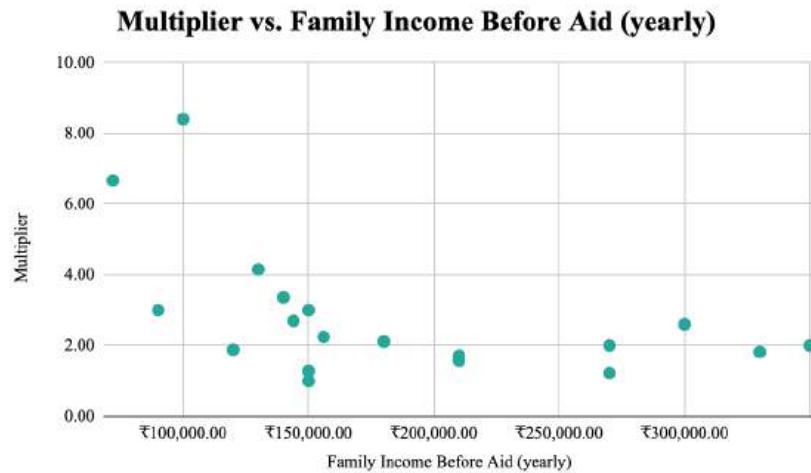


Fig. 4: A scatterplot depicting the multiplier (Column I in Appx. C) as a function of annual family income before aid (Column G in Appx. C). Student 004 did not provide their family income after aid, and Students 001 and 150 did not provide their family income before aid, so they are not included.

5. Discussion

5.1 A Qualitative and Anecdotal Analysis

A common theme seen in almost all students interviewed is the way they perceive the importance of education. As one student said in their interview, “I want to be successful in life, everybody wants that. The only way is education. This...places us in a better position.” Another student echoed this sentiment, saying if you study diligently for “three years or four years of life, [if] you are dedicated to these years, then your life will be super happy.” Yet another student said a very similar thing: “if we study, we will have success in life.” Many of these students see studying and getting an education as integral to their future success. Therefore, the potential for future success is a key motivating factor in working hard despite struggles in their personal and academic lives. This can be observed anecdotally, through the way they talk about their education, but also in the degrees they chose to study. As demonstrated in the Results section, students overwhelmingly selected the most employable college degrees, since their number one goal is ultimately to make enough money to support themselves and their families. They know that, when it comes to their education and career, they often must put aside their personal ambitions for the sake of their families’ prosperity. For example, Student 077 has always had a passion for photography, videography, and VFX. As he said in his interview, he didn’t want to “follow the crowd” and do a course in technology, engineering, or computer science. However, he ultimately decided to do his B.Sc in Maths, Statistics & Computer Science because it would provide him better job opportunities. He is now making 2.20 LPA. He has a small side job in videography and photography editing, but treats it more as a hobby than a job. His multiplier was 2.69, a considerable factor of growth.

Furthermore, upon being asked what motivates them to work hard, some cited their family background, or a desire to gain respect from their relatives and neighbors. One student beamed with pride as she told me that she is the first college graduate in not only her home, but also in her entire family. She is now the main source of income to her family, making about 4.5 LPA now, as compared with less than 1 LPA before getting her education.

Each individual comes from a slightly different background—some grew up with a parent that died, was abusive, or was incapacitated, while others may not have had those experiences—yet their ability to grow from the suffering they began with is remarkable. As mentioned earlier, one student’s father was a carpenter before he became paralyzed due to an accident; her mother, like many mothers in this sample of interviewees, is a housewife. With the father unable to work, the burden of earning was placed on his wife and kids. Moreover, many family relatives cut ties following the accident, leaving her family floundering both financially and emotionally. This student told me that the entrance of PLNRES into her life felt like divine intervention. The fact that PLNRES was willing and able to fund almost half of her education truly felt like a blessing from god.

However, the proportion of educational fees funded by PLNRES’s varied between individuals. Some students received government aid, and some were able to pay for part of their tuition out of their pocket, while PLNRES paid the rest. The average percentage of the cost of students’ education paid by PLNRES was 29.13%, indicating that the organization was willing and able to pay for a sizable portion of their education.

When asked what they would have done had they never received financial aid from the PLNRES, they mention going into debt, taking out loans, or taking time out of their day to work to provide for their own education. By receiving funding from this organization, students are able to focus solely on their education, rather than attempting to earn income simultaneously.

5.2 Scarcity

PLNRES was founded to fund college education for students who otherwise would have dropped out due to financial constraints. The success that the interviewed students have experienced suggests that the full extent of PLNRES’s support isn’t simply the receipt of a check. Rather, its impact is much more long-lasting. With the concern of financing their own education becoming slightly less pressing in their minds, students are able to devote more of their time and mental energy towards their education itself, yielding higher dividends in the future. In *Scarcity: Why Having Too Little Means So Much* by Sendhil Mullainathan and Eldar Shafir, the authors argue that for those who experience scarcity in any valuable resource, whether that’s money, time, or calories (for those on a diet), that scarcity weighs on their brain and taxes what the authors call “bandwidth.” This bandwidth tax impairs cognitive capacity, causing the victim of scarcity to make potentially irrational or even entirely incorrect choices. Mullainathan and Shafir ran an experiment in which they examined sugar-cane farmers, a group of people who are relatively rich immediately following harvest and relatively poor just before harvest. They found that “the same farmer fared worse on fluid intelligence and executive

control when he was poor (preharvest) than when he was rich (postharvest). [...] The very state of having less money in the months before harvest had made [the farmer] perform less intelligently and show less cognitive control” (Mullainathan and Shafir 58-59). When one has scarcity weighing on their mind, their ability to focus on other matters is impaired—when a student is worried about how they are going to pay this semester’s tuition, they may perform worse in school.²⁶ This is the profound impact that PLNRES provides: it removes the financial burden of an education, at least partly, allowing the student to place more effort into their studies, thereby giving them the opportunity to work a high-paying job in the future.

6. Conclusion

Poverty is a multifaceted, complex issue. While there are many ways to address it, one possible avenue is through the education system. In the last few decades, India has proven that it is capable of substantial economic and social development, especially in the context of the education system. While there is still room for growth, the decline in poverty rates and the increased focus on education reform are welcome indicators of positive change in India. PLNRES is unique not because it provides educational financial aid—many organizations do that—but because the relationship between beneficiary and PLNRES extends beyond a monetary transaction. The close emotional relationship that the beneficiaries form with the organization ensure PLNRES’s long-term success. Of the 23 interviewed beneficiaries, 22 have since joined as members and began giving back to the very same organization, to help financially support the educations of future students.

The structure of educational charities like this one is noteworthy because the money they provide is meant specifically to get an education. Rather than paying for basic living necessities such as food, shelter, and water, PLNRES is enabling students to get an education, setting them up for success for the rest of their life by opening up opportunities for higher-paying jobs. Based on the research on education and poverty previously collected, and an analysis of PLNR Educational Society, it is logical to conclude that an increased focus on education—whether on an individual or national level—can aid in poverty alleviation in India.

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Could Gene Editing or New Biotechnology Help Cure Peripheral Neuropathy?

By Vidula Ravindran

Background

The nervous system is divided into two systems: central and peripheral nervous systems. The brain and spinal cord make up the central nervous system where they receive and convey messages to body, whereas the peripheral nervous system (PNS) refers to the parts that are not part of the central nervous system (CNS) which includes the autonomic and somatic nervous system which branches off to the parasympathetic and sympathetic nervous system. The peripheral nervous system carries messages to and from the central nervous system and delivers them to various parts of the body which results in regulation of voluntary movement and involuntary body processes such as breathing, heartbeat, blood flow, and sensory information. The somatic nervous system regulates voluntary movements and reflex movements while also assisting in the process of touch, sound, taste, and smell. The autonomic nervous system controls involuntary actions such as organ function. The somatic and autonomic nervous system work together to coordinate the body's conscious and subconscious processes.

The autonomic nervous system has two anatomically distinct divisions: sympathetic and parasympathetic. When the sympathetic nervous system is activated, it causes a state of overall increased activity and attention known as the "fight or flight" response. During this process, heartbeat and blood pressure increases, with physiological changes like increased adrenaline release, sweating, and rapid breathing. The SNS signals nearly all living tissues in the body. The parasympathetic nervous system promotes the "rest and digest" process, which lowers your heart rate and blood pressure. Your sympathetic and parasympathetic nervous systems play contradictory yet complementary tasks. Your sympathetic nervous system sends messages to systems that activate your body's activities, while your parasympathetic nervous system sends signals that restore those systems to normal activity levels. These two systems inhibit one another so they cannot both be activated at the same time. Over activation of the sympathetic nervous system for an extended period of time can lead to unwanted consequences such as chronic stress which can increase your blood pressure and increase your risk of heart attack or stroke. Signaling the parasympathetic nervous system will allow the body to return back to the normal levels. Because these two systems counteract each other, they help to keep your body in balance.

Although CNS and PNS are both part of the nervous system, their neurons possess different structures and functions. The neurons in the central nervous system contain oligodendrocytes wrapped around their axons, while neurons in the peripheral nervous system have schwann cells surrounding them. In the peripheral nervous system, Schwann cells function similarly to oligodendrocytes. Each Schwann cell can only myelinate one axon, whereas a single oligodendrocyte can myelinate 40-50 axons. Fast signal transmission in the peripheral nervous system (PNS) is essential for adequate sensory and motor tasks. Quick signal transmission provides precise and timely coordination of motions, as well as reactive actions, such as avoiding

a burning surface. Rapid sensory processing enables immediate responses to environmental changes, which is crucial for detecting pain.

The peripheral nervous system has two types of neurons: Motor and Sensory. Sensory neurons collect and transfer information about within and outside the body to the central nervous system for processing. The information transmitted to the brain enables touch, taste, smell, sight, and sound. The Motor neurons receive input from other neurons and send commands to muscles, organs, and glands. There are two types of motor neurons: upper and lower. Upper neurons are found in the brain and link to the lower neurons near the spinal cord and peripheral nervous system. The general structure of a neuron includes dendrites, soma, and an axon. The coating of myelin around axons, similar to the insulation around wires in electrical systems, is often referred to as a "myelin sheath." The myelin coating protects the axons and facilitates the rapid transmission of information and signals throughout the body. When there is a dysfunction in the myelin, demyelination occurs. Demyelination occurs when the myelin sheath surrounding the axons is damaged or unable to develop properly. When there is no myelin coating the axons, signals and information are carried slowly. This can lead to a variety of challenges, including extremely painful demyelinating diseases.

Charcot-Marie-Tooth Disease

Charcot-Marie-Tooth (CMT) disease, also known as hereditary motor and sensory neuropathy, damages nerves in the peripheral nervous system. CMT affects the peripheral nerves in the arms and legs, which control movement and sensation. CMT is characterized by gradual peripheral nerve degeneration, which causes symptoms such as muscle weakness, muscle wasting (atrophy), sensory loss, and foot deformity. CMT affects motor nerves, which regulate muscle movement, as well as sensory nerves, which express sensations like touch, pain, and temperature.

The degeneration of peripheral nerves in CMT is frequently caused by genetic mutations that alter their structure and function. These mutations can interfere with proper signal transmission between the CNS and the muscles and sensory organs, causing the disease's typical symptoms. CMT does not directly impact the central nervous system (CNS), but its impact on peripheral nerves has a profound impact on movement, sensation, and coordination throughout the body.

There are numerous kinds of CMT with similar symptoms, but they differ in terms of inheritance pattern, age of onset, and involvement of the axon or myelin sheath. There are 5 major types of the CMT disease: CMT 1, CMT 2, CMT 3, CMT 4, and CMT X. CMT 1 is the most common and involves abnormalities in the myelin sheath. CMT 2 causes abnormalities in the axon of the neurons rather than the myelin sheath. Lastly, CMT X is the second most prevalent form of CMT. It is an X-linked disorder caused by mutations in a gene that encodes the protein connexin-32. Connexin-32 is important for myelinating Schwann cells. Each neuropathy subtype is caused by a mutation in a distinct gene.

CMT1, a majority of CMT2 cases, and most of the intermediate types are inherited in an autosomal dominant pattern. This inheritance pattern implies that only one copy of the mutated gene in each cell is required to induce the illness. Most affected people have one affected parent. CMT 1 has a subtype which is CMT 1A and is caused by the duplication of a gene on chromosome 17 that encodes the peripheral myelin protein-22 (PMP22). Many of the CMT types are caused by mutations in the PMP22 (Peripheral Myelin Protein). The PMP22 protein is a crucial component of the myelin sheath. Overexpression of this gene leads to aberrant structure and function of the myelin sheath. Many scientists believe that PMP22, rather than other genes that control the duplication, is causing disorders. PMP22 duplications account for the great majority of hereditary CMT cases. Because of its high prevalence, it has been a primary focus of research and clinical diagnosis. Genetic testing for CMT identifies PMP22 duplications.

CMTX is inherited in an X-linked dominant manner which more severely affects males since they have one copy of the X chromosomes. One gene that causes CMTX is GJB1. Mutations in the GJB1 (Connexin-32) gene disrupt signaling and trafficking through gap junctions (Gap junctions facilitate the movement of nutrients, charged particles (ions), and tiny molecules that transmit communication signals between cells), causing an inherited peripheral neuropathy known as X-linked Charcot-Marie-Tooth Disease. Complications include oligodendrocyte and Schwann cell demyelination. This illness causes a variety of symptoms, the most frequent of which are muscle weakness and sensory abnormalities in the outer limbs. Because of the hemizyosity of the X-chromosome in males, the symptoms and difficulties associated with X-linked Charcot-Marie-Tooth illness are more common.

Mitofusin 2 (MFN2) is another essential gene that impacts a particular type of CMT disease. Mitofusin 2 is connected to CMT 2, where abnormalities happen within the neuron's axon instead of within the myelin sheath. It also plays a role in mitochondrial fusion. Since CMT2A is inherited in an autosomal dominant pattern, the disorder can be caused by a mutation in just one copy of the MFN2 gene. There are cases where an individual can get this disease in their adulthood even though the CMT2A inheritance pattern is autosomal dominant. MFN2 is found on the outer membrane of the mitochondria, where it interacts with MFN1, another mitofusin protein, which is present in adjacent mitochondria. MFN1 and MFN2 work in unison to advance the combination of the external membrane of mitochondria, which causes the mitochondria to elongate and mix their contents. Mitofusin 2 (MFN2) is fundamental for mitochondrial fusion, which joins together mitochondria to make a single, interconnected network within the cells. Mitofusin 2 is important because the gene produces a mitochondrial membrane protein that aids in mitochondrial fusion in order to maintain and function the mitochondrial network. This process is vital to preserve distribution, quality control, and function of the mitochondria. The location of MFN2, which is on the outer membrane, is significant because Mutations in the MFN2 gene can cause damaged mitofusin 2 protein in CMT patients. These mutations can occur in a variety of gene areas, such as coding, regulatory, or protein folding. The location and nature of the mutation in the MFN2 gene can have an impact on the severity and specific characteristics of CMT disease. Charcot-Marie-Tooth disease, CMT

disease type 2A, is a disorder that results in axonal degeneration and is caused by abnormalities with mitochondrial activity. MFN2 dysfunction may eventually result within the neuronal loss and peripheral neuropathy observed in CMT2A, as well as alteration to energy synthesis and disturbances to mitochondrial dynamics.

Treatment

No cure exists for all five types of the Charcot-Marie-Tooth illnesses, but therapies are directed towards alleviating symptoms. Patients may engage in physical therapy, occupational therapy, and orthopedic surgery. Pain control is essential for the disorder. Since the illnesses cause physical symptoms and dysfunction, the provided treatments to the illnesses are physical therapies, braces for legs, and more.

My interpretation of potential treatments: there are no cellular treatments for neuron diseases like CMT so we can maybe use CRISPR for gene editing technologies to edit the specific gene or protein to fix the structure of the myelin sheath. Gene editing technology enables scientists to accurately alter DNA sequences within an organism's genome. It is derived from a bacteria's natural defense mechanism that permits it to resist viruses. There are limitations to using CRISPR. CRISPR can mistakenly cut unintended parts of the genome which can cause mutations and lead to different diseases and side effects. There are also many parts of the genes that are not fully analyzed yet and using CRISPR can affect those areas which will lead to a disease that we don't know about or don't have treatments for.

Guillain Barre Syndrome

Background

Guillain Barre Syndrome is an autoimmune disorder targeting myelin sheath and axons. The immune system of the body targets a portion of the peripheral nerve system in Guillain-Barré syndrome. The nerves that convey pain, temperature, and touch sensations can all be impacted in the disease, as well as nerves that control muscular activity. Muscle weakness, numbness in the arms and/or legs, and difficulties breathing or swallowing might arise from this. Guillain-Barré syndrome (GBS) occurs when the immune system mistakenly attacks peripheral nerves, specifically the myelin sheath. This autoimmune response causes inflammation and damage to the nerves and myelin sheath, reducing their capacity to carry messages effectively. The reason for the attack on peripheral nerves is still unknown. GBS is not a hereditary disease; rather, it is a sporadic autoimmune disease caused by infections, environmental factors, and genetic factors, however, the specific reason for the attack on peripheral nerves is still largely unknown. What is known suggests that there are hypotheses on molecular mimicry. Molecular mimicry is one of the primary mechanisms by which infectious or chemical agents can cause autoimmunity. The immune system gets tricked by an infection or other agent into thinking that myelin sheath is a foreign particle that needs to be eliminated.

GBS typically begins with a bacterial or viral infection, with *Campylobacter jejuni* being the most common. Viruses and other bacterial illnesses have been implicated as well. It is thought that the infection-induced immune response goes incorrectly, which results in the attack on the myelin sheath. There are also two types of immune cells that attack the myelin sheath which include T cells and B cells. T cells are white blood cells that play a vital role in the immune system's response to infection. T cells, also known as T lymphocytes and thymocytes, are immune cells that grow from stem cells in the bone marrow. They protect the body by destroying the infection and could also help in cancer treatment. In GBS, some T cells, known as cytotoxic T cells, identify myelin sheath components as foreign or abnormal and generate an attack against them. This immune reaction causes inflammation and damage to the myelin sheath.

Another kind of white blood cell, B cells, may also contribute to GBS by generating antibodies that target myelin sheath components or triggering other immune cells involved in the inflammatory process. B-cells play an important regulatory role in the immune system, creating antibodies, antigen-presenting cells, and directly contributing to inflammatory pathways. The attacks on the myelin sheath leads to paralysis and life threatening progressive muscle weaknesses. The combined consequences of the immune responses cause demyelination, which occurs when the myelin sheath gets damaged or destroyed. Without myelin, nerve conduction fails, resulting in GBS symptoms such as muscular weakness, numbness, and tingling.

Treatment

The two main treatments are intravenous immunoglobulin (IVIG) and plasmapheresis (plasma exchange), which regulate the immune response and remove harmful antibodies. Both work better when started early. Intravenous Immunoglobulin (IVIG) therapy includes injecting immunoglobulins from healthy donors into the patient, which helps to modify the immune system and reduces peripheral nerve attacks. It is most helpful when given within the first two weeks of symptom onset. Plasmapheresis (Plasma Exchange) entails withdrawing the patient's blood, filtering out the antibodies which attack the nerves, and returning the cleansed blood. This decreases the body's immune response and relieves symptoms. IVIG works best when begun early in the disease process. Both treatments seek to speed up recovery and reduce severity. There is also supportive care for the illness. During recovery, physical therapy and rehabilitation can help you restore strength and mobility. A collaborative approach combining neurologists and therapists, as well as regular monitoring, are required for best results. Early treatment enhances GBS patients' chances of recovery and quality of life.

My interpretation of potential treatments

There is a treatment for cancer that can possibly help the Guillain Barre syndrome. The treatment is using nanotubes to slow down cancer. Carbon nanotubes (CNTs) can be used for targeted drug delivery. Currently they are used to deliver chemotherapy drugs directly to tumors, reducing side effects. CNTs can deliver DNA, RNA, or other genetic material into cancer cells to

alter gene expression, either by silencing cancer genes or introducing tumor-suppressor genes. This method can be used in GBS as well. Carbon nanotubes can be altered to carry medicine to T or B immune cells, or even the entire immune system, changing the way it attacks good cells. This technology requires further study because it is not fully developed for cancer; nonetheless, it may be an alternative way to cure GBS using the carbon nanotube. This method can also work for CMT diseases because nanotubes can deliver genetic material to change the gene expression. This gene editing method can alter the CMT mutation and may cure CMT diseases.

Conclusion

The nervous system is divided into two parts: the central nervous system (CNS) and the peripheral nervous system (PNS). The PNS, which includes the autonomic and somatic systems, controls voluntary and involuntary actions. Charcot-Marie-Tooth disease (CMT) is a hereditary condition that affects peripheral nerves, causing muscle weakening, atrophy, and sensory loss. It is classified into two types: CMT 1, which involves abnormality in the myelin sheath, and CMT 2, which affects the axon. Genetic mutations, such as those in the PMP22 and MFN2 genes, are the primary contributors. Guillain-Barré Syndrome (GBS) is an autoimmune condition in which the immune system attacks the peripheral nervous system, causing demyelination and nerve damage. It is generally caused by infections. It causes muscle weakness, numbness, and paralysis. The reaction of the immune system involves T and B lymphocytes that mistakenly target the myelin sheath. There are many methods that can reduce the effects of the diseases but gene editing can be the main method to cure the different diseases, hereditary or autoimmune. There are many gene editing methods that can be proposed and be taken into further research and the methods include CRISPR, Carbon Nanotubes, and CAR T. Although they are used for different illnesses, research can be used to alter the function of the treatments and can be used to cure specific mutations and foreign particles in different diseases. Although the biotechnology treatments are not ready for clinical application yet, these treatments should be further developed by scientists to move towards a cure.

This paper addressed how myelin sheath could be destroyed in disease. Myelin sheath can be destroyed due to hereditary or autoimmune diseases. The same neurons were affected in both diseases and both caused destruction which leads to muscle weaknesses.

CMT

Strikingly, CMT is actually probably multiple diseases and each subtype affects different parts of the neuron.

Understanding the differences in different CMT subtypes lets us think about targeted/specific treatments for each part of the neuron.

GBS

It is interesting that different immune cells (T and B cells) both contribute to disease.

The different immune cells can be altered to treat the GBS disease by using the CAR T cell therapy or nanotubes.

There should be more research on specific treatments and whether they are curative or simply treat symptoms.

Think about the treatments that you'll discuss will be applied to other diseases

Expand the potential treatments to thinking about the neuron and axon

One dream recommendation (if one thing were possible what would you want it to be)

Treatments

Screening

Works Cited

<https://docs.google.com/spreadsheets/d/16WmyjR5yNe917gGuBpED4XbvfHjI3tqpzfxbHWj4v6E/edit?usp=sharing>

(Will cite sources based on the requirement in the journal I am submitting paper to)

The Genetic Basis of Sickle Cell Disease, its Origins, and its Ties to Malaria: A Scoping Review By Aditi Suhas Joshi

Abstract

The research topic is sickle cell disease. Sickle cell disease is a hemoglobin beta mutation that causes regular round blood cells to form a sickled shape and be sticky, blocking the blood flow. This paper seeks to delve into the genetic basis and structure of sickle cell disease, how it has originated, and how it is tied to malaria, as well as available treatments for this disease. This paper finds that sickle cell disease has the most haplotypes/origins discovered in Africa, with one more in Asia. There are many different strands of this recessive monogenetic disease, and it has tied to malaria due to it inadvertently fighting off the bloodborne disease due to sickle cell disease being crescent shape and blocking malaria from passing through the bloodstream. The sources all elaborate on an aspect of what sickle cell is, how it affects the body, its genetic structure or geographic region, its ties to malaria, or treatments. The findings are significant in relation to the literature as a whole because they add crucial information to the paper and this paper is credible due to its sources. The contributions made in this paper are important because it seeks to be more convenient to have groups of information in one place, and researchers can find specific information about the disease. When the findings of this paper are analyzed, it is evident that there are myths about the origins of sickle cell disease and its correlations to malaria. This paper was written so that researchers could learn more about sickle cell disease and have crucial information all in one place.

Background and Objective

Sickle Cell Disease (SCD) is a group of blood disorders resulting from the inheritance of a homozygous and compound mutation in the β -globin gene, which encodes the hemoglobin subunit β (HBB) (Thein, 2013). Hemoglobin (Hb) is an iron containing protein that enables the transport of oxygen in red blood cells. It is a tetrameric protein composed of different combinations of globin subunits; each globin subunit is associated with the cofactor haem, which can carry a molecule of oxygen (Kato et al., 2018). A single nucleotide substitution in the β -globin gene results in the production of a sickle β -globin subunit, which polymerizes and causes the erythrocytes to assume a crescent or sickled shape when the Hb is not bound to oxygen (deoxygenation conditions). Hb tetramers with two of the mutant sickle β -globin subunits, Hb tetramers (HbS) result in more severe polymerization compared to Hb tetramers with a single mutant β -globin subunit (HbAS). Polymerization of erythrocytes reduces their ability to flow through tiny vessels and leads to recurrent vaso-occlusion which is the main feature of SCD. Repeating episodes of vaso-occlusion can be debilitating with numerous negative effects including chronic arthritis resulting from osteonecrosis affecting the joints, progressive retinopathy, chronic renal failure, an elevated risk of stroke, and a shorter lifespan (Damanhour et al., 2014; Manwani & Frenett, 2013; Piel et al., 2017). In addition to vaso-occlusive episodes, other clinical manifestations of the disease include fatigue, anemia, and

increased susceptibility to infections (Platt et al., 2024). These manifestations have significant impact on the physical and psychosocial well-being of individuals with SCD.

The first record of sickle cell disease was by James B. Herrick in 1910, noting red cells that were “sickle shaped” (Herrick, 2001). Sickle Cell disease (SCD) is one of the most common monogenic disorders in the world (Rees & Gladwin, 2010), predominantly affecting people of African, Indian and Arab descent (Weatherall, 2010; Grosse et al., 2011). Global estimates of the disease burden are scarce. Birth prevalence of SCD (children \leq 1-year-old) was highest in several sub-Saharan Africa countries (500–2000/100,000), South America and Caribbean Islands (20-1000/100,000); in the USA and some European countries, birth prevalence was \leq 500/100,000 (Colombatti et al., 2022). Regional data from a systematic review by Colombatti et al. (2022) highlighted sub-Saharan and North-East Africa, India and the Middle East as global SCD “hotspots”. Owing to the similarity of the geographical distribution of malaria and SCD, it has been postulated that the sickle cell trait (SCT) could provide protection against severe forms of malaria (Allison, 1954).

Malaria is a mosquito-borne parasitic disease, carried through the bloodstream (WHO, 2023). Malaria has a higher mortality rate than SCD with about 249 million infections and 600,000 deaths in 2023 (WHO, 2023). Allison (1964) suggested that Hbs conferred a selective advantage against malaria. Friedman (1978) demonstrated that in high oxygen conditions, no sickling of homozygous (SS) and heterozygous (AS) erythrocytes allowed the growth and multiplication of *P. falciparum* identical to growth in normal hemoglobin (AA) erythrocytes. However, cultures under low (1-5%) oxygen showed a clear inhibition of growth. The sickling of SS red cells killed and lysed most or all the intracellular parasites. In AS red cells, the parasites were killed primarily at the large ring stage, probably because of a disruption of the parasite metabolism. This indicated that the mechanism of resistance in vivo could be due solely to intraerythrocytic conditions. In more recent studies, Williams et al. (2005) reported that protection conferred by HbAS is remarkably specific to malaria.

This scoping review will explore the current scientific and cultural understanding of the genetic basis and origin of sickle cell disease and its ties to malaria. Specifically, this review will assess (i) the genetic background of sickle cell disease, (ii) the evolutionary ties between sickle cell disease and malaria, and (iii) the resulting implications of this co-development on the management of the disease. This review will enhance the understanding of SCD and highlight knowledge gaps that can inform future research. Its first objective is to cover the genetic basis of sickle cell disease. This review will delve into the genetic structure and mutations of the condition, and how these affect the body. This paper will scan different viewpoints of the origin and aim to verify an accurate origin for sickle cell disease. Sickle cell disease being created to fight off malaria is a popular belief and then is a product of evolution and internal defense mechanisms gone wrong (Hematol Rep, 2011) On the other hand, there is no evidence to back this claim. The people who had sickle cell disease and could fight off malaria due to having sickle cell disease lived long enough to reproduce and carry on their mutation to the offspring. This cycle would repeat, with people who had malaria but not sickle cell disease passing away

more often. This would cause more people in malaria-borne places, for example, Africa or India, to have sickle cell disease without its origin being linked to malaria.

Results

The literature reviewed highlights the multifaceted impact of SCD, covering its genetic foundation, physiological effects, geographical prevalence, and the connection to malaria resistance. Furthermore, the findings underscore the significance of understanding these diverse aspects to develop effective treatments and management strategies for SCD. The study's contribution lies in consolidating crucial information about SCD, making it easier for researchers to access and comprehend the complex interplay between the disease and its genetic and environmental factors. By addressing myths about SCD's origins and its relationship with malaria, this paper provides a comprehensive and credible resource for future research and clinical applications.

Methods

This study sourced, extracted, and analyzed information on the genetic basis and origins of SCD, its evolutionary ties to malaria and implications on the management of the disease. The methodology involved:

Literature search

Relevant articles were sourced in PubMed, MedLine, Google Scholar, using the search terms 'sickle cell disease', 'sickle cell anemia', 'SCA/SCD evolution' cross listed with key words 'malaria', 'malaria resistance', 'genetic origins', 'treatment'. Additional sources were identified by examining the reference list.

Inclusion and exclusion criteria

The search results were screened based on an inclusion criterion which involved only full-text reports, reviews, web pages and articles written in English focusing on SCD, its genetic origins, evolution, links to malaria and disease management. A total of 19 articles were identified that met the inclusion criteria.

Data extraction

Relevant data including details on the genetic basis of SCD, the evolutionary history of SCD, the disease epidemiology, its evolutionary ties to malaria and mechanism of malaria resistance were extracted from selected articles. Key findings were recorded. Data will be gathered from verified research articles and papers. Two of the major sources that are used in this research paper are PubMed and Nature Journal.

Data analysis

The key concepts were identified from retrieved articles and the extracted data were analyzed to identify key patterns which were structured in line with the specific objectives. Data were categorized into subtopics that linked the key concepts for coherency.,

Discussion

The results confirm that the sickle cell trait provides a protective advantage against malaria, explaining its prevalence in malaria-endemic regions. The genetic mutation responsible for sickle cell disease (HbS) alters the hemoglobin structure, impairing malaria parasite survival within red blood cells. However, individuals with homozygous HbS suffer severe health consequences, including anemia and vaso-occlusive crises. These findings underscore the evolutionary trade-off between malaria resistance and the debilitating effects of sickle cell disease. Future research should focus on therapeutic interventions that mitigate disease severity while maintaining the protective benefits against malaria.

The Origin of Sickle Cell Disease

The SCD mutation is postulated to have arisen between 1100 and 200 BC, in several places in Africa and Asia (101). Four specific haplotypes have been defined in Africa (the Senegal, Benin, Bantu, and Cameroon haplotypes), and one has been defined in Asia (the Arab-Indian haplotype).

This disease was found in a 20-year-old student in Grenada, North America. In 1933, it was concluded that West-Africans were the community that was the most impacted by this disease, and a study of 2,500 people in West Africa proved that West African populations had more cases of sickle cell disease than other regions. Through this experiment and trials, it was determined that the sickle cell trait and disease are separate entities, with sickle cell trait being where a person inherits only one copy of the sickle cell gene and one normal gene. The origin of sickle cell disease was found to be independent of any other diseases, such as malaria, and it became prevalent independently (NHLBI, 2010) A large majority of research papers state that the origin of sickle cell disease was not intertwined to the prevalence of malaria or any malaria outbreaks that happened in the past. Most theories of sickle cell's origin being due to malaria are found in untrustworthy articles.

Genetic Basis and Structure

In this section, I discuss the current knowledge of the genetic basis of SCD, potentially craft diagrams of the genetic structure of SCD, and include information such as what chromosome is affected, symptoms, the mutation, and its effects on the body.

Sickle cell disease is a monogenetic disorder. This means the appearance of the disease is determined by a singular gene. It is also an autosomal recessive disorder, so both parents must carry the trait for the disease to occur in their offspring. (Sanders, 2020). A single base-pair point mutation (GAG to GTG) in the 6th position of the β -chain of hemoglobin referred to as

hemoglobin S (HbS) results in the substitution of glutamic acid (hydrophilic) to Valine (hydrophobic) (Baba P.D Inusa, 2019). There are many different types of sickle cell disease as well as different causes. Hemoglobin S is the most common and the most severe strand of sickle cell disease. 65% of people who have sickle cell disease have the Hemoglobin S strand of it. The child receives a copy of the mutated hemoglobin S from each parent (Onimoe, Rotz, 2020). Hemoglobin SC affects 25% of people who have sickle cell disease and is less severe. The child receives a copy of the mutated hemoglobin S from one parent and the abnormal hemoglobin C from the other parent (Gene, 2024). Other much rarer forms include Hemoglobin SD, SE, and SO.

The single-base-pair mutation causes the phenotype to vary, affecting the hemoglobin and creating the trademark sickle shape. Hemoglobin is a protein found in the red blood cells that carries oxygen from the lungs to other parts of the body. The red blood cells with healthy hemoglobin are round/disc shaped, allowing them to smoothly provide oxygen. When the hemoglobin beta gene, located on chromosome 11 of the hemoglobin, is mutated, it causes abnormal hemoglobin molecules. They are most commonly in the shape of a sickle, but could also be in a long “S” shape (Sanders, 2020).

There are environmental factors that influence sickle cell disease and its evolution. The severity of the disease varies in different geographical locations, with the African population facing more severe effects of the disease than Caucasians. The effects of temperature are also a factor, with colder temperatures resulting in a more painful sickle cell disease. (Tewari, 2015) West Africa holds 63.4% of all sickle cell cases, with the next highest number being in North America with 21%. (Campbell, 2020)

Ties Between Sickle Cell and Malaria

In 1954, it was suggested that the sickle cell trait protected bodies against malaria (NHLBI, 2010). Haldane (1932) was the first to speculate that the genetic makeup of individuals influenced their response to a parasitic infection. When a harmful gene confers some protection to a prevalent disease, its frequency would increase despite the associated risk. Haldane (1939) later proposed the relationship between thalassemia and malaria as an important example. Malaria is a highly lethal parasitic infection, and deaths occur mostly in children. It is estimated to have persisted in several parts of the world for thousands of years where SCD is also dominant. During invasion, the parasite in a vacuole resides within the erythrocyte and during its 48-hour life cycle, 70% of hemoglobin from the red cell cytoplasm is digested to generating amino acids required for protein synthesis (Mohandas & An, 2012). Hence, any red cell abnormalities (as they are for instance in SCD and other thalassemia) likely confers protection against malaria parasite infection and virulence. Haldane’s hypothesis might explain the similarities in the geographical distribution of SCD and malaria, where a continuous exposure to malaria over thousands of years of evolution has selected for several red cell genotypes that offer some protection against severe forms of the disease in the heterozygous state. This includes other

genetic disorders such as thalassemia, G6PD deficiency, blood group polymorphisms and hereditary ovalocytosis.

Malaria is tightly linked to the gametocyte formation in the bloodstream, which means that malaria transfers through the bloodstream. (Meibalan, 2017) Sickle cell disease creates abnormal, sickle-shaped red blood cells, which along other serious effects, block the blood flow. With malaria being a blood-transmitted disease, sickle cell inadvertently fights against it by preventing malaria from travelling through the bloodstream. (Rep, 2011) Several studies have confirmed Allison's (1964) hypothesis that HbS confers a selective advantage against malaria (Williams, 2006). In addition, these studies highlighted that AS heterozygotes are relatively protected from dying of malaria. In essence, AS heterozygotes can be infected with malaria parasites, but they tend to show less parasitemia and they have a lower incidence of cerebral malaria and malaria with severe anemia which can be immediately life threatening (Luzzatto, 2012). Moreover, AS heterozygotes rarely die of malaria, even in the rare cases of cerebral malaria (Olumese, 1997). Although HbS isn't absolutely resistant to malaria, clinical epidemiologic data (Taylor et al., 2012) are consistent with increased fitness of AS heterozygotes in an environment with malaria. This suggests that the mechanism of increased fitness is not a result of failure to invade the cells but rather is based on subsequent interventions. Beet (1946) first suggested that sickling may be responsible; and subsequently Friedmann (1978) demonstrated that sickling of SS red cells killed and lysed most or all the intracellular parasites. Quantitative invitro studies showed significantly higher sickling of AS red cells parasitized in vivo compared to non-parasitized red cells within the same blood sample (Luzzatto, 1970). This indicates that the parasite triggers sickling, and the sickled cells are removed by macrophages (Luzzatto, 1990). This protective hypothesis is known as the sickling-phagocytosis model where there is increased phagocytosis of parasitized sickled cells. However, various other mechanisms of protection have been proposed including premature hemolysis and parasite death (Roth et al., 1978), impaired hemoglobin digestion (Pasvol et al., 1978), impaired cytoadherence (Cholera et al., 2008), acquired host immunity (Williams et al., 2005), translocation of HbS specific parasite growth inhibiting microRNA's (LaMonte et al., 2012), induction of heme-oxygenase-1 (Ferreira et al., 2011) and oxygen-dependent HbS polymerization (Archer et al., 2018).

In contrast to the heterozygote state (AS), the homozygote SS are susceptible to malaria. Thus, selection for the carrier state has occurred at the cost of the loss of the homozygote (SS). Homozygote sickle cell anemia (SCA) patients have a prototype congenital hemolytic anemia and are susceptible to malaria which is an acquired hemolytic anemia. This combination is highly lethal as malaria makes SCA worse and can trigger a SCA pain crisis. In addition, impaired spleen function in patients with SCA limits the filtering and removal of parasitized red cells (Adeloye et al., 1971). A population study in Kenya showed that malaria is no more common in SCA children than in controls; however, mortality rates were 10 times higher in children with SCA than in controls (McAuley et al., 2010).

Post WWII Malaria affected West Africa the most, and many who had malaria died before being able to reproduce. Many people with malaria, before there was treatment, died

before puberty (Gelband, 2020). The average age of death for someone with sickle cell disease before treatment was 43–47 years (Payne, 2023). People with sickle cell disease were able to fight against malaria and live long enough to reproduce. Although sickle cell disease is a recessive gene, the majority of people able to reproduce during the malaria epidemic were people who had the disease, so the chances of their offspring having the disease got higher and higher, which is why West Africa has a higher population of people with sickle cell disease than other populations. (Eridani, 2011) The origin of sickle cell disease is not believed to be because of malaria. Sickle cell disease was discovered later, but became so prevalent because it evolved because of malaria being prevalent in certain areas.

Resulting Implications on the Management of SCD

Management of malaria and SCA remain a major challenge and are both major public health problems. The understood protective effect in AS heterozygotes against malaria doesn't essentially provide information on potential ways to protect susceptible patients. Especially since the mechanism of protection is not clearly understood and pharmacologically mimicking the proposed mechanisms in healthy individuals would be difficult and ethically questionable. It is important to ask what the prevalence of malaria means for the selective propagation of the carrier form of sickle cell trait and prevalence of SCA. On the other hand, malaria substantially contributes to the early mortality of patients with SCA (McAuley et al., 2010). It is thus imperative that these patients be protected by life-long antimalarial prophylaxis.

Treatments for sickle cell disease were not found until the 1980s, as when sickle cell disease originated, it was an unknown and confusing entity. Starting from the 1980s, treatments have ranged from penicillin injections to Doppler screening, with none of the treatments being fully effective. (Ocshner, 2018) In 2014, it was suggested that CRISPR, a gene editing tool, could be used to remove the mutation on the beta-globin gene and prevent sickle cell disease altogether. (Sanders, 2023) On December 8, 2023, the gene editing software was approved by the FDA to be used on patients 12 years and older to remove stem cells, modify them, and place them back in the patients' bloodstream. (Pflaum, 2023) Two types of gene editing were approved for sickle cell disease in 2023-- Casgevy and Lyfgenia. These both modify the stem cells to help treat sickle cell disease, but while Casgevy uses CRISPR Cas-9 to edit the genes, Lyfgenia adds healthy stem cells without using gene editing. (Day, 2023) The outcomes of these treatments have been positive so far. On May 1st, 2024, The first patient has undergone Casgevy treatment for sickle cell disease. While the process will take months, the trajectory is reported to be successful, with clinical trials having a 93% success rate. (Dawson, 2024)

Conclusion

Sickle cell disease originated due to a mutation of hemoglobin S. It requires both parents to be carriers in order to be prevalent in the offspring. While sickle cell disease can play a factor in blocking malaria through its sickle-like shape, the origin of sickle cell disease is not reliant on

the origin of malaria. Gene editing resources have begun to be utilized for the treatment of this disease.

Further research that could not be accessed by this author but could be important for further research could be interviews and surveys with patients of sickle cell disease, as well as ancestry charts.

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Engineering Targeted Immune Modulators for the 2B Variant of Mpox: A Revolutionary Approach in Chemical Engineering and Therapeutics By Kurlus Kurlus

Abstract

The emergence of the 2B variant of Mpox represents a significant global health challenge, characterized by enhanced virulence and immune evasion. This study proposes a revolutionary approach using chemical engineering and molecular chemistry to design and synthesize targeted immune modulators that inhibit the NF- κ B and interferon pathways. By integrating chemical synthesis, process engineering, and biophysical analysis, we aim to develop potent, highly specific therapeutics that disrupt viral replication and modulate the immune response. This interdisciplinary approach offers a dual benefit: immediate therapeutic interventions against Mpox and a broad framework for developing next-generation antivirals. The research is poised to redefine how we approach viral infections, making it an exceptional breakthrough in both chemical engineering and therapeutic development.

1. Introduction

The 2B variant of Mpox, identified in 2024, has challenged existing antiviral strategies due to its ability to evade immune responses and increase disease severity. Traditional treatments have shown limited effectiveness, necessitating new approaches that can address these challenges. Chemical engineering provides a powerful platform for designing molecules that precisely target the molecular mechanisms of viral infections. By focusing on the NF- κ B and interferon pathways, which are critical to the host immune response, this study aims to develop new therapeutic agents that can effectively counteract the 2B variant's virulence.

2. Hypothesis

Hypothesis: Chemical engineering can be harnessed to design, synthesize, and optimize small molecules and biologics that specifically inhibit NF- κ B and interferon pathways, leading to reduced viral replication and improved clinical outcomes in Mpox infections caused by the 2B variant.

3. Methodology

3.1 Molecular Design and Chemical Synthesis

- Rational Design: Computational chemistry and molecular dynamics simulations will be used to design inhibitors that bind specifically to key regulatory proteins in the NF- κ B and interferon pathways. This includes targeting the I κ B kinase (IKK) complex in the NF- κ B pathway and the interferon regulatory factors (IRFs) in the interferon signaling cascade.

- Synthesis Techniques: The designed molecules will be synthesized using advanced organic chemistry techniques such as transition metal-catalyzed coupling reactions, asymmetric

synthesis, and solid-phase peptide synthesis. These techniques allow for precise control over molecular architecture, ensuring the production of high-purity, active compounds.

- Bioconjugation: Antibody-drug conjugates (ADCs) will be engineered to deliver these molecules specifically to infected cells. Chemical linkers will be used to attach the synthesized inhibitors to monoclonal antibodies, ensuring targeted delivery and reduced systemic toxicity.

3.2 Biophysical Characterization and Engineering

- Structural Analysis: NMR spectroscopy, X-ray crystallography, and cryo-electron microscopy (cryo-EM) will be employed to elucidate the structure of the synthesized inhibitors and their complexes with target proteins. Understanding the binding interactions at an atomic level will inform further optimization.

- In Vitro and In Vivo Testing: The efficacy of these compounds will be tested in vitro using MpoX-infected cell lines and in vivo using humanized mouse models. These studies will measure the compounds' ability to inhibit viral replication and modulate immune responses, with a focus on cytokine production and viral load reduction.

- Optimization through Process Engineering: Scalable chemical processes will be developed to manufacture these therapeutics. Continuous flow chemistry and other process intensification techniques will be used to optimize the synthesis and purification steps, ensuring that the production of these molecules can meet clinical demand.

3.3 Nanotechnology-Enhanced Delivery Systems

- Nanoparticle Formulation: Nanoparticles will be engineered to enhance the delivery and bioavailability of the therapeutic agents. These particles will be designed to encapsulate the drugs, protect them from degradation, and ensure controlled release at the site of infection. Surface modification of nanoparticles will be performed to improve targeting to infected tissues.

- Liposome Carriers: Liposomes will be developed as alternative delivery systems, particularly for hydrophobic small molecules. Liposome formulations will be optimized for stability, drug loading efficiency, and targeted delivery, leveraging chemical engineering principles to ensure effective transport and release of the therapeutic agents.

4. Results (Expected)

We expect the successful synthesis and optimization of highly specific NF- κ B and interferon pathway inhibitors that demonstrate significant efficacy in reducing MpoX viral replication and modulating immune responses in both in vitro and in vivo models. The use of nanotechnology-enhanced delivery systems is anticipated to further improve the bioavailability and therapeutic index of these compounds. Process engineering approaches will ensure that these therapeutics can be produced at scale, making them viable for clinical use.

5. Breakthrough and Implications

This research represents a significant breakthrough by merging chemical engineering, molecular chemistry, and virology to address a critical global health challenge. The ability to engineer molecules that can precisely modulate immune pathways in response to viral infections opens up new possibilities for the treatment of not only Mpox but also other viral pathogens. The interdisciplinary nature of this work, combining advanced chemical synthesis, biophysical analysis, and process engineering, positions it as a foundational study for future therapeutic development. The implications extend beyond immediate therapeutic interventions, offering a scalable and adaptable platform for responding to emerging infectious diseases.

6. Discussion

This study highlights the transformative potential of chemical engineering in the development of targeted therapeutics. By focusing on the molecular mechanisms of viral pathogenesis and using advanced engineering techniques to design and deliver therapeutic agents, we have created a model that can be applied to a wide range of viral infections. The integration of nanotechnology and process engineering further enhances the scalability and efficacy of the proposed treatments, making them not only innovative but also practical for large-scale application. This research sets a new standard for how chemical engineering can contribute to the field of infectious diseases, offering solutions that are both scientifically robust and clinically relevant.

7. Conclusion

The successful application of chemical engineering principles to the design and synthesis of targeted immune modulators marks a new era in the treatment of viral infections. This study has demonstrated that by leveraging the power of chemistry and engineering, we can develop highly specific and potent therapeutics that address the unique challenges posed by the 2B variant of Mpox. The interdisciplinary approach taken in this research ensures that the findings are not only innovative but also have the potential for broad application in the fight against infectious diseases.

8. Future Directions

Future research will explore the application of these principles to other viral infections, with a focus on identifying common molecular targets that can be exploited for therapeutic intervention. Long-term studies will also investigate the potential for resistance development and the durability of the immune response modulated by these therapies. Further integration of bioinformatics and machine learning could enhance the precision of molecular design, leading to even more effective and personalized therapeutic options.

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A Review of Current FDA-Approved PCSK9 Inhibitors Against Atherosclerotic Cardiovascular Disease By BaoMeng Mei

Abstract

Atherosclerotic cardiovascular disease is currently the world's leading cause of death, predominantly caused by elevated levels of low-density lipoprotein cholesterol (LDL-C). Current lipid-lowering therapies include first-line treatment with statins, followed by ezetimibe and if both strategies fail, the introduction of pro-protein convertase subtilisin/kexin type 9 (PCSK9) inhibitors. Statins are ineffective for around two-thirds of patients and ezetimibe only lowers LDL-C levels by around 20%. By offering a novel therapeutic target, PCSK9 inhibitors can reduce LDL-C levels by as much as 70%. There are currently three FDA-approved PCSK9 inhibitors: two monoclonal antibodies (alirocumab and evolocumab), and one small interfering RNA drug (inclisiran). Through the analysis of data, alicumab has been found to be the most effective in reducing cardiovascular risk, evolocumab lowers LDL-C and PCSK9 levels the most and inclisiran offers the longest maintenance of these reductions. This review encompasses the PCSK9 protein mechanism and compares the mechanisms of action, clinical efficacies, and safety of currently FDA-approved PCSK9 inhibitors.

Keywords: Atherosclerosis, ASCVD, LDL-C, Statins, PCSK9 inhibitors, Alirocumab, Evolocumab, Inclisiran

1. Introduction

As the main risk factor for cardiovascular disease, atherosclerosis (Jebari-Benslaiman et al.) is currently the world's leading cause of death, responsible for an estimated 20.5 million deaths every year (Piñeiro et al.). Atherosclerotic CVD (ASCVD) refers to diseases of the cardiovascular system due to cholesterol plaque build-up (Esau et al.), which is implicated by the major carrier of blood cholesterol, low-density lipoprotein cholesterol (LDL-C) (Kruth). The accumulation of oxidized LDL-Cs in the subendothelial space of arteries calcifies and develops into lesions (Salekeen et al.). These lesions either reduce blood flow in the arteries or change the mechanical characteristics of the arterial wall, increasing the risk of plaque rupture (Berliner et al.). This often leads to ischemic damage (Salekeen et al.) and thrombosis (Ambrose and Bhullar), respectively. Hence, hypercholesterolemia, a condition that is characterized by high levels of LDL-C, has been established as a significant cardiovascular risk factor (Mortensen et al.). Multiple studies have demonstrated a correlation between lowered LDL-C levels and reduced risks for ASCVD (Catapano et al.), making various cardiovascular therapeutic options available by targeting LDL-C.

Current treatment for hypercholesterolemia involves initiation with statins, preceded by a combination of statins and ezetimibe as second-line treatment. If both therapies fail, then there is the introduction of pro-protein convertase subtilisin/kexin type 9 (PCSK9) inhibitors (Ilut et al.; Mach et al.).

2. Discussion

Statins, currently the most used treatment for lowering LDL-C levels (Rossini et al.), are a class of drugs that are competitive inhibitors of hydroxymethyl-glutaryl-CoA (HMG-CoA) reductase enzyme (Ilut et al.). By inhibiting this cholesterol biosynthetic cascade from HMG-CoA to mevalonate (Buhaescu and Izzedine), statins can reduce hepatic cholesterol synthesis. Despite being the first-line cholesterol-lowering therapy, statins are associated with adverse effects including myalgia (the most common), myopathy, and rhabdomyolysis (the most severe) (Ramkumar et al.). Statins can lower LDL-C levels by as much as 60% (Feingold et al.) but nearly two-thirds of ASCVD patients are not achieving their risk-specific LDL-C goals despite maximally tolerated doses of statin therapy (Ramkumar et al.), which is the result of the patient being either statin resistant or intolerant. Statin resistance has been associated with polymorphisms in several genes; statin intolerance is the inability to tolerate a full dose due to adverse effects (Reiner). In contrast, PCSK9 inhibitors are effective in lowering LDL-C levels in both statin resistant (Pokhrel et al.) and intolerant patients (Voutyritsa et al.).

Ezetimibe has been introduced as an add-on therapy to statins to further lower LDL-C levels in patients already on maximally tolerated statin therapy (Vavlukis and Vavlukis); it inhibits the intestinal absorption of cholesterol in the jejunum by blocking the Niemann-Pick C1-like protein (Rosenson). Where ezetimibe is used in combination with statin therapy, LDL-C levels are lowered by 26-46%, PCSK9 inhibitors combined with statins reduce LDL-C levels by 54-74% compared to placebos (Coppinger et al.; Toth et al.).

PCSK9 inhibitors decrease LDL-C levels by binding to the PCSK9 protein, thereby increasing the availability of LDL-Receptors (LDL-Rs) on the surface of hepatocytes (Handelsman and Lepor). Current US Food and Drug Administration (FDA) approved PCSK9 inhibitors include: alirocumab (Praluent[®]; Sanofi) (Raedler), evolocumab (Repatha[®]; Amgen) (Fala), and inclisiran (LEQVIO[®]; Novartis) (Lamb). PCSK9 inhibitors have shown great effects at lowering major adverse cardiovascular events (MACE) in high-risk patients with LDL levels ≥ 70 mg/dL and in populations that are statin intolerant or statin resistant (Coppinger et al.; Turgeon et al.). Thus, PCSK9 inhibitors, along with their minimal side effects (Kosmas et al.) and a novel therapeutic target, provide a great milestone on the road to treating ASCVD.

3. PCSK9 Inhibitors

ASCVD is associated with familial hypercholesterolemia (FH) (Tada et al.), which is primarily caused by the loss-of-function (LOF) mutations in the LDL-R gene (Bjornsson et al.) or in the apolipoprotein B gene (Innerarity et al.), but in 2003, another protein was found to be associated with FH: PCSK9 (Dubuc et al.; Abifadel et al.).

The human PCSK9 gene, which is located on chromosome 1p32.3, encodes for a 692 amino acid protein (Seidah and Prat) that is mainly expressed in the liver but also the small intestine and kidney (Seidah et al.). PCSK9 overexpression leads to elevated LDL-C levels by accelerating the degradation of LDL-R by binding to the epidermal growth factor-like repeat A (EGF-A) domain (Dubuc et al.; D.-W. Zhang et al.).

3.a. History

In 2003, PCSK9 was first identified in cerebellar neurons undergoing apoptosis and it was originally called neural apoptosis-regulated convertase-1 (NARC-1) (Seidah et al.). As the ninth member of the proprotein convertase family, this protein was also named PCSK9 (Guo et al.). In the same year, through the genetic investigation of two French patients with FH, gain of function mutations in PCSK9 were related to abnormally high plasma LDL-C levels (Abifadel and Boileau). Conversely, LOF mutations in the PCSK9 gene resulted in life-long low levels of LDL-C, thereby reducing risks for ASCVD (Page and Watts).

In 2007, scientists from Amgen disclosed for the first time that they had successfully retrieved the crystal structure of PCSK9 and demonstrated that PCSK9 could function through close binding with LDL-R (Piper et al.). Subsequently, PCSK9 inhibitors were quickly developed to lower the circulating LDL-C levels. Currently, there are three PCSK9 inhibitors approved by the FDA for the treatment of hypercholesterolemia (Pokhrel et al.), but there are also other therapeutic approaches targeting PCSK9 in preclinical and clinical stages (Katzmann et al.).

3.b. PCSK9 Mechanism of Action

LDL-Rs, transmembrane proteins on the surface of liver hepatocytes, are the primary mechanism for regulating LDL-C levels (Fig. 1). Once a circulating LDL-C particle has been bound to the LDL-R, the resultant complex is endocytosed into the hepatocyte through a clathrin-coated pit. Inside the endosome, the LDL-C particle separates from the LDL-R; the LDL-R recycles back to the hepatocyte cell surface while the LDL-C particle is transported to a lysosome for degradation, thereby lowering plasma LDL-C levels (Roth and Davidson).

PCSK9 is firstly secreted into the circulation, which then binds to the EGF-A on the LDL-R (D. W. Zhang et al.). After the binding of LDL-R to PCSK9, an LDL-R-PCSK9 complex is formed by the high affinity caused by the low pH within endosomes (Tveten et al.). The complex is shuttled to the lysosome for the degradation of both the LDL-R and the LDL-C (Poirier and Mayer). Less LDL-R recycling to the hepatocyte surface therefore results in fewer LDL-Rs and higher LDL-C levels. Hence, PCSK9 is a proteolytic enzyme that regulates LDL-C levels by regulating the number of LDL-Rs (Roth and Davidson).

Thus, PCSK9 inhibitors decrease the concentration of circulating PCSK9, so less PCSK9 binds to LDL-R, resulting in less LDL-R degradation. Fewer LDL-Rs on the hepatocyte cell surface allow for more LDL-C degradation, resulting in lowered plasma LDL-C levels.

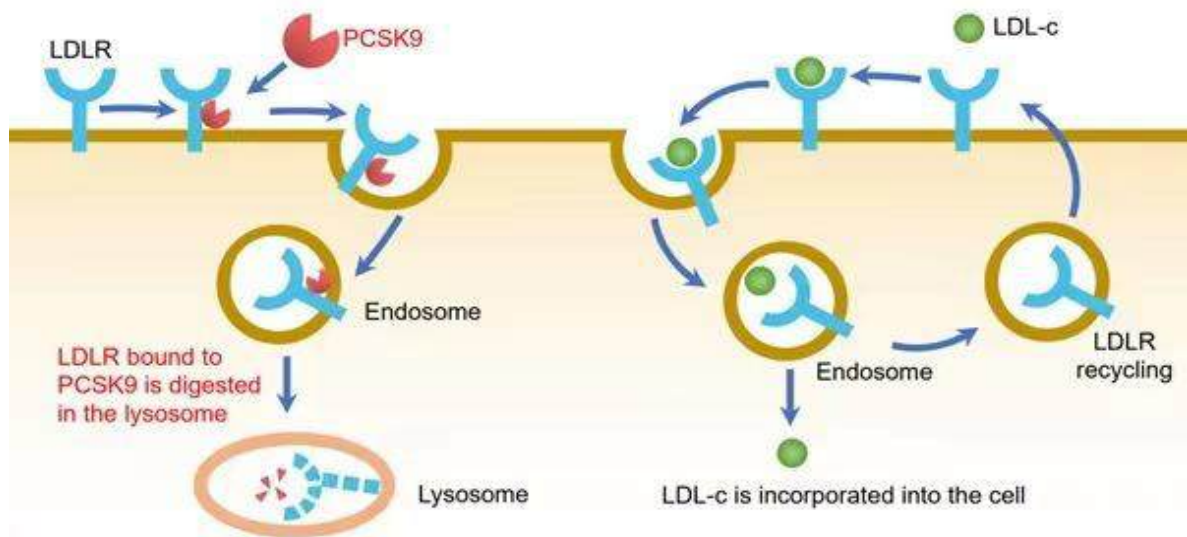


Fig. 1 Biology of LDL-R and PCSK9 in liver hepatocytes.

Right: LDL-R mechanism of action. LDL-C particles bind to the hepatic transmembrane protein, LDL-R, forming a complex that is endocytosed. Within the endosome, the LDL-C and LDL-R separate; the LDL-C is transported to a lysosome for degradation and the LDL-R is recycled to the cell surface for more LDL-C degradation. Left: PCSK9 mechanism of action. Circulating PCSK9 in the bloodstream binds to the EGF-A on LDL-R, promoting PCSK9-mediated LDL-R degradation. The PCSK9-LDL-R complex is endocytosed and shuttled to a lysosome for the degradation of both PCSK9 and LDL-R.

4. Brief Overview of FDA-approved PCSK9 Inhibitors

Alirocumab and evolocumab were the first two PCSK9 inhibitors approved by the FDA in 2015, followed by the FDA's approval of inclisiran in 2020 (Pokhrel et al.).

4.a. Alirocumab

Alirocumab (Praluent[®]; Sanofi) is the first PCSK9 inhibitor to be approved by the US FDA on 24th July 2015 (Raedler). It is used for the treatment of adults with heterozygous FH (HeFH) or ASCVD patients who require additional lowering of LDL-C as an adjunct to maximally tolerated statin therapy (FDA, Praluent (Alirocumab) Injection [Prescribing Information]).

Alirocumab is a monoclonal antibody (MAB) that is administered subcutaneously once every 2 weeks. The initial dose is 75 mg every 2 weeks but if the response in LDL-C levels is insufficient, the dosage can be increased to 150 mg every 2 weeks, which is the maximum recommended dose (FDA, Praluent (Alirocumab) Injection [Prescribing Information]).

4.b. Evolocumab

Approved by the US FDA on 27th August 2015, evolocumab (Repatha^o; Amgen) is the second PCSK9 inhibitor to be approved for heterozygous or homozygous FH or those who require additional LDL-C lowering (FDA, Repatha (Evolocumab) Injection [Prescribing Information]).

As a MAB, evolocumab is administered by subcutaneous injection to the abdomen, thigh or upper arm that is administered every 2-4 weeks. The recommended dose for patients with ASCVD or HeFH is 140 mg every 2 weeks or 420 mg monthly for homozygous FH patients (FDA, Repatha (Evolocumab) Injection [Prescribing Information]).

4.c. Inclisiran

Inclisiran (LEQVIO^o; Novartis) is a small interfering RNA (siRNA) conjugated to triantennary N-acetylgalactosamine carbohydrates (GalNAc) (German and Shapiro) that was approved by the US FDA on 22nd December 2021. Inclisiran is also used for the treatment of adults with hypercholesterolemia, statin resistance or intolerance (FDA, Levqio (Inclisiran) Injection [Prescribing Information]).

As a twice-yearly subcutaneous injection, inclisiran is given as a single injection on day 1, day 90, and afterwards, every 6 months (FDA, Levqio (Inclisiran) Injection [Prescribing Information]). The recommended dose is 284 mg, with each pre-filled syringe supplied as 284 mg/1.5mL (Pokhrel et al.).

Table. 1 Overview of FDA-approved PCSK9 inhibitors.

Drug name:	Alirocumab	Evolocumab	Inclisiran
Brand name:	Praluent ^o	Repatha ^o	LEQVIO ^o
Company:	Sanofi	Amgen	Novartis
FDA approved date:	24 th July 2015	27 th August 2015	22 nd December 2021
Drug type:	Monoclonal antibody	Monoclonal antibody	Small interfering RNA
Method of administration:	Subcutaneous injection	Subcutaneous injection	Subcutaneous injection
Administrative duration:	Every 2 weeks	Every 2-4 weeks	3 months after initial dose then every 6 months thereafter
Recommended dosage per administration:	75 mg	140 mg every 2 weeks/ 420 mg monthly	284 mg

5. Mechanism of Drug Action

5.a. Monoclonal Antibodies

While alirocumab is a MAB IgG1 isotype (FDA, Praluent (Alirocumab) Injection [Prescribing Information]), evolocumab is an IgG2 isotype (FDA, Repatha (Evolocumab) Injection [Prescribing Information]); both MABs bind to the circulating PCSK9's binding site for LDL-R, which prevents the binding of PCSK9 to LDL-R to form the PCSK9-LDL-R complex. This mechanism prevents PCSK9-mediated LDL-R degradation, allowing LDL-R recycling back to the hepatocyte surface for more LDL-C clearance from blood, lowering plasma LDL-C levels (Fala).

5.b. Small Interfering RNAs

Inclisiran is a double-stranded siRNA consisting of a 21-base passenger strand and a 23-base guide strand (Scicchitano et al.), which is specific to PCSK9 mRNA. The passenger strand is conjugated to GalNAc (Khvorova), enabling selective uptake by the liver through the interaction with asialoglycoprotein receptors (ASGPRs) that are highly expressed by hepatocytes (Lamb). Thus, this siRNA design facilitates liver-specific siRNA delivery and uptake, lowers the risk of off-target effects, and allows for lower cumulative doses (Wilkinson et al.).

After inclisiran's subcutaneous administration into the body, it is transported across the interstitial space into the bloodstream and then to the liver (Migliorati et al.) (Fig. 2). Upon reaching the liver, GalNAc binds to the ASGPRs expressed by hepatocytes to induce endocytosis of the ASGPR-inclisiran complex (M. M. Zhang et al.). Once endocytosed, GalNAc is degraded, resulting in the release of the double-stranded inclisiran into the cytoplasm and ASGPR being recycled back to the hepatocyte surface. The guide strand of the double-stranded siRNA is then loaded onto the RNA-induced silencing complex (RISC), leading to the removal of the passenger strand (Wilkinson et al.).

Since the guide strand is complementary to PCSK9 mRNA, these bind together, activating the argonaute enzymes of RISC to catalyze the cleavage of PCSK9 mRNA (Henney et al.). By degrading the PCSK9 mRNA, inclisiran effectively silences the PCSK9 gene by preventing protein translation. Reduced PCSK9 protein synthesis results in less PCSK9 binding to the LDL-R, so less LDL-R is degraded. More LDL-Rs present on the cell surface are available to bind to more LDL-C particles for endocytosis, thereby lowering LDL-C levels.

The long-lasting efficacy of inclisiran is due to the intact inclisiran-RISC complex after PCSK9 mRNA cleavage and the complex's long half-life (Henney et al.; Fitzgerald et al.).

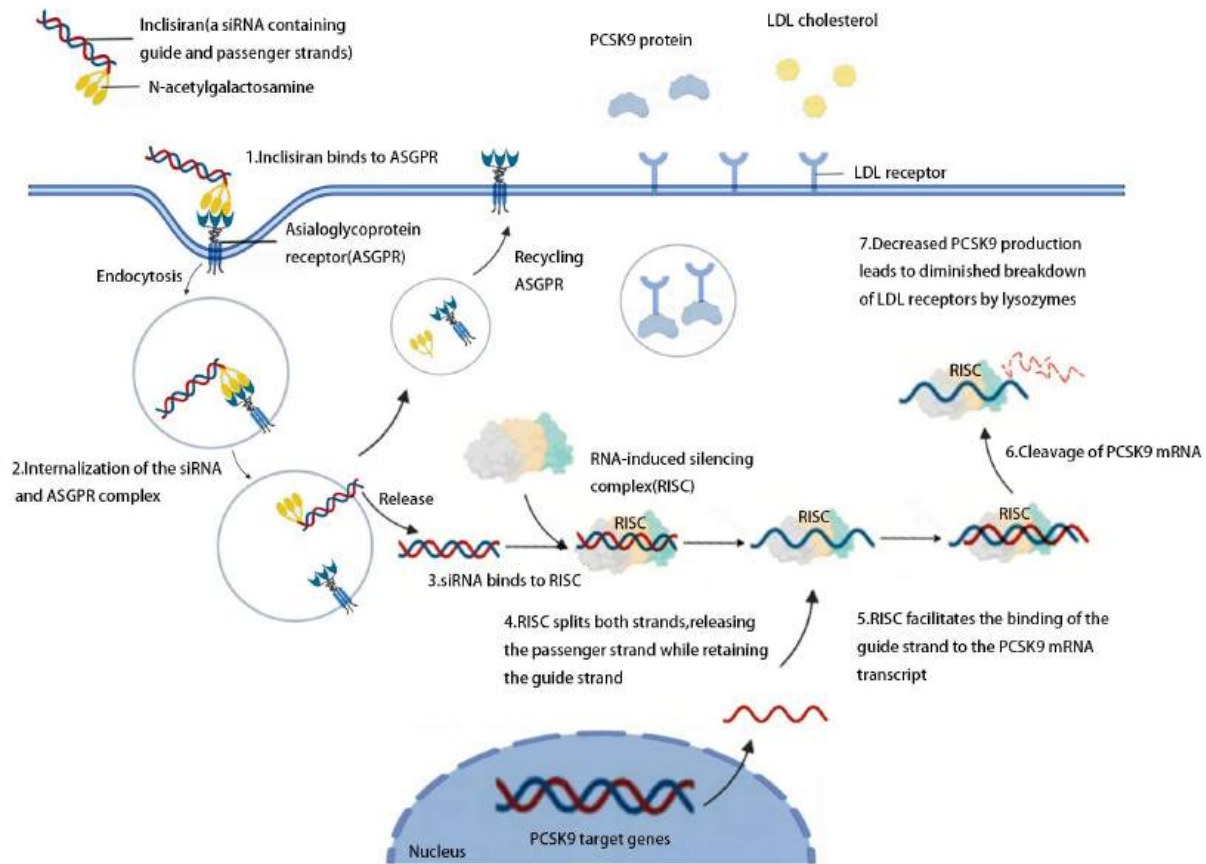


Fig. 2 Inclisiran's mechanism of action.

The GalNAc conjugate of inclisiran interacts with the ASGPRs on the surface of liver hepatocytes, leading to endocytosis of the siRNA-ASGPR complex. Once endocytosed, GalNAc is degraded, ASGPR recycles back to the cell surface and double-stranded siRNA is released into the cytoplasm. Inclisiran binds to RISC, which retains the guide strand and releases the passenger strand. RISC first facilitates the binding of the guide strand to the PCSK9 mRNA, then catalyzes PCSK9 mRNA cleavage, resulting in less PCSK9 mRNA and PCSK9 protein synthesis. Decreased PCSK9 levels result in more LDL-C clearance from blood by LDL-Rs, thereby lowering plasma LDL-C levels.

6. Clinical Efficacy

PCSK9 inhibitors are recommended treatments for lipid-lowering and reducing the risk of ASCVD composites (Wilson et al.; Lloyd-Jones et al.). Hence, PCSK9 inhibitors reduce PCSK9 levels, LDL-C levels, and cardiovascular risks.

6.1.a. PCSK9 levels – Alirocumab

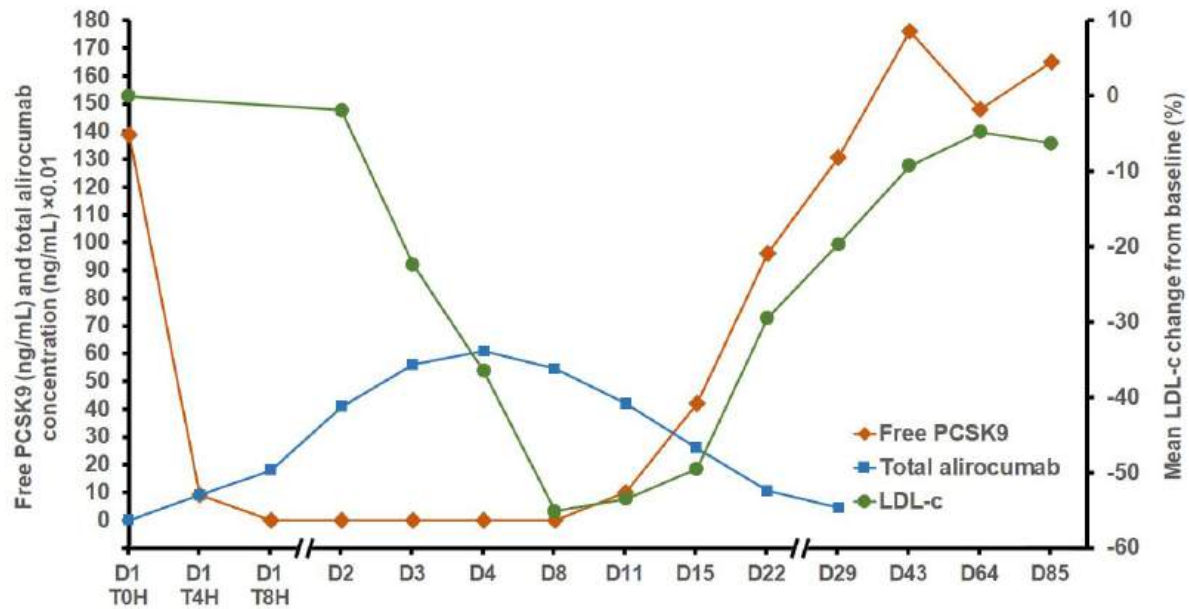


Fig. 3 Relationship between free PCSK9 concentration, mean LDL-C change from baseline, alirocumab after a single subcutaneous administration of 75 mg alirocumab. Adapted from (Li et al.).

After the first day of alirocumab administration, PCSK9 levels for 75 mg, 150 mg, and 300 mg alirocumab dosages can be reduced by 92.85%, 86.66%, and 100% respectively, which can be maintained for 8, 15, and 22 days (Kosmas et al.) (Fig. 3).

In another study, for patients receiving 75 mg Q2W dosage, PCSK9 levels continually decreased, approximately from 260 ng/mL at Week 0 to 125 ng/mL at Week 12, which was maintained until Week 20. Among patients who received the 300 mg Q4W dosage, a similar trend was seen where free PCSK9 concentrations decreased by approximately 200 ng/mL between weeks 0 and 12 (Roth et al.).

6.1.b. PCSK9 levels – Evolocumab

4 hours after the initial administration of 140 mg, and 420 mg of evolocumab, PCSK9 levels were lowered by 92.2% and 92.9% respectively. The lowered PCSK9 levels for the 140 mg dosage were maintained for 15 days and the PCSK9 level for 420 mg dosage was maintained for 22 days (Liu et al.).

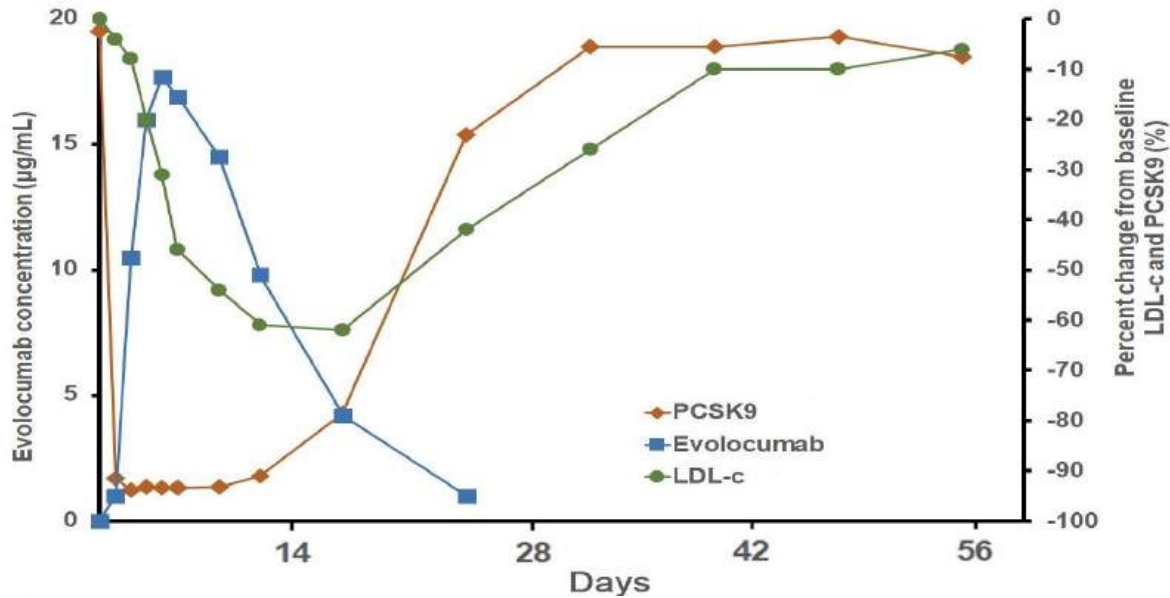


Fig. 4 Relationship between the change of LDL-C, PCSK9 from baseline and evolocumab concentration after a single administration of 140 mg evolocumab. Adapted from (Kasichayanula et al.).

Single dose evolocumab of 140 mg resulted in -95% of LDL-C levels for around 12 days and started to increase back to its baseline level, which was roughly reached on Day 28 (Fig. 4). The increase of LDL-C levels back to the baseline after Day 14 can be explained by the administrative period of 2 weeks by 140 mg of evolocumab (Kasichayanula et al.).

6.1.c. PCSK9 levels – Inclisiran

For a 300 mg dosage of inclisiran, PCSK9 reached its lowest level of -74.5% from the baseline on Day 25, which was maintained for the entirety of the 180-day study (Fitzgerald et al.).

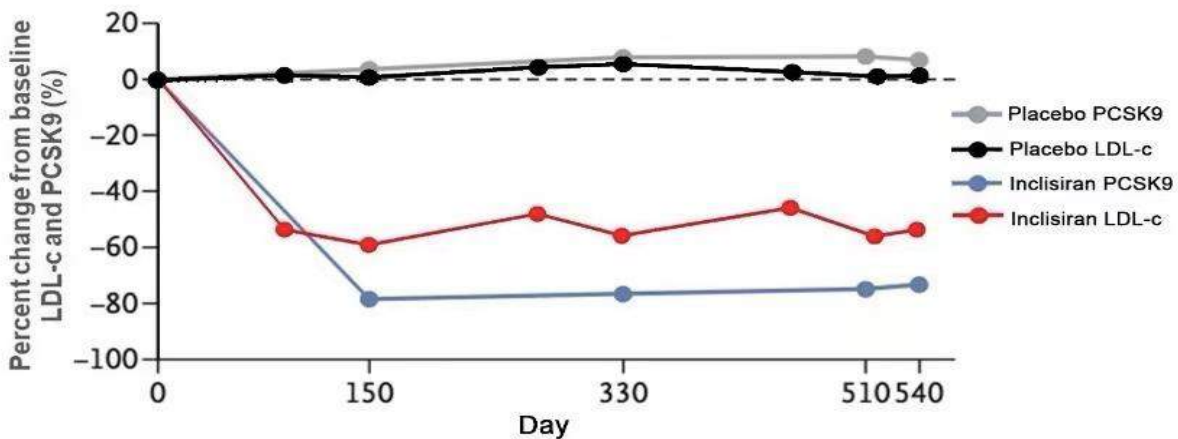


Fig. 5 Efficacy of inclisiran and placebo in lowering PCSK9 and LDL-C levels. Adapted from (Ray et al.).

PCSK9 levels had decreased by 80% from the baseline level on day 150, which was maintained until the end of the study (Ray et al.) (Fig. 5).

6.1.d. PCSK9 levels - Summary

Where MABs lower PCSK9 expression extracellularly by preventing the binding between PCSK9 and LDL-R, inclisiran, a siRNA, inhibits both the intracellular and extracellular pathways (Xia et al.). However, evolocumab, alirocumab, and inclisiran all reduce PCSK9 levels by a similar degree, with evolocumab being slightly more sufficient. With the FDA-recommended doses, alirocumab can reduce PCSK9 levels by at most 85%, evolocumab by 95% and inclisiran by 80%.

In comparison to siRNA, MABs are better and quicker at lowering the PCSK9 levels. Within 1 day after the initial dosage, MABs can reduce PCSK9 expression by over 90% but inclisiran only reaches its lowest PCSK9 level on Day 25, which is only by 74.5% in comparison. Conversely, inclisiran has longer intervals between dosage arms due to its ability to maintain lowered PCSK9 levels for 180 days, whereas MABs can only maintain their levels for 22 days.

6.2.a. LDL-C levels – Alirocumab

Patients receiving Q2W 75mg dosage had a mean decrease in LDL-C levels of 55% by the 4th week (Fig. 3). This lowered LDL-C level was maintained until week 20 (Roth et al.).

After the initial administration of 300 mg, in the 4th Week, LDL-C levels were reduced by roughly 60% and continued to decrease to -75% by Week 6. These lowered LDL-C levels were then maintained throughout the remaining 18 study weeks (Roth et al.).

6.2.b. LDL-C levels – Evolocumab

By the 4th week, treatment with evolocumab reduced LDL-C levels by 60 mg/dL and by the 12th week, LDL-C levels reached their lowest point of 28 mg/dL, which was roughly maintained for the remainder of the study (Sabatine et al.) (Fig. 4).

6.2.c. LDL-C levels – Inclisiran

Patients receiving inclisiran had a decrease in PCSK9 levels of around 55% and reached their lowest on day 60, approximately -60% from baseline levels on day 150 (Fig. 5). Despite slight fluctuations in LDL-C levels, LDL-C levels were maintained relatively low throughout the 540-day study (Ray et al.).

6.2.d LDL-C levels – Summary

Like PCSK9 levels, all three PCSK9 inhibitors demonstrated similar LDL-C reductions, with evolocumab lowering it by the greatest magnitude. The greatest percentage changes in LDL-C levels in comparison to the baseline for alirocumab, evolocumab, and inclisiran were

68%, 70%, and 60% respectively. However, the maintenance time of alirocumab and evolocumab is inferior to that of inclisiran, 22 days vs 180 days.

6.3.a. Cardiovascular Risk Reduction

Apart from lowering PCSK9 and LDL-C levels, PCSK9 inhibitors have also been demonstrated to lower the risk of ASCVD events (Imran et al.). ASCVD is associated with elevated risks of MACE, defined by the occurrence of myocardial infarction (MI), stroke, and cardiovascular (CV) mortality (Berger et al.).

The data below was taken from the meta-analysis by Imran, Tasnim F et al. (Imran et al.), which included 22 studies (n=11,751) on treatment with alirocumab, 21 studies (n=41,361) on the treatment with evolocumab and 4 studies (n=9,522) on treatment with inclisiran (Imran et al.).

6.3.b. Cardiovascular Risk Reduction – Data Analysis

Alirocumab, a 75 mg dosage, significantly reduced the risk of MI, odds ratio (OR) 0.57 (95% CI: 0.38, 0.86), CV mortality OR 0.35 (95% CI: 0.16, 0.77) overall MACE OR 0.35 (95% CI: 0.16, 0.77). However, the risk of stroke was not significantly reduced OR 0.99 (95% CI: 0.48, 2.04).

The administration of 140 mg evolocumab significantly reduced the risk of MI OR 0.72 (0.64,0.81), stroke OR 0.79 (0.66, 0.94), and MACE events OR 0.85 (0.80, 0.89). The OR for CV mortality, 1.04 (0.87, 1.23) is clinically insignificant.

Treatment with inclisiran, 284 mg and 300 mg, showed no significant changes in ASCVD events. MI and stroke had OR 0.72 (0.29, 1.76), and OR 0.69 (0.11, 4.21) respectively. CV mortality had a non-significant OR of 1.06 (0.52, 2.15) and the overall MACE OR -0.21 (-1.11, 0.69).

Table. 2 Odds Ratio of ASCVD events (95% Confidence Intervals) after PCSK9 inhibitor administration

Drug name:	Myocardial infarction	Stroke	Cardiovascular mortality	Overall MACE
Alirocumab (75 mg)	0.57 (0.38, 0.86)	0.99 (0.48, 2.04)	0.35 (0.16, 0.77)	0.35 (0.16, 0.77)
Evolocumab (140 mg)	0.72 (0.64, 0.81)	0.79 (0.66, 0.94)	1.04 (0.87, 1.23)	0.85 (0.80, 0.89)
Inclisiran (284 mg and 300 mg)	0.72 (0.29, 1.76)	0.69 (0.11, 4.21)	1.06 (0.52, 2.15)	-0.21 (-1.11, 0.69)

6.3.c. Cardiovascular Risk Reduction – Summary

Both alirocumab and evolocumab displayed significant reductions in overall MACE, with alirocumab having a lower OR than evolocumab. A further study revealed that alirocumab

reduced all-cause mortality (RR 0.83), whereas evolocumab (RR 1.04) and inclisiran did not show significant associations (RR 1.00) (Wang et al.). The clinical data for inclisiran was shown not to significantly lower cardiovascular risks. However, there was a minimal sample size, number of MACEs observed in the analysis and a lack of long-term data.

Thus, this demonstrates that where both MABs lower the risk of ASCVD events, alirocumab can further lower the risks. Inclisiran showed no significant data in lowering cardiovascular risks, but it requires larger sample sizes and more long-term data before any conclusions can be drawn.

7. Safety and Toxicity

In addition to prominent PCSK9, LDL-C, and cardiovascular risk lowering, PCSK9 inhibitors are well-tolerated. The most common adverse events are often mild, uncommon and resolve over time (Gürgöze et al.).

Table. 3 The four greatest occurring adverse reactions in alirocumab-treated patients (FDA, Praluent (Alirocumab) Injection [Prescribing Information])

	Placebo	Alirocumab
Nasopharyngitis	11.1%	11.3%
Injection site reactions	5.1%	7.2%
Influenza	4.6%	5.7%
Urinary tract infection	4.6%	4.8%

Table. 4 The four greatest occurring adverse reactions in evolocumab-treated patients (FDA, Repatha (Evolocumab) Injection [Prescribing Information])

	Placebo	Evolocumab
Nasopharyngitis	9.6%	10.5%
Upper respiratory tract infection	6.3%	9.3%
Influenza	6.3%	7.5%
Back pain	5.6%	6.2%

Table. 5 The four greatest occurring adverse reactions in inclisiran-treated patients (FDA, Levqio (Inclisiran) Injection [Prescribing Information])

	Placebo	Inclisiran
Injection site reactions	1.8%	8.2%
Arthralgia	4.0%	5.0%
Urinary tract infection	3.6%	4.4%
Diarrhea	3.5%	3.9%

The risk of death was not significantly different between PCSK9 inhibitors versus placebo groups in clinical outcomes studies OR 0.99 (0.84, 1.17). Moreover, PCSK9 inhibitors

were found to have no difference compared to placebo in affecting all-cause serious adverse events (SAEs), OR 0.98 (95% CI: 0.91, 1.05).

The most common adverse events (AEs) associated with statin use are musculoskeletal pain (McKenney et al.), and myalgia, which are frequent causes of statin discontinuation (Selva-O’Callaghan et al.). As a result, studies have shown that only 50% of statin-treated patients continue their therapy after two years, resulting in notable residual risks of ASCVD (Schonck et al.). However, PCSK9 inhibitors have shown no adverse effects on muscle enzymes (Schonck et al.; Guedeney et al.)

Table. 6 Percentage occurrence of myalgia and musculoskeletal pain in patients treated with alirocumab (FDA, Praluent (Alirocumab) Injection [Prescribing Information])

	Placebo	Alirocumab
Myalgia	3.4%	4.2%
Musculoskeletal pain	1.6%	2.1%

Table. 7 Percentage occurrence of myalgia and musculoskeletal pain in patients treated with evolocumab (FDA, Repatha (Evolocumab) Injection [Prescribing Information])

	Placebo	Evolocumab
Myalgia	3.0%	4.0%
Musculoskeletal pain	3.0%	3.3%

For inclisiran-treated patients, the occurrences of myalgia and musculoskeletal pain were below 3% of patients (FDA, Levqio (Inclisiran) Injection [Prescribing Information]). This analysis contrasts the 9.4% vs 4.6% (statin vs. placebo) prevalence of myalgia in atorvastatin-treated patients (Tournadre) and musculoskeletal pain in 22% vs 16.7% in statin users (Buettner et al.).

Hence, through similar AE occurrences to control groups in randomized clinical trials, PCSK9 inhibitors provide a safe and well-tolerated therapy in treating patients with high LDL-C levels.

Conclusion

Where statins and ezetimibe lower LDL-C levels by targeting the HMG-CoA reductase enzyme and the jejunum respectively, PCSK9 inhibitors offer a novel therapeutic target by preventing the binding of PCSK9 protein to the LDL-R. This facilitates LDL-R recycling for more LDL-C degradation, lowering plasma LDL-C levels. PCSK9 inhibitors can decrease LDL-C, PCSK9 levels and cardiovascular risks without substantial adverse effects, highlighting their potential as effective treatment options for patients who are unable to reach their risk-specific goals despite maximally tolerated doses of traditional therapy.

Alirocumab was the first PCSK9 inhibitor to be approved by the US FDA in 2015 as a subcutaneously administered injection every 2 weeks. As a MAB, alirocumab works as an anti-PCSK9 antibody to prevent PCSK9-mediated LDL-R degradation. Out of currently

FDA-approved PCSK9 therapies, alirocumab is the most proficient in lowering cardiovascular risk.

The second FDA-approved PCSK9 inhibitor, evolocumab, is a MAB that is subcutaneously administered every 2-4 weeks. Evolocumab is currently the most effective PCSK9 inhibitor in lowering LDL-C and PCSK9 levels by the greatest magnitude.

Inclisiran is a siRNA drug which inhibits PCSK9 synthesis on translational levels by degrading the PCSK9 mRNA. As the first siRNA in cardiovascular medicine, inclisiran provides a much longer duration of drug action compared to pre-existing lipid-lowering therapies. Unlike other PCSK9 inhibitors, inclisiran demonstrated a non-significant association with lowering cardiovascular risks while lowering LDL-C, despite the direct correlation between LDL-C and ASCVD risk lowering; this could be either due to small sample sizes, lack of long-term data or statistical errors, the result of inclisiran's rather recent approval in 2021.

In conclusion, PCSK9 inhibitors are highly effective and well-tolerated in clinical practice by providing an alternative therapeutic target, PCSK9. By using PCSK9 inhibitors, more patients will be able to reach their lipid targets, which will lower their residual risk of cardiovascular events and help reduce the global burden of the leading cause of death, ASCVD.

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A Review on Methods Used to Strengthen Bioplastics and Applications By Eric Jia

Abstract

With the rise in plastic consumption, concerns about its environmental impact have been at the forefront of conservationist discussions. Bioplastics are links of starch-derived polymers synthesized by bacteria. Since these plastics are made from organic material, they are also biodegradable, allowing them to be a promising avenue to decrease our ecological footprint. However, these plastics are rather weak without any kind of strengthening treatment and thus are not suitable for everyday usage. Such treatments usually alter bioplastics' structure, making changes in their tensile strength and durability. This research analyzes the effects of different treatment methods – thermal treatments, hot-pressing, and material addition – on strengthening the mechanical properties of bioplastics and proposes possible applications of the treated plastics. Various studies regarding these treatment types concluded that they increased the crosslinking degree in bioplastics, thereby improving their tensile strength. However, these treatments also induced lower water uptake capacity due to the resulting stiffening. Based on current developments, thermally treated bioplastics can find usage in the medical or packaging industry. Hot-pressed bioplastics are also a promising material for food packaging. Bioplastics reinforced by other materials may find their way into the construction industry. This research can help future researchers in selecting the most appropriate methods to optimize bioplastics for specific industrial applications.

Keywords: bioplastics, dehydrothermal, hot-pressing, reinforced bioplastics

Introduction

Since the inception of plastic, its production and consumption have soared to new heights, and its adverse impact on the environment has been increasingly apparent. Globally, around 46% of plastic waste ended up in landfills, 22% ended up as litter, 17% got incinerated, and only 15% got recycled [1]. As plastics degrade in landfills, microplastics and other toxic substances are released into the surrounding soil and water. As a result, microplastics act as carriers for pathogenic organisms. For instance, microplastics are shown to influence the behavior of earthworms, causing them to borrow differently and affecting their fitness, thus altering the soil quality [2]. Furthermore, soil with microplastics present displays a decrease in the diversity of species of microarthropods, negatively impacting the fertility of the land [3]. However, these effects are mostly from plastics in landfills. Plastics that end up as litter are often swept away by wind or rain into the ocean, presenting a new host of problems. Aquatic organisms can become entangled by plastic bags or ingest smaller pieces of plastic, resulting in the buildup of toxic chemicals in their body [4]. This becomes problematic since it allows plastics to end up in food. Ingesting plastics may cause metabolic disorders, immune responses, or even neurotoxicity [5].

Given the inherent dangers of plastics, more environmentally friendly alternatives are becoming more appealing than ever, as exemplified by the increased attention given to plastic-free shopping experiences in supermarkets [6]. Besides recycling, the development of biodegradable plastics, or bioplastics, can potentially solve the issue with contemporary plastics. Made from biopolymers, bioplastics are a class of plastics derived from renewable sources or through microorganisms. Generally, these biopolymers stem from starch originating in corn, potatoes, cassavas, cellulose, and agricultural biowaste [7]. Certain bacteria can link monomers of these starches together through metabolic reactions, creating a polymer that can be linked together even further to form plastics. While some bioplastics are biodegradable, nonbiodegradable bioplastics still exist, though they are often obtained from corn, sugarcane, and biomass [8]. Nevertheless, the production and circulation of biodegradable bioplastics from renewable sources reduce dependence on conventional plastics, alleviating some of their environmental strains.

Currently, bioplastics find use in packaging, agricultural films, and medical devices due to their biodegradability [7]. Bioplastic degradation was shown to display no statistically significant impact on soil's bacterial diversity, thus decreasing the environmental impact compared to conventional plastics [9]. In agricultural films, bioplastics are used as mulch films, which can help regulate temperature and moisture near plant roots [10]. This promotes the growth of nitrogen-fixing bacteria, which are beneficial to soil and plants. With these factors in mind, bioplastics also offer biodegradability as a bonus, furthering their appeal. In the medical field, bioplastics have the potential to be used as materials for drug delivery, therapeutic devices [11], 3D implants, and scaffolds for tissue regeneration [12]. Since they are made from biodegradable polymers, these plastics can be safely degraded in the human body once entering and are either absorbed into the body or expelled from it.

However, despite their promises, bioplastics face numerous challenges before they can be suitable for mass production and consumption. The conditions to make certain bioplastics are difficult to meet. For bacteria to produce polymers optimally, the salinity, temperature, pressure, and pH environment must be perfect for specific strains of bacteria [7]. As a result, it is difficult to upscale production since the optimal setup for one strain of bacteria might not work for another strain. Furthermore, the cost to maintain these perfect conditions is also relatively high. Despite these problems, tentative solutions have been proposed. Seawater is often used as a low-cost cultivation environment for bacteria as it is abundant and allows for some control over osmotic pressure and pH [7]. With developments in gene editing, the strain selection process of bacteria might become more efficient [13]. Desirable genes may be inserted into the bacteria, allowing them to survive conditions they normally cannot survive or to improve polymer production.

Other than challenges in the industrial production of biopolymers, another important issue is the lack of durability in bioplastics. Due to their biodegradable nature, bioplastics display weaker mechanical properties, such as lower ductility, toughness, and flexibility. Bioplastics are also generally more hydrophilic in nature, thus they absorb less moisture than petrochemical

plastics before their structural integrity gets compromised. While these features do not make bioplastics inherently bad, they severely limit the applications of these plastics. In its current state, bioplastics are best suited for short-shelf life products.

Given this context, the present work aims to discuss the various methods of synthesizing and treating bioplastics and their effects on the products' mechanical properties such as tensile strength and water uptake capacity. Many treatment methods have been discovered to improve the durability of bioplastics, such as using ultrasound, dehydrothermal treatment, hot-pressing, and material addition. With most of the research focusing on the latter three treatments, papers detailing the effects of thermal treatment, hot-pressing, and the addition of other materials on mechanical properties are evaluated.

Methods

A literature review was conducted on articles regarding treatment methods that strengthened bioplastics. Articles were gathered from websites such as Google Scholar, PubMed, Nature, Science, and ACS Journals. A heavy emphasis was placed on accruing primary research papers within the past 15 to 20 years to best reflect the strides recent advances in bioplastics are making. Keywords used included mold temperature, dehydrothermal, hot-pressing, material addition, tensile strength, water uptake capacity, and degree of crosslinking. Both papers on conventional plastics and bioplastics were considered.

Results

Thermal Treatment

Two common bioplastic strengthening treatments involve the usage of heat: dehydrothermal treatment and high molding temperature. Under dehydrothermal treatment (DHT), newly produced bioplastics are subject to high temperatures, usually higher than 90°C. This intense heating removes water between the polymers, initiating a condensation reaction that creates crosslinks between polymer chains. For this type of treatment, the temperature and time spent at said temperature are key factors. An experiment done by Jiménez-Rosado et. al [14] explored the effects of this treatment on the mechanical properties of soy-based bioplastics. Soy protein was chosen since it favored the formation of hydrogen bonds and was cheap to produce. These bioplastics were put in an oven at 50°C for 4 and 24 hours. The bioplastics placed in the oven for 24 hours displayed a higher degree of crosslinking since longer times favor the formation of covalent bonds, indicating that dehydrothermal treatment improved tensile strength and elasticity, which are critical properties of durable bioplastics. The results also imply that a longer DHT time results in a lower water uptake capacity, which might compromise the strength of bioplastics.

Perez-Puyana [15] studied the effects of heat treatment on bioplastics sourced from another legume. After pea protein-based bioplastics were produced, they were placed in ovens at 50 and 120°C for 4 and 24 hours. The exposure to heat would denature the collagen in the

bioplastic, increasing its amino groups' reactivity. Higher temperatures and lengthier times significantly increased the crosslinking degree. However, these conditions also decreased the bioplastic's critical strain and water uptake capacity. The reduction in water uptake could be explained by the rigidity and brittleness of the bioplastic after DHT; more brittle bioplastics will not be able to absorb water and swell as much. Both results indicate that thermal treatment needs to be optimized to achieve a balance of tensile strength, elasticity, and water uptake capacity for durable bioplastics.

While dehydrothermal treatments are done post-synthesis, high mold temperatures are conducted as the bioplastics form. Like dehydrothermal treatments, higher mold temperatures induce the formation of crosslinks between polymer chains. In the same experiment conducted by Jiménez-Rosado et. al [14], bioplastics that were usually made in molds at 70°C were made at 130°C instead (70°C was set as the base temperature since it emphasized the water uptake capacity and 130°C was the maximum temperature the bioplastics could withstand before their proteins disintegrate). The bioplastics produced at 130°C exhibited greater tensile and flexural strength than their 70°C counterparts. However, the increased mold temperature also resulted in a significant decrease in water uptake capacity; the rigid, structured bioplastic had a greater difficulty swelling, thus it was unable to retain large amounts of water. The paper suggests that higher mold temperatures produce flexible, compact, and non-absorbent bioplastics suitable for applications in packaging.

Alonso-González et. al [16] detailed the effects of mold temperatures on bioplastics sourced from rice bran. After preparing blends containing rice bran, water, and glycerin, they were put into molds at 100, 130, and 150°C (the selected temperatures were based on previous studies). Again, higher mold temperatures resulted in tougher bioplastics with higher maximum tensile strengths and strains. Elongation percentage also improved with increased temperatures. As the bioplastics became more compact at higher temperatures, they could not absorb as much water.

Hot-Pressing

With hot-pressing, plant fibers or proteins are pressed between two hot plates. This process exerts both pressure and temperature onto the fiber, compacting the material and removing any imperfections it may have. This reduces the material's porosity, strengthening its structure. A study by Wang et. al [17] detailed the effects of hot-pressing on cellulose-based bioplastics. Cellulose was chosen as a major component due to its strength and toughness (as found in maintaining structural integrity and preventing desiccation in plant cells). Cellulose was dissolved in solutions, producing cellulose hydrogels. The hydrogels were then hot-pressed at a temperature of 110°C and a pressure of 60 MPa, resulting in the production of a bioplastic film. During this process, the polymer chains were reorganized, changing their structure to become a more uniformly oriented structure. The results of this experiment displayed that hot-pressing significantly increased the tensile strength, flexural strength, and thermal stability of the bioplastic when compared to other common synthetic polymers, like acrylonitrile–

butadiene–styrene copolymer, polycarbonate, polyethylene, polyvinyl chloride, and polybutylene terephthalate.

Another experiment detailing the effects of this treatment on cellulose-based bioplastics was conducted by Rumi et. al [18]. This study dissolved cotton fibers into solutions and baked them to produce films. These films were plasticized with varying concentrations of glycerol before being hotpressed at 120°C for 15 minutes. Hot-pressing transformed the cellulose hydrogel into a smooth, flexible, and strong film. The cellulose chains were also rearranged to form more densely packed structures. The results indicate that a combination of plasticizing and hot-pressing significantly improved the tensile strength, flexibility, and elongation percentage. However, hot-pressing did not play a significant role in improving the water absorption of the cotton bioplastic; instead, this improvement was largely attributed to the plasticizer, which was able to impart a hydrophilic character to the bioplastic. While the authors did not optimize the conditions of hot-pressing in this paper, their results suggest combining hot-pressing with other plasticizers can further improve durability without compromising water absorption capacity.

Chen et. al [19] conducted a study regarding the effects of hot-pressing on wheat proteins. Since wheat protein is inexpensive, abundant, and renewable, it was chosen as the source of bioplastics in the experiment. Gluten, gliadin, glutenin with starch, and pure glutenin were first mixed with glycerol. The gluten, glutenin, and pure glutenin films were compressed at 40,000 lbs for 6 minutes at 120, 150, and 180°C while the gliadin films were compressed for 2 minutes under the same conditions (these compression times provided optimum tensile properties). The results indicated that 150°C was the optimal temperature for the hot-pressing of these materials. There was no significant improvement in mechanical properties as temperature increased from 120 to 180°C.

Material Addition

The addition of other materials helps strengthen the bioplastic fibers by stiffening the bioplastics, making them more resistant, or stimulating reactions that form crosslinks between the polymer chains. In an experiment by Judawisastra et. al [20], polyvinyl alcohol (PVA) was added to bioplastics to reduce their brittleness. Due to its abundance of starch, tapioca was the material of choice for this experiment. Four mixtures with differing percentages of PVA by weight were prepared for the experiment: one with 0% PVA for reference, one with 17% PVA, one with 29% PVA, and one with pure PVA for reference. The results from this experiment show that the pure tapioca starch bioplastic had higher water absorption than the 17% and 29% PVA bioplastics. Since PVA and the starch interact to decrease the amount of free OH groups, the bioplastics with PVA content are more hydrophobic, making them less sensitive to water and thus absorb less of it. The results also showed a negative correlation between PVA content and tensile strength. Change in PVA content also resulted in no statistically significant difference in the bioplastic's stiffness retention and elongation. However, after these bioplastics were submerged in water, the addition of PVA displayed a significant increase in elongation, decrease in tensile modulus, and no significant change in tensile strength.

An experiment from Amri et. al [21] proposed the addition of graphene oxide (GO) to tapioca starch bioplastics. Different percentages of GO by volume (0, 5, 10, and 15%) were mixed with the tapioca solution and stirred for two different durations (30 and 60 minutes). When the concentration of GO increased, tensile strength increased as well. This is attributed to the increased interaction between the OH groups in the starch and GO. However, as the tensile strength increased, elongation percentage decreased along with it. The same trend was observed in stirring time as well. Longer stirring times yielded higher tensile strengths but decreased elongation percentage. The increase in tensile strength results from the even spread of GO particles due to the long stirring time. Based on these results, a solution with 15% GO mixed for 60 minutes would yield the highest tensile strength.

In Harunyah et. al [22], clay nanoparticles were added during the bioplastic film production process as reinforcements. The starch used for the bioplastic came from cassava, another abundant source of starch. The resulting bioplastics contained clay nanoparticles at 0, 0.2, 0.4, 0.6, 0.8, and 1.0% by weight. They were also plasticized with glycerin concentrations of 25, 30, and 35%. This experiment demonstrated that the addition of clay nanoparticles increased the tensile strength to an extent before diminishing and decreased the percentage of elongation. The presence of clay nanoparticles contributed to a higher thermal stability in the bioplastics. An increase in the glycerin plasticizer concentration tended to increase elongation percentage while reducing the tensile strength. The results concluded that the optimum conditions for tensile strength was at a clay nanoparticle concentration of 0.6% and a plasticizer concentration of 25%.

Discussion

Paper	Treatment Type	Crosslinking Degree/Structure	Tensile Strength	Water Uptake Capacity
Jiménez-Rosado et al. [14]	DHT	Longer time spent at high temperatures induced a higher degree of crosslinking. However, it took several hours before higher degrees are shown, making this treatment more costly.	DHT improved maximum stress, strain at break, and Young's modulus, though there wasn't a significant difference between the two treatment times of 4 hours and 24 hours.	DHT significantly decreased the bioplastic's water uptake capacity under longer periods of time. The bioplastic was still functional and super absorbent.
Perez-Puyana et al. [15]	DHT	The results demonstrated that higher temperatures and longer time spent under these temperatures improved degree of crosslinking in a statistically significant way.	Higher temperatures and longer heat exposure time resulted in a more elastic character. However, these conditions significantly reduced the critical strain value. When the treatment was implemented at higher temperatures and for a longer duration, the bioplastic got stronger but more rigid.	Higher temperatures and longer time also compromised the water uptake potential of the bioplastics. At 120°C at 4 hours and 24 hours, water absorption was reduced from 22% and 50%, respectively.
Jiménez-Rosado et al. [14]	Mold Temperature	High mold temperatures at 130 °C significantly improved in the degree of crosslinking when compared to DHT (32.2% increase compared to DHT's 0.8% and 22.5%).	A mold temperature of 130°C improved maximum stress, strain at break, and Young's modulus. This improvement isn't statistically different from DHT's effects within the same experiment.	The high mold temperature made the bioplastic lose its super absorbent quality by more than 2000% when compared to the reference bioplastic.
Alonso-González et al. [16]	Mold Temperature	Not available	Bioplastics obtained from higher mold temperatures improved Young's modulus, maximum tensile strength, and elongation at break. However, they become stiffer.	Higher mold temperatures resulted in lower water uptake capacities.
Wang et al. [17]	Hot-pressing	Hot-pressing stretched the cellulose chains in the bioplastics, reducing their crystallinity. This treatment also resulted in a layered dense packing structure in the bioplastics.	The tensile strength of cellulose bioplastics significantly improved compared to those of conventional plastics.	Not available
Rumi et al. [18]	Hot-pressing	Hot-pressing disrupted the crystalline structure of the cotton cellulose. The cellulose structure became more amorphous.	Results imply that plasticizing and hot-pressing the cellulose significantly improved tensile strength but reduced elongation at break.	Hot-pressing did not play a major role in improving water absorption.
Chen et al. [19]	Hot-pressing	Not available	There was no significant improvement in modulus and breaking point elongation as the temperature increased from 120 to 180°C.	Not available
Judawisstra et al. [20]	Material Addition	As more PVA gets added, the bioplastics' structure becomes more similar to PVA.	Bioplastics with a higher PVA to tapioca starch ratio decreased in tensile strength and tensile modulus while increased in elongation percentage.	As the PVA to tapioca starch ratio increased, the bioplastics absorbed less water.
Amri et al. [21]	Material Addition	Not available	Higher concentrations of graphene oxide and longer stirring times resulted in an increase in tensile strength and Young's modulus but a decrease in elongation percentage.	The addition of graphene oxide increased water uptake capacity while longer stirring times decreased it.
Harunyah et al. [22]	Material Addition	Not available	The addition of clay particles (from 0% to 0.8%) tended to increase tensile strength before decreasing it at a clay nanoparticle concentration of 1%.	Not available

Table 1. Treatment types and their effects on crosslinking/structure, tensile strength, and water uptake capacity.

Desired Properties of Bioplastics

It is evident that a heavy emphasis is placed on the tensile strength and water uptake capacity of the bioplastic. When judging the effectiveness of each treatment in enhancing these mechanical properties, the degree of crosslinking is a key factor. The more linkages the treatments can induce, the stronger the bioplastics become. Crosslinking creates a network structure among polymer chains, increasing their uniformity. As a result, the bioplastic can evenly dissipate stress, reducing the occurrence of localized stress concentrations. Furthermore, this crosslink network limits the movement of polymer chains under stress, preventing the chains from deforming. The effects of these improvements are displayed in a study by Martinez et. al [23], which demonstrated how higher degrees of crosslinking significantly increased a polymer's resistance to strain.

However, the rigidity induced by crosslinking also reduces the bioplastic's water absorption. The crosslink network prevents the bioplastic from swelling, thus limiting the plastic's ability to retain the water it absorbs. Without these networks, water absorption will change the dimensional shape of the bioplastic, making it more vulnerable to warping and distortions. Water uptake can result in the extraction of water-soluble components in plastics, altering the structure of the polymers drastically [24]. Furthermore, Jones et. al [25] mentioned how water absorption can change the elastic modulus of the bioplastic, thus compromising the tensile strength of the original plastic as weak spots become more prevalent.

Even though a higher water uptake capacity has a negative impact on tensile strength, that is not to say it is an undesirable quality. The unstable dimensional shape heightens the degradability of the bioplastic. In a world where efforts are constantly made to produce eco-friendly materials, bioplastics with high water uptake capacities are an enticing option.

Challenges and Applications of Durable Bioplastics

The results of the treatment methods discussed in this paper demonstrate the promising future of bioplastics. Across all methods, the amount of crosslinks and tensile strength increased while water uptake capacity decreased. Although both dehydrothermal treatment and higher mold temperatures resulted in decreases in water uptake capacity, DHT produced bioplastics with a controllable decrease in water uptake capacity while higher mold temperatures produced bioplastics without being superabsorbent. The strong and non-absorbent bioplastics made using high mold temperatures are thus suitable for applications in the packaging industry, where tensile strength is heavily valued and where moisture absorption is unlikely to occur. Meanwhile, the strong and rather absorbent bioplastics made from DHT may find uses in biomedical implants. These implants require high tensile strength since they act as scaffolds for tissues or sutures. The super-absorbent nature of bioplastics made from DHT allows these scaffolds to degrade in the body, allowing for the regeneration of tissue without needing surgical removal. Studies have shown that DHT-treated bioplastic scaffolds hold potential for medical usage. In a study done by Haugh et. al [26], the enhanced durability of DHT-treated collagen scaffolds was deemed

sustainable for use *in vitro* and *in vivo*. Hot-pressed bioplastics show promising results for future applications in consumer products. The rigid structure of these bioplastics improves their tensile strength and durability, making them suitable for being used for food packaging. In a review done by Harnkarnsujarit et. al [27], hot-pressed bioplastics are reported to greatly improve oxygen barriers due to the crystallization and restructuring of the base bioplastics. This suggests that hot-pressed bioplastics are viable materials for food packaging as they can effectively block out oxygen, preventing the oxidation of food. Bioplastics strengthened by the addition of other materials might see usage in industries with stiff parts and unmovable parts. These bioplastics may find their way into the construction industry. As stated by Ivanov and Stabnikov [28], bioplastics can provide a source of cheap and biodegradable raw materials. Coupled with their strength, stiffness, and inelasticity, these reinforced bioplastics are suitable for parts like wall claddings or facades. However, in its current state, the quality of bioplastics in regards to life-span and response to biodeterioration agents, as stated in a paper by Friedrich [29].

Conclusion

Bioplastics are a promising avenue for reducing environmental impact. Although they are rather weak, different treatment methods can be used to strengthen bioplastics. The effects of three methods including thermal treatment, hot-pressing, and material addition on the mechanical properties of bioplastics were discussed.

Thermal treatment can be done in two ways; through dehydrothermal treatment and using high mold temperatures. DHT is conducted during the synthesis of the bioplastic, relying on heat to cause condensation reactions to join polymer strands. High mold temperatures are done post-synthesis and also use high temperatures to connect polymer strands. Based on other research, both DHT and high mold temperature treatments increased bioplastic durability and decreased absorbency. However, the absorbency of bioplastics decreased far greater under high mold temperatures than under DHT. It is speculated that DHT bioplastics have potential in the biomedical industry due to their ability to maintain some level of absorbency in bioplastics. Bioplastics that underwent high mold temperatures may see usage in food packaging because of their high tensile strength and low absorbency.

Hot-pressing applies pressure and heat to fibers, thus compacting the materials and forming a strengthened structure. The resulting bioplastics are rigid and durable. These factors coupled with improvements in bioplastic structure that block oxygen, bioplastics made from this treatment are also suitable for the food packaging industry.

Bioplastics reinforced through the addition of other materials rely on these additions to become stiffer or stimulate the formation of crosslinks. Bioplastics made from this treatment are stiff and inelastic, making them a great potential substitute for construction materials.

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**Severe Wage Stagnation and Cost of Living Growth Since 1980:
An Analysis of Young Americans' Financial Distress and Potential Policies for Addressal
By John Wu**

Abstract

This paper draws from data from various national sources to analyze and properly extrapolate conclusions regarding the United States' growth in wages in comparison to the growth in prices of multiple significant expenditures for urban citizens throughout the last 44 years. Through this study, the cost of living growth for the majority of Americans is explored with the perspective of real dollar cost indexed to 1980 to show the past 44 years of economic change. Using computer modeling, a linear regression prediction is made to model how these changes will develop in the future if left to continue in their trends, keeping in mind recessions and the volatility of certain expenditures. This paper shows that specific categories of cost, especially the main ones included in the basket of goods and services, as well as widely prevalent and essential costs like health insurance, college tuition fees, groceries, utilities, and housing, have far outgrown and outpaced the wage growth over the same period. This paper also offers potential government solutions in policies to help alleviate the stresses plaguing young citizens that limit economic mobility and widen the income and quality of life disparities between classes.

Introduction

Wage stagnation in America has grown to be more and more well-known as a concern to America's people due to increased publicity and is now a popular topic of discussion when discussing the future of America's economy, the success of its citizens, and the policies that govern how our country should proceed forward. In order to better understand this subject, its history and possible consequences of allowing the observed trends to continue, and how far wages have grown relative to relevant expenditures for urban citizens, this research paper will delve into the phenomenon of wage stagnation in the United States starting from the past 44 years and expand our viewpoint a decade into the future while accounting for recent recessions for modeling.

While there is much commotion about how prices have jumped significantly, many places only use nominal dollars for their comparisons. While this is effective and can provide some information as to the difference in wages and costs, it neglects to consider inflation. Adjusting nominal dollars to constant dollars will allow for a real evaluation of the power, growth, and/or stagnation of data points. Thus, in this paper, we will compare the real buying and spending power increases of the average salary to the real cost growths in significant and prevalent spending areas. Using the data found, this paper will analyze the results, extend a prediction into the future with relevant statistics, and offer potential targets to address these issues.

The implications of these findings are profound, not only for the workforce but for the broader socio-economic fabric of the United States. As wage stagnation persists, the ripple effects are felt across consumption patterns, savings rates, and even social mobility, thereby reinforcing the cycle of inequality. This necessitates a multi-faceted policy approach that not only addresses the symptoms of wage stagnation but also its underlying causes, promoting a more equitable and sustainable economic growth model.

Problem & Reasoning

Starting from 1980, the past 44 years have witnessed significant stagnation in median and mean income and weekly earnings for urban citizens, contributing to the widening gap between wage growth and the skyrocketing costs of essential expenditures such as healthcare, college tuition, and housing. Wages in the United States have risen consistently slower in comparison to the much faster rise in the costs of essential goods and services like the widely recognized rights to life of food, shelter, and healthcare (Shaak, 2022). This discrepancy highlights a growing concern over the affordability and accessibility of basic needs for urban citizens, further exacerbating the challenges of wage stagnation. The disparity between slow wage growth and rapid increases in living costs underscores the pressing need for policy interventions aimed at bridging this expanding gap. On top of this, college tuition fees are ever more expensive and ever more prevalent as a leading cause of concern and financial stress among young people in America, as college education has become essentially a requirement for young urban citizens to access careers and, subsequently incomes to support themselves in a society with ever-growing costs of living (Meyer, 2024). College degrees are often considered to be a basic requirement for living in a modern society, yet come saddling new members of the American workforce with tens to hundreds of thousands of dollars of student debt (Shireman and Fast, 2023). This has long-lasting repercussions for graduates, as these monthly payments limit the amount of leftover money to be spent on housing, car payments, car and health insurance, and various necessities like groceries, clothing, utilities, and technology. Due to living on a stricter budget, modern graduates are unable to pursue the same lifestyles as their parents and grandparents; often, they are forced to share apartments for more affordable rent, wait until their forties to afford a home, purchase lower-quality products and thus be forced to spend more on replacements, are unable to buy in large bulk and reach discounts, eat more cheaply while sacrificing often more expensive healthier restaurants and groceries, and finally neglect long-term investments without spare time or money to spend on common generational wealth builders like real estate and stocks. These effects have led to and will lead to continued repercussions across generations as those uninhibited by these financial stresses continue to grow while the lower economic classes are held in place without significant chance for mobility.

Since 1959, we have seen a significant increase in spending allocated towards housing (Meyer, 2024) and diverted away from living necessities like groceries and clothing, which points towards an unwelcome development: American citizens are facing a future entrenched in debt without reaching the same qualities of life their predecessors only a couple generations ago

could. If these trends continue without serious and immediate addressing through widespread government policies of limiting price growth in privatized essential industries, costs of essential items and services could strengthen their chokehold on future Americans' prosperity.

To properly make positive social progress and push towards changing the private systems currently in place, our biggest tool is voting. A large portion of American people are unable to boycott essentials due to their already precarious financial position - they, both metaphorically and literally, cannot afford to simply not purchase these goods and fund the industry exploiting them. Thus, to undertake the process of real change, we must look towards politics. American roots are found deeply entrenched in democratic solutions, and our country is set up such that this is one of our only options for bringing change to our society. Voting for the right candidates who will put into motion the bills and policies that will benefit the people most is extremely important, as well as knowing the effects, history, and logic of proposed policies and solutions. In these cases, awareness is vital - awareness is what will propel the engine of the people to move together in a positive and productive direction. Having a fully educated perspective on relevant subjects in the political and socio-economic spheres is highly important for each citizen to fulfill their civic duty to the greatest capacity. This paper aims to serve as a wide-spanning source of information for the subjects covered, such as to spread awareness of the current situation as well as the potential consequences and solutions.

Literature Review

A study from the Economic Policy Institute provides a detailed analysis of the near stagnation of hourly wage growth and its consequences for living standards for the majority of American workers. This source also delves into the causes of wage stagnation and inequality, providing valuable insights into the factors contributing to this issue, arguing that rising income inequality and slow growth in living standards for low- and moderate-income Americans are key economic challenges that need to be addressed. Hourly wage growth has significant consequences for American living standards, as the majority of Americans rely on their paychecks for their income, with many having fewer and fewer leftover real income compared to what those many decades ago were able to save away. They argue that wage stagnation for the majority of American workers was not caused by abstract economic trends but rather by policy choices that favored those with the most income, wealth, and power.

Furthermore, these shocking wages greatly target and affect recent college graduates, showing that many of them are experiencing stagnant hourly wages and declining access to employer-sponsored health insurance (Mishel, Lawrence, et al., 2015) Thus, on top of having to vie for even more competitive career opportunities and being faced with overwhelming experience requirements and a huge field of competitors, they are often left with less real pay despite an increased base hourly from the past. Greatly growing expenditures are forcing many to fork over a large portion of their income and be stripped of their ability to progress higher up the economic ladder without money to spend on long-term investments or better living conditions and opportunities.

We additionally see that while worker productivity has actually risen in modern years, the minimum wage has been sorely left behind, which points towards a stagnation in the middle class as CEOs and the top one percent of society have had their wages grow to exorbitant levels, with CEOs now making 296 times what a typical worker makes, and top one percent wages growing by 138 percent since 1979 (Mishel, Lawrence, et al., 2015). These show an unfortunate and unfair imbalance between the elite of society and those working under them, with little chance to make their way any higher up the social ladder.

Specific errors, such as CAFE standards and monopolies in the auto dealership industry, lead to inefficiencies that can impact consumers negatively and drive up cost of living (Furth, 2015). The report also suggests that American industries should reevaluate these policies to understand the full effects and see where policy changes can create more favorable outcomes, with a key example being local land-use laws - a key area for potential reform to alleviate financial burdens on residents. Local officials have the power to influence costs through zoning regulations and permitting processes. By adopting less stringent rules and expediting projects, local leaders can make living more affordable for their constituents.

The report emphasizes proactive measures to rectify costly policy mistakes and enhance economic efficiency, calls for a shift towards market-based policies, and highlights the potential benefits of such approaches for consumers. By advocating for smart policy changes and a broader perspective on costs and benefits, we can bring improvements in living standards and affordability for all.

In order to tackle the cost-of-living crisis and protect workers from inflationary pressures, the government has many possible methods and policies; one approach is to implement benefit systems that redistribute income more equitably, like imposing windfall taxes on corporate profits similar to what countries like the United Kingdom and Spain have done in the energy sector (Janssen, 2023). Targeting excessive corporate profit-making could help redistribute the earnings back to workers, social benefits, or public investments. Furthermore, a global minimum tax of 15% on profits of multinational enterprises, as part of the OECD agreement signed by numerous countries, could potentially generate substantial additional global tax revenues annually.

Janssen discusses that by strengthening wage institutions and supporting collective bargaining and minimum wage agreements with social partners, like what has been implemented in Portugal, societies can increase the flow of national income to wages and provide fiscal incentives to companies that enhance wages through collective bargaining. In Colombia, minimum wage indexation is enforced in the absence of agreements between social partners. This emphasis on improving wages and social safety benefits aims to curb the disproportionate burden of the cost-of-living crisis on low-income earners, especially in terms of basic necessities like energy and food. Another key player in this issue is trade unions, as they advocate for government actions to bolster social benefit systems; there is a clear correlation between trade union density and the effectiveness of income redistribution through social benefits. Efforts to make social benefits poverty-resistant are underscored as crucial in protecting vulnerable

populations from the financial strains of inflation.

Overall, a collaborative approach between accountable governments, responsible social partners, and supportive monetary policies is key to facing the challenges posed by the cost-of-living crisis and ensuring the well-being of workers.

Methods and Materials

The goal of this investigation was to answer the following question: How severely have wages stagnated in comparison to the real growth of costs of living in America? To accurately gain a perspective on the comparison of today's wages and expenditures to the past, the study looks at the past 2 generations of employees and workers, thus starting the data collection from 1980 and choosing data from urban cities due to the concentrated nature of cities more accurately showing the growths of costs and incomes in different sectors. All programming and analysis was done using Python and can be accessed here:

<https://github.com/wu0122/Indexed-Expenditures>

Throughout the collection of data, it was difficult to find proper data ranging back to 1980 for the average annual health expenditure per capita. As mentioned in the methodology, this was remedied by taking the total national health expenditure and adjusting it to the population growth. Since what we are measuring is an index of growth, the actual data values of health expenditure are not as important as the fact that the national health expenditure is roughly proportional to the growth of health costs for an individual. For the actual values predicted, we use modern annual household mean healthcare expenditures for dates we do know and base the past and future values on them.

Another limitation of this study is that using a linear regression can be less accurate for more complex patterns and curves. Using a polynomial regression of a higher degree would provide more accurate prediction results but also comes with the caveat of potentially creating an artificially high future prediction thanks to not being able to inherently predict the ups and downs of price growth in a complicated system. Polynomial regressions of higher degree, unlike sinusoidal functions, are not waves and end up with quickly-growing functions and slopes on the future side. In the end, I decided to use linear regression for a more understandable and less bold claim for the volatile future. One can see when comparing the real index data to the prediction data that the index values do not line up. This is due to the nature of an approximation, so these predictions are not to be taken as completely exact but rather ballpark estimates. These estimates do not account for any potential spikes or drops in the next ten years; only the overall growth rate of the 44-year or more recent years data.

Choosing and Sorting Data

As a general rule, except for when not applicable to datasets, the study chooses seasonally adjusted data to showcase trends in order to avoid seasonal fluctuations in prices and expenditures, as well as chooses to use constant dollars to account for inflation such that the growth in expenditures done by indexing is accurate to the real amount of money spent. Seasonal

adjustment is made when data is measured monthly/quarterly as opposed to annually; the adjustment is made by removing the predictable seasonal influences such as school schedules, holidays, or the seasons themselves that affect various rates of spending through taking unadjusted monthly or quarterly data, dividing by the seasonality factor, and multiplying by 12 or 4 respectively. The seasonality factor is the ratio of the amount in one month/quarter divided by the average amount per month/quarter over an entire year to get the relative amount of the data that fluctuates seasonally. In selecting data, the study chooses from several datasets that can give a reflection of how multiple sources of expenditure have grown in comparison to the growth in mean wages, selecting the Consumer Price Index (CPI), average yearly expenditures per capita for healthcare expenditures, groceries (food away from home), gasoline, house sale prices, and tuition costs and fees. One exception to this was rent, instead taken as median, as consistent average rent price data was not found for years spanning to 1980. Since we have used averages for these expenditures, we chose the mean income as our marker of growth for this study. Although the mean income is influenced by outliers more than the median, mean measurements for the expenditures are also affected in this way, and thus for a proper comparison, we must use the same measure of center.

For CPI, we use CPILFESL, or CPI, considering all items less food and energy, as those prices are often volatile and prone to extreme change that makes a good analysis of real, constant dollars less accurate. Since the analysis seeks to measure growth, cost indexes will be used as the data. Indexing is a common practice to measure growth in data, where one data point or value corresponding to a certain time or other value is chosen as a base and set as 100. Then, the other values are indexed to be relative scales of the base value. The general form of this process is:

$$\left(\frac{\text{New Value}}{\text{Base Value}} \right) * 100. \quad (1)$$

However, before one can set these indexes for analysis since this study is looking at the real change in the spending power and costs in relation to the salary and not simple price growth, the dollar costs should all be within the same constant dollars. Constant dollars is simply a term for when one takes nominal dollar data, which is expressed as the price one paid for a certain good or service at that time, and adjusts according to the inflation change such that we see the real cost or amount of money reflected in today's dollars. By adjusting to constant dollars, this study can provide insights into the actual burden of cost growth, as pure growth without inflation adjustment only tells half of the story. When we look at constant dollar growth, it can tell us that the real value of purchasing power as several expenditures have changed to become greater portions of the wage. To get constant dollars from the more common nominal dollar data, one iterates the CPI inflation adjustment process over the data values. This method is taking a base year's currency to adjust the rest of the values by taking:

$$\text{Nominal Dollar Value} * \left(\frac{\text{CPI Value of Base Year}}{\text{CPI Value of Nominal Year}} \right). \quad (2)$$

As mentioned earlier, the CPI used in this calculation is CPILFESL to more accurately reflect dollar values and disregard variable energy and food prices. For one source, the healthcare expenditure, since national spending was the only measure of data found, this study accounted

for the growth in total national healthcare expenditure that comes with an increasing population by adjusting the nation’s population in a manner similar to CPI adjustment. The process was:

$$\text{National Health Expenditure} * \left(\frac{\text{Base Year Population}}{\text{Nominal Year Population}} \right). \quad (3)$$

Predictive Models

Finally, for a predictive model, we use a simple linear regression program from the data provided. Linear regression is a statistical process that uses a line of best fit to model the relationship between two or more variables and estimate the value of a response. The line of best fit is a straight line that most closely correlates to all the values in a dataset with the equation $y = \beta_0 + \beta_1 x$, with β_1 representing the slope and β_0 representing the y-intercept, or where the y-value is when $x = 0$. To do this, you first find the sum total of “x” and “y” values, in this case, x representing time in years and y representing dollars. Using the following expression and finding the rest of the values in this expression, where \sum represents the summation of the variables and “n” represents the number of data points provides β_0 .

$$\frac{((\sum y)(\sum x^2)) - (\sum x)(\sum xy)}{(n(\sum x^2) - (\sum x)^2)} \quad (4)$$

Expression 5 gives us the best-fit slope of the line, β_1 .

$$\frac{n(\sum xy) - (\sum x)(\sum y)}{(n(\sum x^2) - (\sum x)^2)} \quad (5)$$

This was done separately for data patterns that were significantly affected by recessions, such as the housing recession of 2008, creating a new line after the recovery. To find values that follow the pattern of growth, one takes the equation and uses the following year's values as “x” to calculate the corresponding “y” values. For the actual future values, we have used a formula taking modern data that tells the current annual mean expenditures and based the other values of the dataset using that real data using the process of:

$$\text{Real Measured Data in Dollars of Year} * \frac{\text{Nominal Index Data}}{\text{Current Index Data}}. \quad (6)$$

Results

The following figures and table are the results of the data collected being analyzed and processed to present as index values and future prediction values. The five graphs are graphs of the index data of each expenditure, with the last three providing highlights along with linear regression lines. The table shows the coefficient and intercept for each expenditure’s data, along with R^2 , adjusted R^2 , and standard error. For additional pre-calculated data for regression predictions, see other resource documents. It is to be noted that all dollar values for future predictions are in 2024 constant dollars. For more data on the flat base indexes from 1980 without adjustment for real dollars, one can multiply the index data by the CPILFESL index data from the year one would like to see.

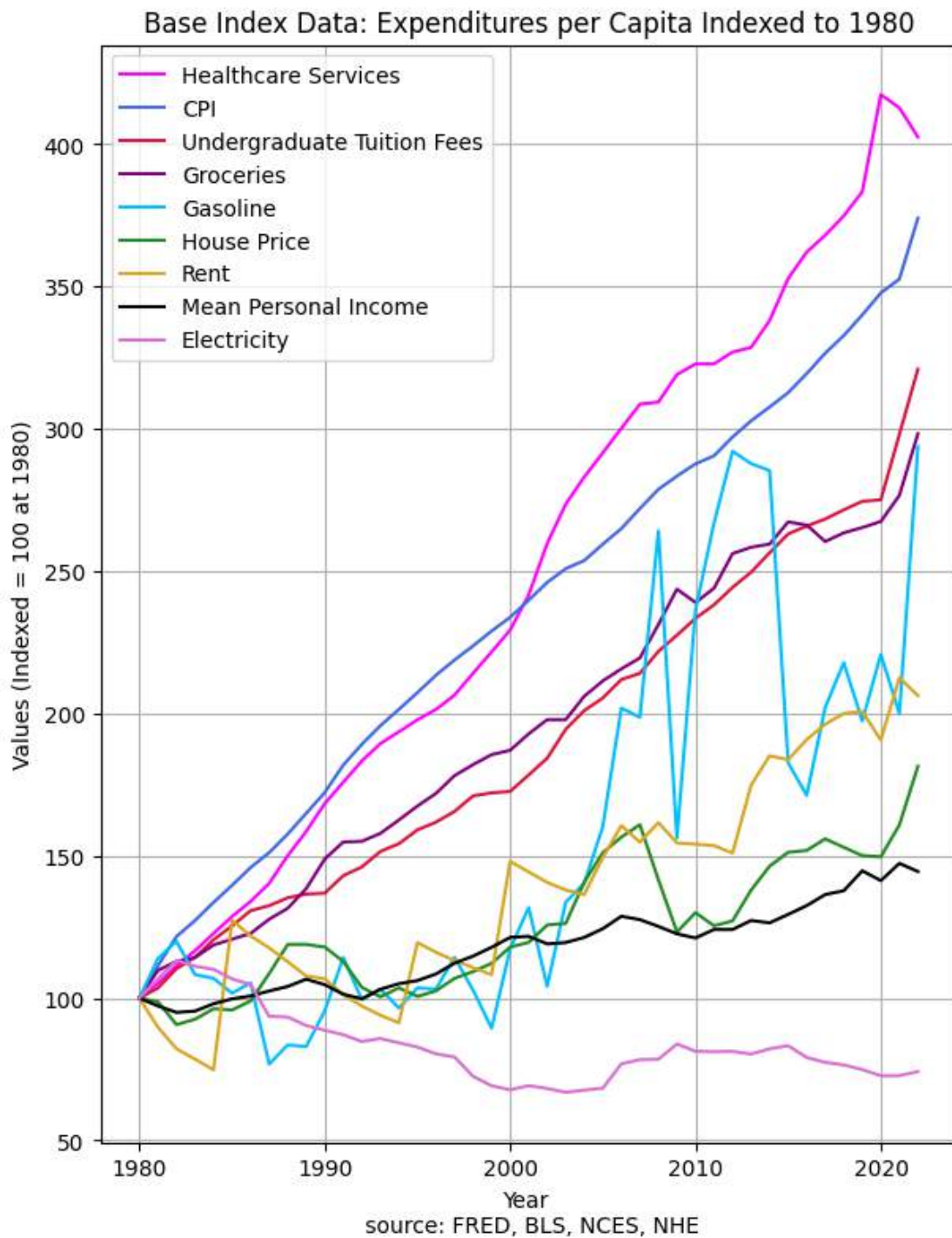


Figure 1:

This figure shows several common expenditures compared to the mean personal income through CPI adjusted dollars indexed to 1980.

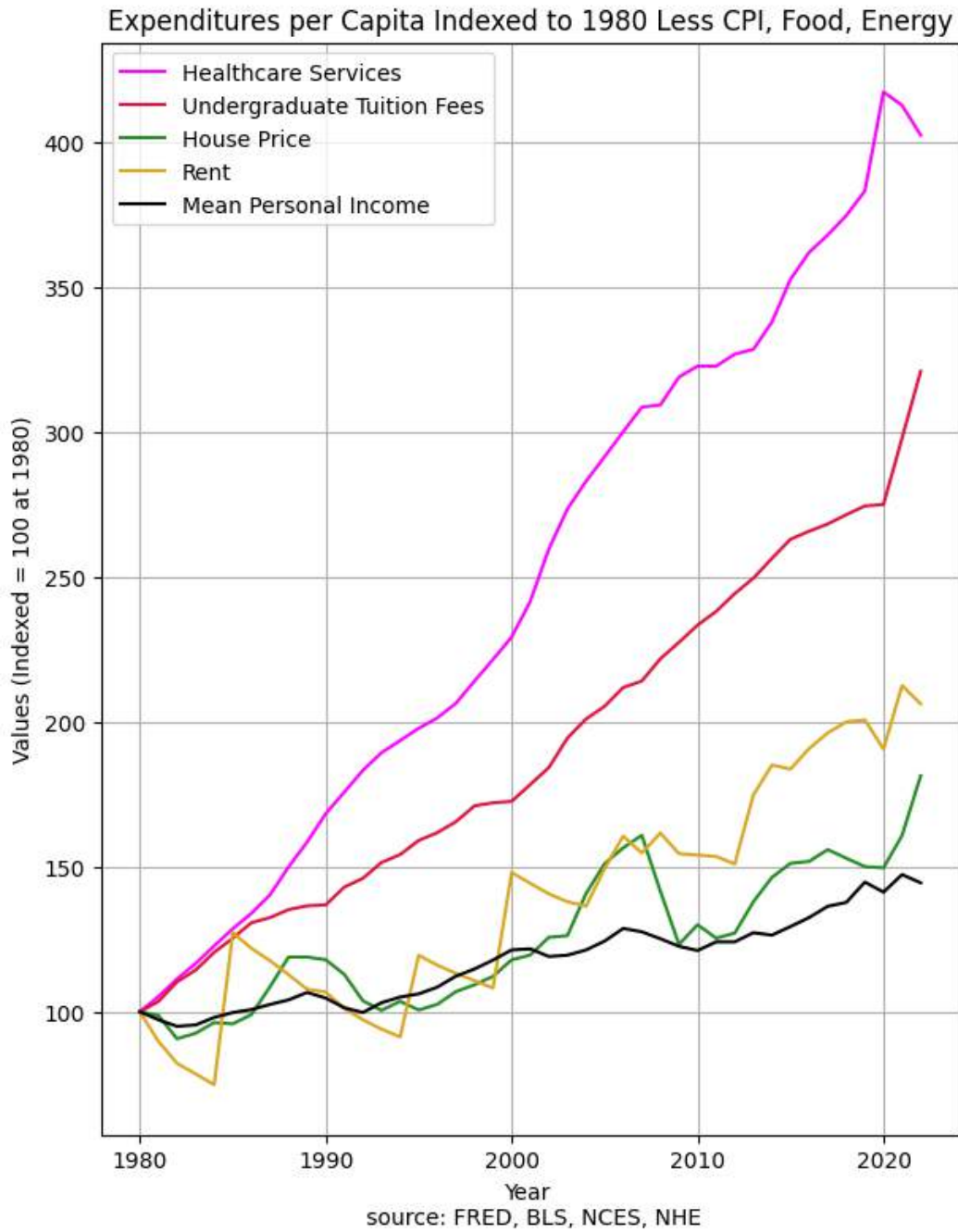


Figure 2:

This figure shows healthcare, undergraduate tuition fees, house price, and rent compared to the mean personal income through CPI adjusted dollars indexed to 1980, taking out the more volatile utilities and groceries values.

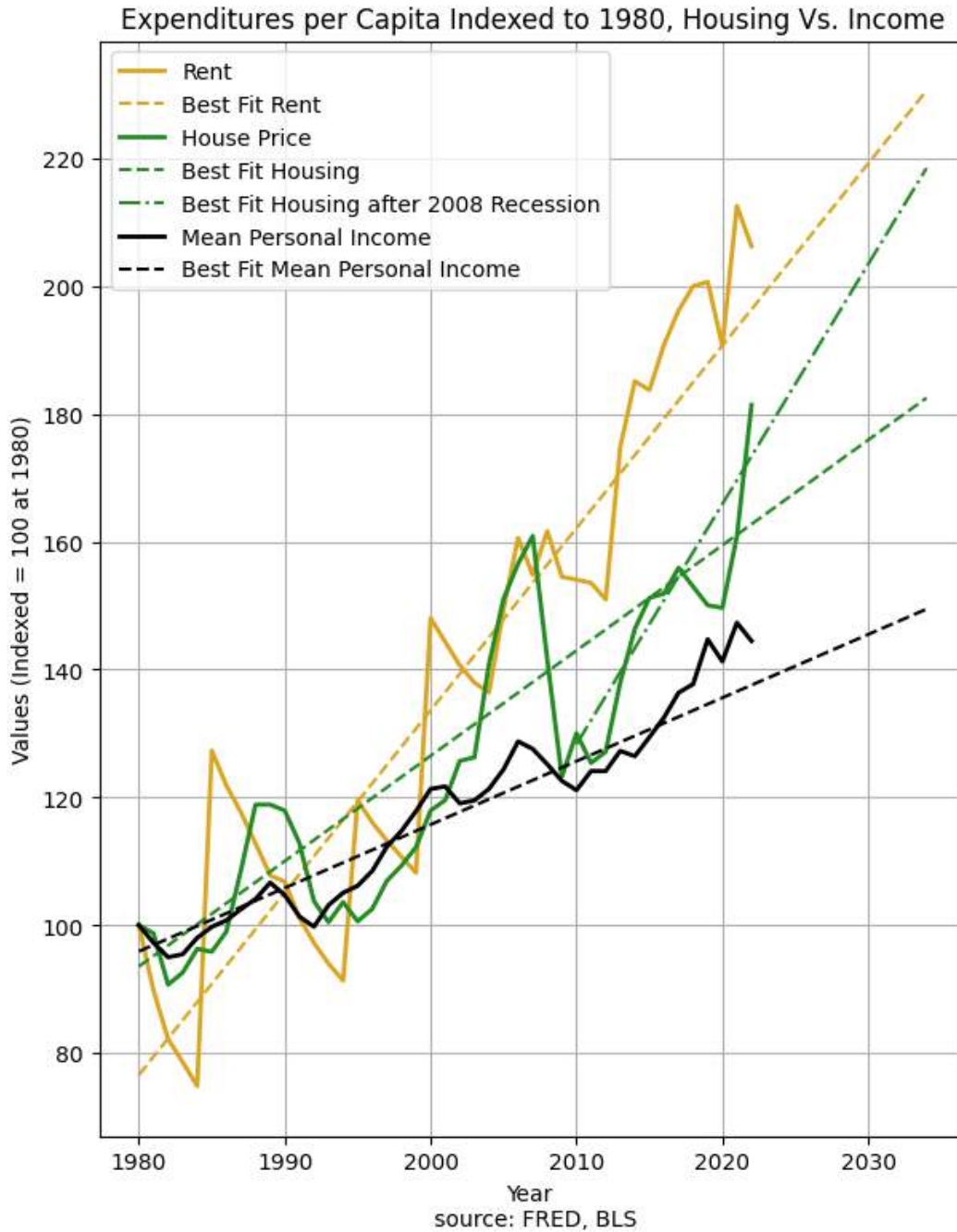


Figure 3:

This figure shows house and monthly rent prices compared to the mean personal income through CPI adjusted dollars indexed to 1980. The three are fitted with linear regression models, with house price having a second estimation based on data after the 2008 recession.

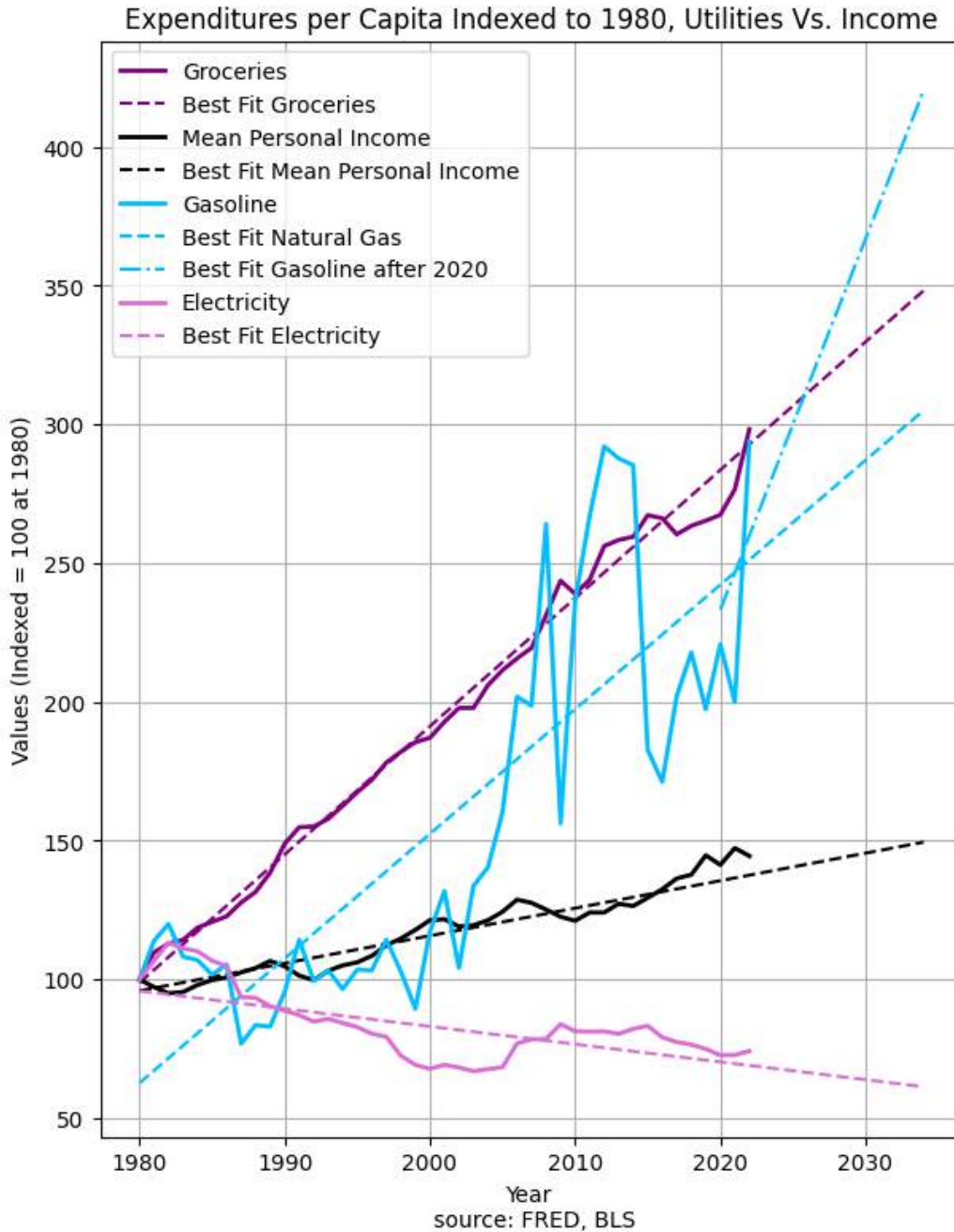


Figure 4:

This figure shows the more volatile groceries, natural gas, and electricity prices compared to the mean personal income through CPI adjusted dollars indexed to 1980. All are fitted with linear regression models, with gasoline having a second line fit with data after 2020.

Expenditures per Capita Indexed to 1980, Tuition & Healthcare Vs. Income

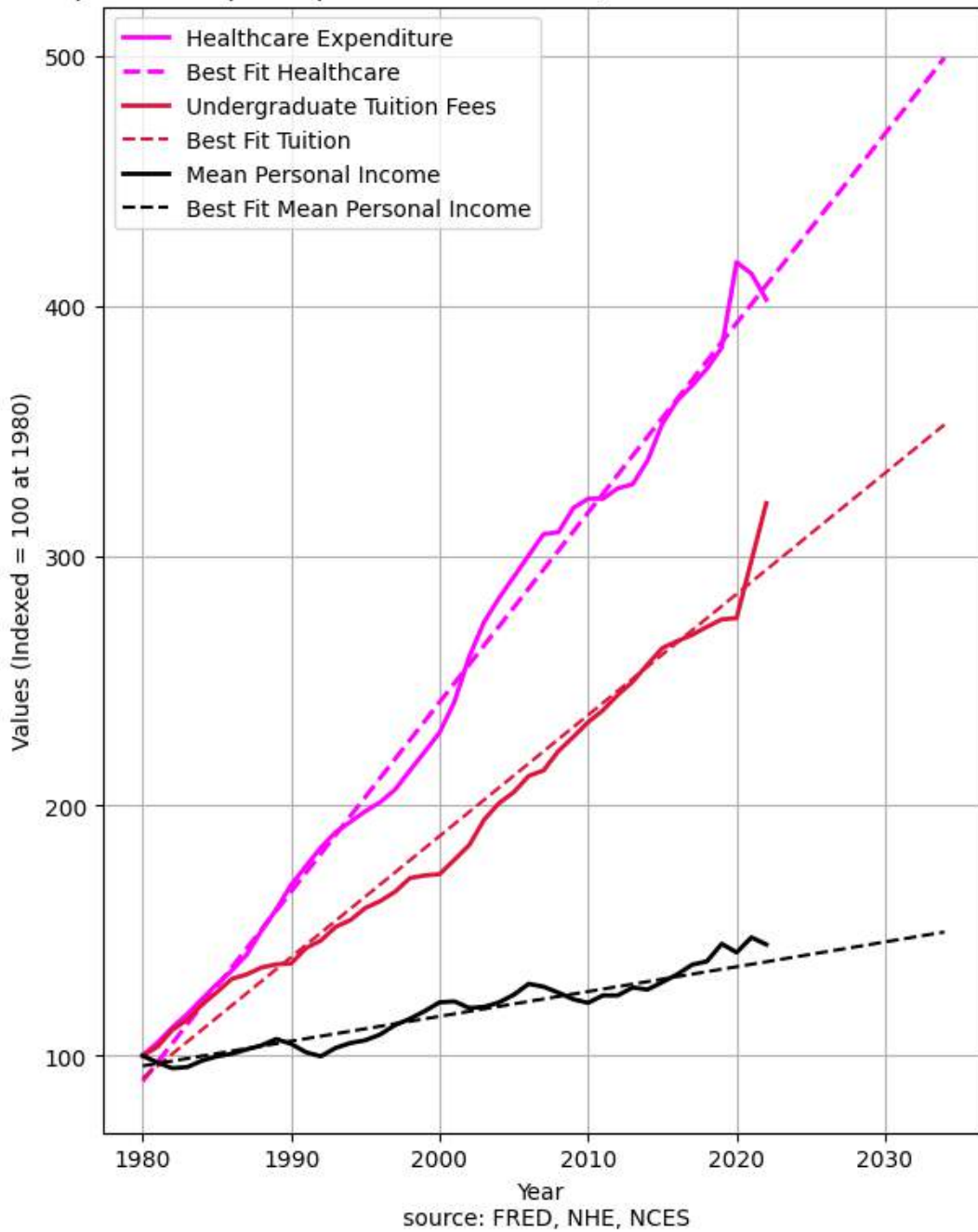


Figure 5:

This figure shows the healthcare expenditure and tuition fee prices compared to the mean personal income through CPI adjusted dollars indexed to 1980. All are fitted with linear regression models.

Table 1:

Expenditure	Coefficient	Intercept	R ²	Adj. R ²	Std. Error
Electricity	-0.639208	1361.482156	0.470093	0.469086	8.634252
Groceries	4.618517	-9045.828084	0.982798	0.982388	7.941739
Healthcare	7.580076	-14918.74671	0.992051	0.992036	8.436395
Rent	2.852678	-5571.740312	0.882791	0.882568	13.224463
House Price	1.647638	-3168.775351	0.886161	0.885692	10.484637
House Price After 2010	3.756203	-7421.644261	0.748875	0.744309	9.112354
Income	0.991952	-1868.160022	0.880116	0.879911	5.186285
Natural Gas	4.488673	-8824.900205	0.684289	0.676947	40.506998
Natural Gas After 2020	13.363616	-26761.29842	0.740172	0.707694	25.426253
Tuition	4.840003	-9492.158502	0.971435	0.970756	10.787006

Costs of almost all measured values have increased significantly, most especially healthcare expenditures and tuition fees. This reflects and validates the common idea held among American young adults that college tuition and, subsequently, student debt is growing extraordinarily quickly and burdening a large majority of the 20-30-year-old population. Tuition fees rose quite high to 3.2 times their average amount in 1980, and they could reach a 15.9 percent increase within 10 years based on the calculated linear model (“Average Undergraduate Tuition”, 2023). Additionally, we have seen that healthcare in the United States has exorbitant prices for those with health insurance and is completely unreasonable for those without it, which is made even more evident when compared to other highly modern countries like Canada and Taiwan. In 44 years, healthcare has grown the quickest out of all data points at 4.02 times their 1980 amount, or around 2.87 times the amount that the average income grew (Centers for Medicare & Medicaid Services, 2023). If the predictive model is an accurate reflection of the health insurance and expenditure values that have been quite steady in their growth rate, then Americans could be looking at an 18% increase in costs over the next 10 years. As shown in Figure 5, the spread of data points is quite close to the linear regression line, telling us not only that the rate of growth has been relatively stable over the past 44 years but also that a future prediction based on this model is more accurate. As discussed in the Literature Review, not only are many college graduates having a harder time getting jobs, but those jobs are less likely to provide healthcare as a benefit for their employees, leading to a great financial burden being placed on these citizens already dealing with student debt and entry-level wages (Mishel, Lawrence, et al., 2015).

Housing is a hugely relevant and commonly discussed issue today - and it has been shown that the growth in housing affordability is severe, and thus many cannot afford to rent or own the same housing as one could one or two generations ago without sharing the space or saving for multiple decades. However, in comparison to the rest of the expenditures, rent and

housing, pictured in Figure 4, only grew by around 100 and 80 percent, respectively (“Historical Census”, 2021). Although this may seem less significant in comparison to the high rises in tuition fees and healthcare costs, this is still a huge growth. Since all of these index values are based on constant dollar values, this shows us that the burden of these expenditures on the wage has nearly doubled in comparison to the portion of the wage housing costs in 1980, reflected by the decline in homeownership and the rise of homeowner ages (Meyer, 2024). Housing is, of course, necessary for life, and these growing costs only further sap the life and potential of wealth accumulation from modern workers.

Houses and apartments are becoming less affordable by a considerable factor - rent could reach around 23 percent higher based on the model than what is currently the median rent in America: \$1,837 (Statista Research Department, 2023). Housing, around \$420,800 in 2024, could grow around 11 percent (“Historical Census”, 2021). Housing and rent in recent years have grown more slowly than the rest of the expenditures we have analyzed, with house prices being slower than rent. However, due to the nature of how expensive rent and house prices are, these percent increases reflect huge differences on top of already nearly unaffordable costs. To be noted, as can be seen in Figure 3, is that rent’s regression line is lower than the actual rent prices from 2020 onwards; the same is the case for housing from 2021 onwards. Due to the recurring spikes and falls of both housing prices and rent, the linear regression models can be less accurate in predicting volatile data like this for short periods of time, but the overall growth over long periods is roughly similar.

Student debt, as mentioned earlier, is huge when considering the financial stresses placed upon the American citizens of today - college tuition has been growing extremely quickly, especially so in more recent years starting in 2020. The overall student debt of today is 1.7 trillion dollars, only beaten by mortgage debt at 12 trillion dollars - which should show how radically financially burdened American citizens are by these 2 large expenditures. Tuition fees have risen to be around 3.2 times the amount spent in constant dollars in 1980, or around 13 times the amount spent directly from 1980. These exorbitant prices for tuition are weighing down heavier than ever on today’s students, and these fees are essentially a rite of passage in society - it’s difficult to find a stable career and income without a bachelor’s degree or higher, and it’s been shown that jobs pay higher when one has a higher education. However, even with wage increases, the build-up of debt is hard to keep up with as interest on a higher price makes the debt grow faster, and with the high starting amount, it’s likely one would be unable to finish the payment quickly enough to avoid being saddled with long term financial detriments.

Utilities are by far the most volatile and least predictable of the three sorted categories discussed. Electricity, almost unsurprisingly, actually fell in price by a significant amount. Out of all expenditures indexed and compared, electricity prices have become more accessible for Americans - and it’s easy to see that truth in everyday life. Electricity has become a necessity for urban environments and is used everywhere; one can extrapolate that electricity efficiency in collection and distribution has played a role in this 16 percent decrease. Taking the regression, we can see around a 7.1 percent decrease from today - however, in reality, this means only a

one-cent difference in dollars per kWh (U.S. Bureau of Labor Statistics, 2024). Due to the fact that we are comparing from 1980, it's uncertain if the innovations in electricity efficiency from those times will continue at the same speed as the innovative speed from today to 2034.

As pictured in Figure 4, gasoline prices spike and fall to extreme degrees and have been for the past forty-four years (U.S. Bureau of Labor Statistics, 2024). Thus, it's quite difficult to get an accurate gauge of the future short-term growth. However, it's still evident that over the past four decades, the costs have still soared to much higher levels of cost, even when accounting for inflation. At 2.98 times their 1980 price, gasoline grew faster than housing and rent, but due to the nature of gasoline not taking up a huge portion of real expenditures, this fast growth hasn't affected Americans the same way tuition, healthcare, and housing have. If one takes the large picture as an indicator of a prediction of gas prices, then we can see around a predicted 14.7 percent increase from today's prices. However, since 2020, gas prices have spiked at an incredible rate - hence the second regression line, pointing instead at a predicted 46.8 percent increase. However, as seen on the same figure and mentioned before, spikes and falls happen quite often, so it is not with absolute certainty that the paper makes this prediction - it's quite likely we see another fall in the near future, similar to 2010-2012.

Groceries, even more necessary than gasoline, however, are a different story from gasoline. Its slope has remained relatively constant, showing consistent growth and reaching 2.93 times its 1980 price (Federal Reserve Economic Data, 2024). Groceries have alarmingly grown at a speed close to gasoline, all the while being much more expensive. Thanks to the consistent growth rate, the prediction of a 15% total increase in monthly expenditure seems to be more accurate. However, after 2020, likely due to the COVID-19 recession, groceries have risen at an even faster speed. This small spike was not as pronounced as the housing and gasoline spikes, and thus this paper did not create another regression for what could end up being inconsequential.

Finally, income has been shown to be dwarfed by almost every other expenditure - only electricity fell below, and house prices were closest in total growth. However, despite only growing to only 1.4 times the 1980 mean salary, this does reflect a difference of about 15,000 constant 2024 dollars (Federal Reserve Economic Data, 2024). However, when faced with consistent expenditure growth at levels far higher than the mean income growth, the leeway for comfort, financial freedom and mobility, and ease of access to high-quality goods and services is shrinking quickly. The slow rate reflects a lower prediction in the linear regression, only rising 7.1 percent - around 4,500 dollars - to \$64,572.

The rate at which costs have grown, especially when looking directly at the comparatively slower growth of the mean income, is incredibly concerning and could potentially lead to great poverty rates, lowered quality of life until after many more (in comparison to 1 and 2 generations ago) years working, and/or a stagnant middle class and growing lower class.

Discussion

A focus on affordability is necessary for government policy to curb these growth rates and allow for the slow catch-up of wages. The cost of living is a serious concern that has accelerated due to inflationary shocks from global supply disruptions. However, prevention with caps of key goods and targeting corporate “greedflation” may help to alleviate this issue (Janssen, 2023). While expenditures and wages for the middle-class stall, corporations and CEOs grow ever richer and gain more control, which allows them to push profits with no real repercussions (Mishel, Lawrence, et al., 2015). Implementing anti-monopoly measures and taxing those with excessive net worths to a greater degree could not only prove to create a felt increase in wages due to lowered costs but also help in creating more infrastructure and policies to benefit the masses of America, like lowered tuition costs and more affordable housing, as will be covered. Additionally, windfall taxes on corporate profits, which have already been shown to be productive in the United Kingdom and Spain, and fighting excessive corporate profit-making and pushing the wealth towards policies and functions that serve the majority of the public could help redistribute the earnings back to workers (Janssen, 2023).

With the current housing crisis, a lack of supply, and constantly increasing demand, it’s recommended that the government both construct more affordable housing and enact policies that stop landlords and other multiple property owners from holding onto so many apartments and houses (Furth, 2015). It’s been shown that the lowered amount of home-owners among millennials (only 18% of homebuyers and only 60% of older millennials at ages 40-42 own homes while 73% of the Silent Generation and 68% of Baby Boomers did at that age) is interconnected with other costs of living - alongside inflated house prices, the burden of student debt prevents 63% of millennials from saving for a down payment for a home (Meyer, 2024). A simple cap to their growth is hard to imagine, so the more elegant solution would be to increase supply and thereby reduce the amount that prices are allowed to rise due to more competition.

In healthcare and tuition, these industries have become almost entirely privatized into corporations with such quickly generated money. In this case, a hard government-enforced cap and reduction of these high prices, as well as a reduction in student debt through loan forgiveness, would largely help the people. In the past, drugmaker company Eli Lilly capped the cost of insulin at \$35 a month in the middle of the COVID-19 pandemic and were able to support themselves without real losses, showing these companies do have the capacity to perform these caps to save lives, yet choose not to (Ricks, 2020). College affordability and student debt is a significant financial issue that affects many Americans, and the government can step in with a tuition cap to help solve this pressing problem (Shireman and Fast, 2023). Without any real incentive to control tuition, private colleges can set their barrier to entry however high they want and force the class disparity and student debt issue to grow worse for the sake of profits. Caps are a similar solution to gasoline spikes - in Germany, a price cap was implemented in the natural gas changes sparked by the Russo-Ukrainian War, having a deflationary effect in an effort to protect households and businesses from soaring costs (Wehrmann, 2023). Electricity, although not currently a large issue, could follow the same path. As for groceries, it’s a difficult situation -

Americans see food prices as their top economic concern (Strachan, 2024), but with the decrease in gas prices comes a decrease in transportation costs that plays a large role in the grocery prices. We see that the U.S. is lacking protections for farmers and grocery stores, which allows large monopoly corporations like Nestle or PepsiCo to institute higher pricing and thus drive grocery costs up as they provide many of the products Americans consume.

Overall, caps and reductions in key items have, in the past, shown useful and productive in slowing inflation and rising household costs. Alongside proper fund diverting and increased wealth taxes, the American government can slow and prevent the potentially disastrous future of a lack of economic mobility and lowered ease of access to the same comparative quality of life past generations have had.

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Advances and Challenges in Antibody-Drug Conjugates For Cancer Therapy: A Focus On Conjugation Methods By Aneesh Mamidi

Abstract

Over the last few years, the field of Antibody-drug conjugates (ADCs) has undergone remarkable changes, and conjugation methods have been central to this development. While lysine and cysteine conjugations have been traditionally used to link cytotoxic payloads to antibodies, novel forms of conjugation are emerging. Aiming to address the limitations of conventional methods, these novel strategies, including the incorporation of non-natural amino acids and the introduction of SMARTag® technology, aim to improve the overall selectivity, efficacy, and safety of ADCs for cancer treatment. In this review, we examine these recent advancements in conjugation methods and their implications for the future of the field.

1. Definition and General Purpose

For thousands of years, drugs have played a crucially important role in the development of medical science and human health: from the use of natural remedies in ancient civilizations to the discovery of penicillin in the 20th century, pharmacotherapy has marked a significant impact on healthcare. Furthermore, the advent of biotechnology supplemented this expansion in medicine, introducing biologics and personalized treatments.

Now, in the 21st century, the integration of biotechnology and pharmacology has led to novel therapies, particularly ADCs. ADCs combine the specificity of antibodies with the potent cytotoxic effects of drugs, primarily to target rapidly proliferating cancer cells, minimize collateral damage to healthy tissues, and deliver drugs directly to the tumor. This targeted approach lowers the minimum effective dose (MED) required, offering significant advantages over traditional chemotherapy (Maecker et al.). Over the past six years, the field of ADCs has grown remarkably, with 11 FDA-approved ADCs, including the recent approval of Elahere in November 2022 for ovarian cancer (Gogia et al.).

Currently, there are more than 300 ADCs that have reached clinical development. This growth is met with an increase in clinical trials, financial investments from major pharmaceutical companies, and scientific interest in challenging diseases pointing to a bright future for ADCs in cancer therapeutics.

2. Core Components of ADCs

The design of ADCs involves four core components: the antibody, the attachment site, the linker, and the payload. The attachment site ensures precise conjugation of the drug-linker to the antibody, the linker is essential for maintaining stability during circulation, and the payload is the cytotoxic therapeutic agent that induces cell death upon release (Maecker et al.) see Fig 1 below.

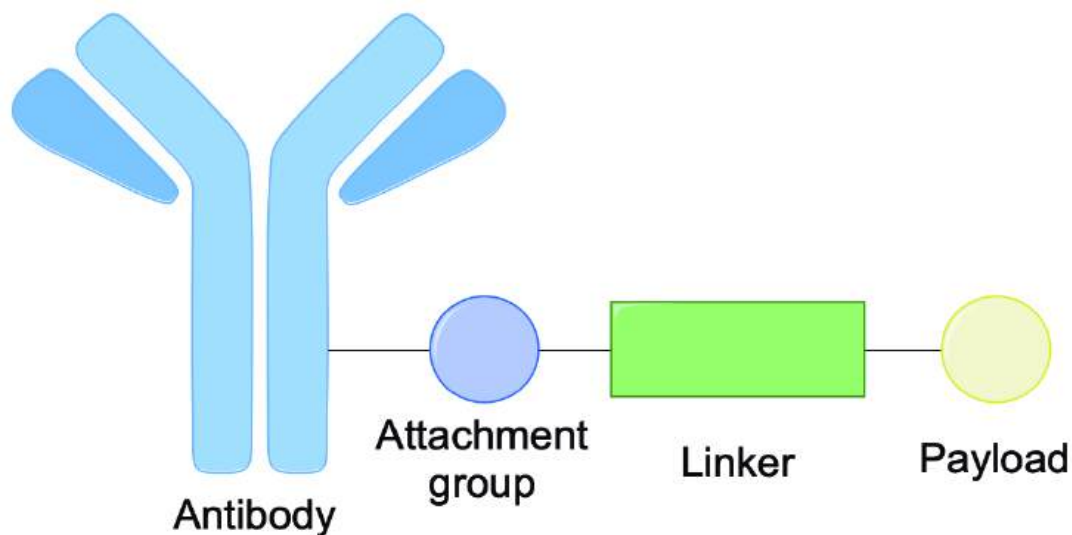


Fig 1. The four core components of ADCs.

3. Mechanism of Action

All the core components of ADCs work together to execute the delivery of the cytotoxic payload to the target cell. As mentioned earlier, ADCs combine the specificity of monoclonal antibodies with the potent cytotoxicity of small-molecule drugs: the Antibody recognizes the target cancer cell through the specific antigen expressed on the cell's surface and binds to it. When binding to the target antigen, the ADC complex is internalized into the cancer cell through receptor-mediated endocytosis. Once internalized, the ADC complex either can be recycled back to the cell's surface or can undergo lysosomal degradation, digested by lysosomal enzymes such as hydrolytic enzymes, which cleaves the linker connecting the antibody to the cytotoxic drug. Moreover, this cleavage, or release, can occur through various mechanisms, such as acidic pH conditions, enzymatic cleavage, or reduction, depending on the nature of the linker used in the ADC design and the tumor environment.

Once the linker is cleaved, the cytotoxic payload is released and triggers cell death. As mentioned earlier the mechanism by which cell death is induced varies based on the type of payload used. Commonly, the cytotoxic payload interferes with critical cellular processes including binding to tubulin, disrupting microtubule dynamics, inhibiting topoisomerase, or inducing DNA crosslinking. These actions result in cell cycle arrest and apoptosis, leading to the selective killing of cancer cells. The specificity of the antibody ensures that the cytotoxic drug is delivered predominantly to cancer cells, sparing normal, healthy tissues and reducing the adverse effects typically associated with conventional chemotherapy(Maecker et al.).

4. Current Clinical and Therapeutic Applications

While ADCs are mostly known for cancer therapy, their use extends to a wide range of applications: as mentioned earlier, over 300 ADCs are in various stages of clinical trials (Maecker et al.), targeting not only solid tumors but also conditions like autoimmune diseases.

The FDA has approved 11 ADCs, including notable examples like brentuximab vedotin for Hodgkin lymphoma and trastuzumab emtansine for HER2-positive breast cancer, highlighting their efficacy and potential (Tong et al.). Furthermore, in clinical trials, ADCs have also shown promising results, with some achieving response rates of up to 80% in certain cancers (W. Q. Li et al.). However, it is important to note that while there exist ADCs that have achieved high efficacy, there have been several examples of discontinued ADCs. In the 26 years of the development of this field, over 1200 clinical trials were conducted. However, there have been over 54 ADC programs formally discontinued and 38 ADCs removed from company pipelines (Maecker et al.).

5. Importance of Conjugation Methods

The field of antibody-drug conjugates is studied thoroughly; however, conjugation methods often need to be more studied. While the antibody, payload, and linker components are extensively researched, there is comparatively little literature on conjugation chemistry. This oversight is significant because the method of conjugation at the attachment site can influence the ADC's overall efficacy, stability, and safety.

Conjugation methods determine how the drug is linked to the antibody at specific attachment sites, influencing the pharmacokinetics (PK) of the ADC. A stable conjugation at the attachment site ensures that the complex remains intact in the bloodstream until it reaches its target cell, preventing premature release of the payload and reducing off-target cytotoxic effects. On the other hand, an unstable conjugation or poorly chosen attachment site can result in premature drug release, leading to reduced therapeutic outcomes. This instability not only lowers the efficacy of the ADC by reducing the amount of drug delivered to cancer cells but also poses significant safety risks, as the cytotoxic payload can affect healthy tissues and cause adverse side effects (Maecker et al.).

Conjugation chemistry of ADCs encompasses several critical concepts, including the drug-to-antibody ratio (DAR) and site-specificity. The DAR refers to the number of drug molecules attached to each antibody or rather the number of drug attachment sites, and it is a crucial factor that impacts both the efficacy and safety of ADCs. Furthermore, conjugation chemistry can be categorized into site-specific and non-site-specific attachments. Site-specific attachment involves linking the drug to predetermined sites on the antibody. On the other hand, Non-site-specific attachment results in a more heterogeneous mixture, which can lead to variability in the therapeutic and safety profiles of the ADC. To comprehensively understand the importance of attachment groups, the concepts of DAR, site-specificity, and homogeneity will be explored in greater detail in subsequent sections of this paper.

In the current clinical landscape of ADCs, several conjugation methods are being utilized, each with distinct implications for the efficacy and stability of the ADC molecule. Currently, the most popular used chemical methodology for the conjugation of drugs to monoclonal antibodies involves either cysteine or lysine residues. Specifically, a widely used conjugation involving such cysteine residues is cysteine-maleimide. Likewise, there exist many forms of lysine

conjugations with more being developed.

However, the field of conjugation chemistry is not just limited to cysteine and lysine residues. In addition to these traditional methods, novel conjugation techniques are emerging, aiming to address the limitations of conventional approaches. These novel conjugation techniques are designed to enhance specificity, stability, and overall efficacy, marking a significant advancement in the field of antibody-drug conjugates.

This paper aims to focus on conjugation techniques, evaluating each method's pros and cons, exploring potential improvements, and introducing emerging methods. By providing a thorough analysis, this paper seeks to offer valuable insights to ADC developers, ultimately aiding in the enhancement of ADC design and efficacy for better clinical outcomes.

6. Evaluation of the Current Clinical Landscape

To evaluate the current clinical landscape, both FDA-approved drugs as well as non-FDA-approved drugs were chosen for analysis. For the non-FDA-approved drugs, ADCs from clinical trials over the last 10 years on clinicaltrials.gov were examined. The selection of the 10-year period is due to the fact that many novel conjugation techniques have been developed and refined over the period. Therefore, including this timeframe ensures that these innovative approaches are adequately represented in the analysis.

Of the 11 FDA-approved ADCs, 4 of them (36.36%) utilize lysine conjugation, with all of them being non-site-specific attachments, and 7 of them utilize Cysteine-maleimide conjugation (63.63%), out of which 2 utilize site-specific attachment and 5 utilize non-site-specific attachment. Note that Lumoxiti has been excluded from the list as we consider it an immunotoxin.

Of the 52 clinically active ADCs (part of clinical trials in various phases), 34 ADCs utilize Cysteine-maleimide conjugation (65.38%), of which 24 have undisclosed site specificity, 8 have site-specific conjugation, and 2 have Non-site-specific cysteine-maleimide conjugation. Furthermore, of these 52 ADCs, 9 ADCs present lysine conjugation (17.30%), with 7 having undisclosed site specificity and 2 having site-specific conjugation. Lastly, 4 ADCs present novel conjugation methods (methods that have never been utilized before), and 5 ADCs have undisclosed conjugation methods. This information about the conjugation methods of the current clinical landscape raises some important trends regarding the direction of development of the field. These trends will be analyzed and evaluated in later sections of the paper.

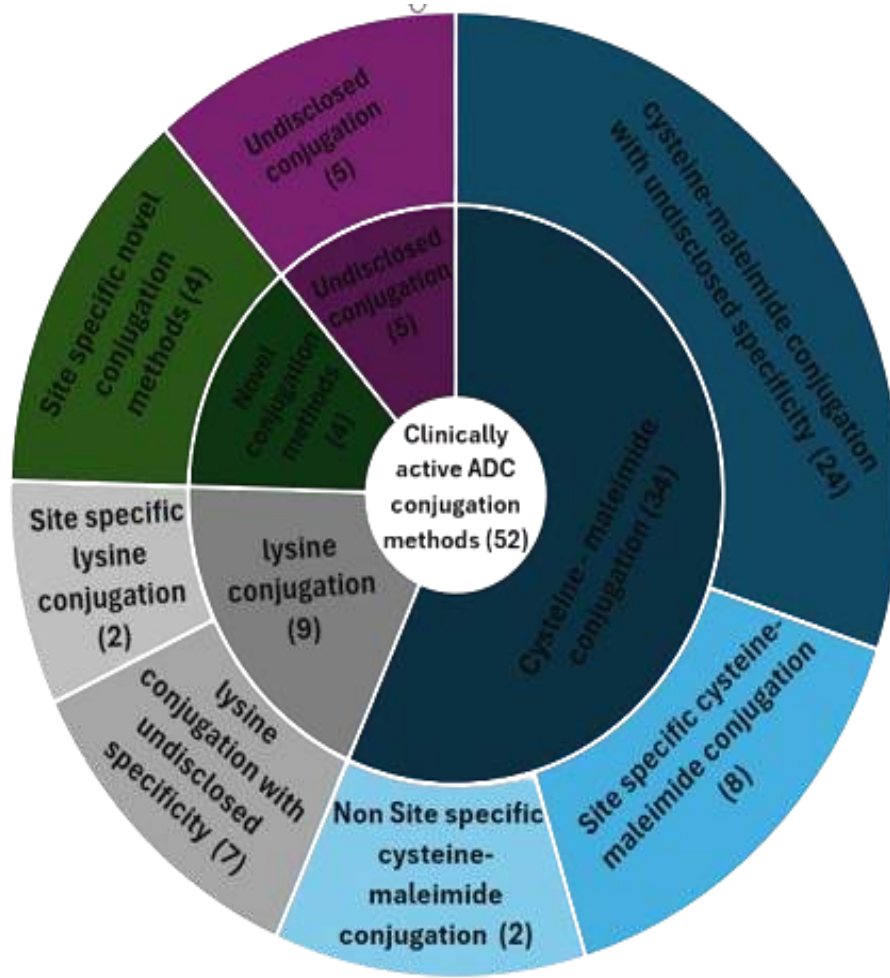


Fig 2. Conjugation Methods used in clinically active ADCs over the past 10 years that are listed on clinicaltrials.gov. Numbers of ADCs utilizing different attachment groups are shown in the outer ring.

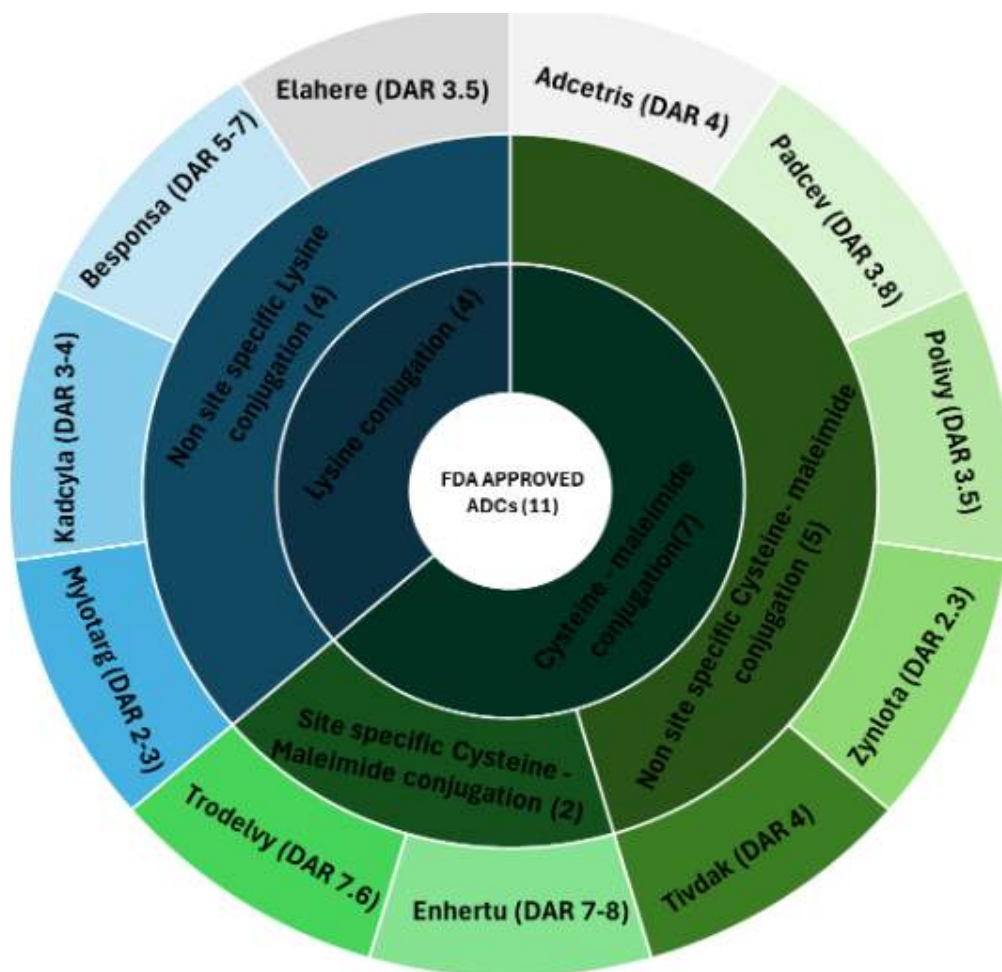


Fig 3. Conjugation Methods Used in the 11 FDA-approved ADCs. Names of ADCs utilizing different conjugation methods along with their site specificity are shown in the outer ring.

7. Concepts in Attachment Chemistry

7.1 DAR(Drug to antibody ratio)

As mentioned before, DAR refers to the average number of drugs linked to each antibody. A higher DAR can increase the potency of the ADC by delivering more cytotoxic payloads to the cancer cells, however excessively high DARs may interfere with antibody structure, stability, and antigen binding, therefore lowering ADC activity. Conversely, low DARs might result in an insufficient amount of drug being delivered to the cancer cells. Therefore, optimizing DAR is crucial for an effective ADC design. However, while many techniques are being developed, controlling DAR proves to be a challenge due to the complex chemical and physical requirements. Currently, several analytical techniques are being developed for DAR measurement, including UV/Vis Spectroscopy and hydrophobic interaction chromatography(Tang et al.). Challenges in accurate monitoring of DAR often result in varied DARs per ADC molecule, hence ADC manufacturers often report a range. For example, below,

is a list of FDA-approved ADCs(11), categorized by type of conjugation, with their average DAR values listed.

FDA APPROVED ADCs (11)			
	Non-site-specific lysine (4)	site-specific cysteine-maleimide (2)	non-site -specific cysteine-maleimide (5)
1	Mylotarg (DAR 2–3)	Enhertu(DAR 7–8)	Adcetris (DAR 4)
2	Kadcyla (DAR 3–4)	Trodelvy (DAR 7.6)	Padcev (DAR 3.8)
3	Besponsa (DAR 5–7)		Polivy (DAR 3.5)
4	Elahere (DAR 3.5)		Zynlota (DAR 2.3)
5			Tivdak (DAR 4)

Table 1. The 11 FDA-approved ADCs with their DAR values.

7.2 Site Specificity

Site specificity in ADC attachment chemistry refers to the precision with which the cytotoxic drug is linked to specific sites on the antibody. This can be categorized into site-specific and non-site-specific attachment methods. Site-specific attachment involves conjugating the drug to predetermined sites on the antibody, such as engineered cysteine residues (non-naturally occurring). This method of conjugation allows for a more homogenous product, with each molecule being uniform and having a consistent DAR, which as mentioned earlier has significant implications for efficacy (Junutula et al.). Moreover, this allows precise control over the stoichiometry of the drug conjugation. On the other hand, non-site-specific attachment involves conjugating the drug's naturally occurring amino acids such as lysine residues or cysteine residues. This conjugation is random due to the abundant distribution of such naturally occurring amino acids. This leads to more heterogenous molecules, with varied DARs.(Junutula et al.)

Just like other critical parameters, the site specificity of an ADC has various implications. Firstly, while site-specific conjugation results in a more homogenous product, allowing for more predictable pharmacodynamics, achieving it requires advanced engineering which can be time-consuming and resource-intensive. On the other hand, while non-site-specific conjugation can introduce variability in clinical outcomes, it is generally simpler and less expensive to execute, making it more accessible for early-stage research and development. Each approach has its advantages and disadvantages and should be selected depending on factors such as therapeutic target and desired clinical outcomes. Future research aims to optimize both methods of conjugation, continuing to advance the safety of ADCs.

8. Conjugation Methods and their Implications

8.1. Cysteine-Maleimide

In general, any conjugation that uses a maleimide attached directly to an antibody is referred to as a cysteine-maleimide conjugation. This type of conjugation is widely used in the field of ADC due to its synthetic accessibility, and excellent reactivity (Ravasco et al.). Moreover, cysteines have been one of the first choices among ADC manufacturers for site-selective modification. As of today, the relevance of this conjugation in the field of ADCs is greatly significant, with notable examples being Brentuximab vedotin and trastuzumab emtansine.

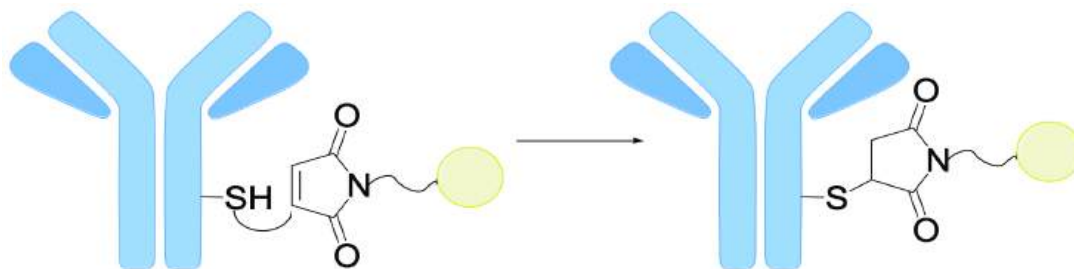


Fig 4. Cysteine-maleimide conjugation

Mechanism of conjugation

The core principle of this conjugation involves the formation of a thioether bond between the cysteine residues on the antibody and a maleimide attached to a payload. However, this bond formation has various structural implications in relation to the stability of ADCs: thiosuccinimide linkages are now thought to be less optimal than once believed (Ravasco et al.), impacting the pharmacodynamics and pharmacokinetics of ADCs. The linkage formed is a result of Michael's addition of cysteine to the double bond of a maleimide.

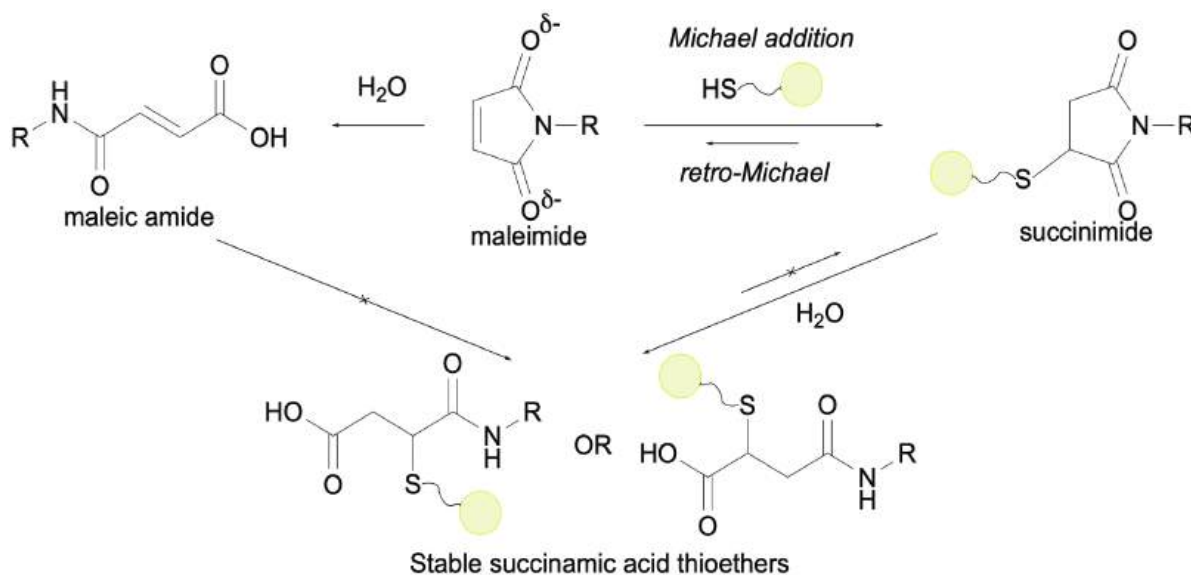


Fig 5. Pathway of cysteine-maleimide synthesis

The Michael addition reaction occurs when a nucleophile attacks an electrophilic α,β -unsaturated carbonyl compound. In the context of ADCs, the thiol group of cysteine residues on antibodies serves as the nucleophile. When the maleimide-functionalized payload is introduced, the thiol group of the cysteine residue attacks the double bond of the maleimide, resulting in the formation of a stable thioether bond. This addition reaction is typically done under a controlled environment, for example, a specific pH (Ravasco et al.) to maintain the integrity of the antibody and the drug, as these are often pH sensitive.

Structural implications

However, the stability of this thioether bond is compromised due to the retro-Michael reaction, which occurs under certain physiological conditions (Wu). Pharmacologically, this has various implications with one of the most notable being premature thiosuccinimide cleavage, resulting in the payload having decreased cytotoxic activity and off-target cytotoxicity.

Therefore, this type of conjugation has various implications. Structurally, the site-specific attachment of drugs to cysteine residues favors a uniform DAR, which is crucial for maintaining the consistency of therapeutic efficacy and safety. However, non-site-specific cysteine-maleimide conjugation, while simpler and more scalable, leads to heterogeneous ADC populations with drugs attaching to various cysteine residues.

Overall, the stability of this thiosuccinimide linkage has been subject to scrutiny. Due to its high sensitivity to pH conditions, this conjugation may not be appropriate for all regions of the body. Furthermore, it is important to note that this reaction typically is difficult to achieve full conversion. This commonly results in excess reagents being added, but often results in over-modification (Ravasco et al.).

Improvements in the conjugation method

To address the limitations of cysteine-maleimide conjugation, the field is rapidly advancing and developing new technologies aimed at improving the therapeutic index, reducing toxicity, and enhancing pharmacokinetics.

a) thiosuccinimide hydrolysis

As mentioned earlier, maleimides are more reactive than other typical Michael acceptors, leading to maleimides being susceptible to the retro-Michael reaction. A method to mitigate this is hydrolysis. Hydrolysis allows for the succinimide thioether bond to be much more stable, resisting retro-Michael reactions due to its high energetic cost. This stability is crucial for ensuring that the ADCs remain intact in the bloodstream until they reach the target cells. Moreover, this increased stability can lead to a more predictable therapeutic outcome, as the drug payload is released in a controlled manner at the target site.

The idea of hydrolysis is rapidly growing in the industry: one notable ADC that utilizes this technology is the emerging antibody-drug conjugate, ABBV-3373 (Buttgereit et al.).

b) Maleimide scaffolding

As seen through hydrolysis, Addressing the retro-Michael reaction can be crucial in enhancing therapeutic effect and reducing off-target toxicity. The idea of hydrolysis has influenced ADC manufacturers to further research ways to mitigate this retro-Michael reaction, and one of the ways is through maleimide scaffolds. As of now, there exist many different novel maleimide scaffolds with more under development.

i) N-Aryl Maleimides: N-Aryl maleimides are designed to be more resistant to the retro-Michael reaction. By attaching an Aryl group to the nitrogen of the maleimide, the resulting conjugate exhibits greater stability. This is due to the electron-donating effect of the Aryl group, which stabilizes the thioether bond. (Ravasco et al.)

ii) Exocyclic maleimides: These are maleimides that feature a five-membered ring structure that locks the maleimide in a configuration that is resistant to retro-Michael reactions. This type of maleimide scaffold ensures that the thioether bond remains intact even with competing nucleophiles. Therefore, the rigidity of these exocyclic maleimides prevents the retro-Michael reaction(Ravasco et al.)

In conclusion, cysteine-maleimide conjugation is vital in ADC development. While this conjugation offers several challenges to maintaining stability during circulation, it offers several advantages in terms of site-selective modification, cost, and reproducibility. Moreover, this conjugation is undergoing significant development, with various structural modifications to address its shortcomings.

8.2 Lysine Conjugation

Another widely used conjugation approach is lysine conjugation, which typically involves the formation of an amide bond. As of today, this conjugation is rapidly growing in popularity, with 36.36% out of the 11 FDA-approved ADCs utilizing this technology, with notable examples being Mylotarg for the treatment of acute myeloid leukemia, and Besponsa for acute lymphoblastic leukemia, see Fig 3. This conjugation method is favored for its extreme simplicity in manufacturing due to its well-established chemistry. However, this type of conjugation, just like cysteine-maleimide, also presents several unique challenges that influence ADC efficacy and safety.

Mechanism of conjugation

Lysine residues are naturally existing amino acids on antibodies for linking substrates. However, unlike cysteine residues, lysine residues exist in great abundance. These lysine residues contain amino groups (-NH₂) that can react to form various conjugations. The general mechanism of conjugation occurs when an activated drug reacts with the amino groups of lysine residues, forming a stable bond(Haque et al.).

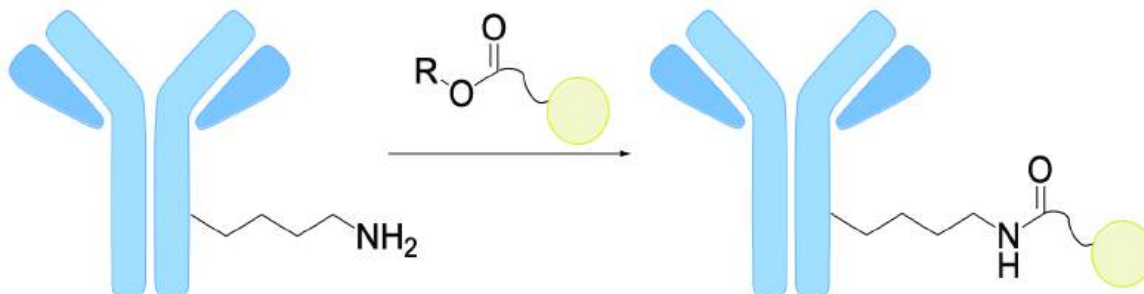


Fig 6. Lysine conjugation

Structural implications

A critical implication of this method of conjugation is that a high drug-antibody ratio is achieved due to the abundance of lysine residues typically present. As mentioned earlier, this high DAR has several positive and negative implications. One major implication is a lack of control of homogeneity. The abundance of lysine residues leads to heterogeneous ADC populations, with varying DARs and inconsistent pharmacodynamics. This variability can impact the therapeutic efficacy and safety of the ADC. However, what is more significant is the fact that this high drug loading results in decreased control of the exact location of drug attachment.

Therefore, site selectivity is often a parameter that presents a challenge. The lack of site-specificity may introduce potential difficulties regarding the antibody's binding affinity and overall stability. Despite the potential challenges with a high DAR, a high DAR may also be favorable due to increasing therapeutic potency by delivering more copies of the cytotoxic drug (Chudasama et al.).

Improvement in conjugation technique

a) Enzyme-directed modification

As mentioned earlier, due to the natural abundance of lysine residues on antibodies, one of the primary challenges of lysine conjugation is achieving site selectivity. One of the methods to address this problem involves enzyme-directed modification (Haque et al.). Enzymes can provide excellent selectivity under suitable biological environments, specifically pH and temperature, due to their highly specific active sites and binding sites. Currently, various enzymes are being investigated in the use of bioconjugation.

i) Sortase A (SrtA) (Haque et al.): Sortase A from *Staphylococcus aureus* is an enzyme that catalyzes the covalent ligation between amino acids via an isopeptide bond. This enzyme can recognize the LPZTG sequence, cleaving the bond between threonine and glycine, and forming a thioester intermediate. This thioester intermediate can be substituted by an amino group, forming an amide linkage. Moreover, this enzyme has been clinically tested, with Chilkoti and colleagues demonstrating that SrtA could achieve site-specific lysine modification on the anti-HER2 antibody 4D5, achieving a high conversion rate and homogeneity in the conjugate (Bellucci et al.).

ii) Lysine Acylation Using Conjugation Enzymes (LACE): This approach involves

combining Sortase A and Ubc9, a small ubiquitin-like modifier (SUMO)-conjugating enzyme, for dual modification. Ubc9 targets the IKXE sequence, while Sortase A recognizes LPETGG. This method was reported to achieve high site selectivity and efficiency, demonstrating 95% conversion in short time frames (Haque et al.). This LACE method allows for precise modification of lysine residues, retaining the antibody's binding affinity and enabling dual modifications with complementary drugs.

b) Covalent tethering (Haque et al.)

This method is a form of Proximity-Induced Site-Selective Lysine Modification and involves a temporary covalent attachment to a nearby site, followed by modification of the target lysine residue. Covalent tethers enable an initial, often reversible, interaction on a temporary binding site, which positions another reactive group closer to the target modification site. Moreover, chemical reagents can be designed to exploit differences in amino acid sidechain reactivities under varying conditions, such as pH, enhancing site selectivity. One notable practice utilizing covalent tethering is Native chemical ligation (NCL), which uses a thioester-mediated chemo-selective reaction that leverages the varying reactivities of amino acids to fine-tune the selectivity of modification sites, ensuring efficient and targeted protein synthesis.

Lysine conjugation is crucial in ADC development, offering broad applicability due to lysine's abundance in proteins. Despite challenges in site selectivity and potential heterogeneity, it provides advantages in stability and ease of modification.

c) thiosuccinimide hydrolysis-SMCC

One particular improvement involves the application of succinimide hydrolysis, which is traditionally associated with cysteine-maleimide conjugation, to lysine-based conjugation, particularly when using the SMCC linker.

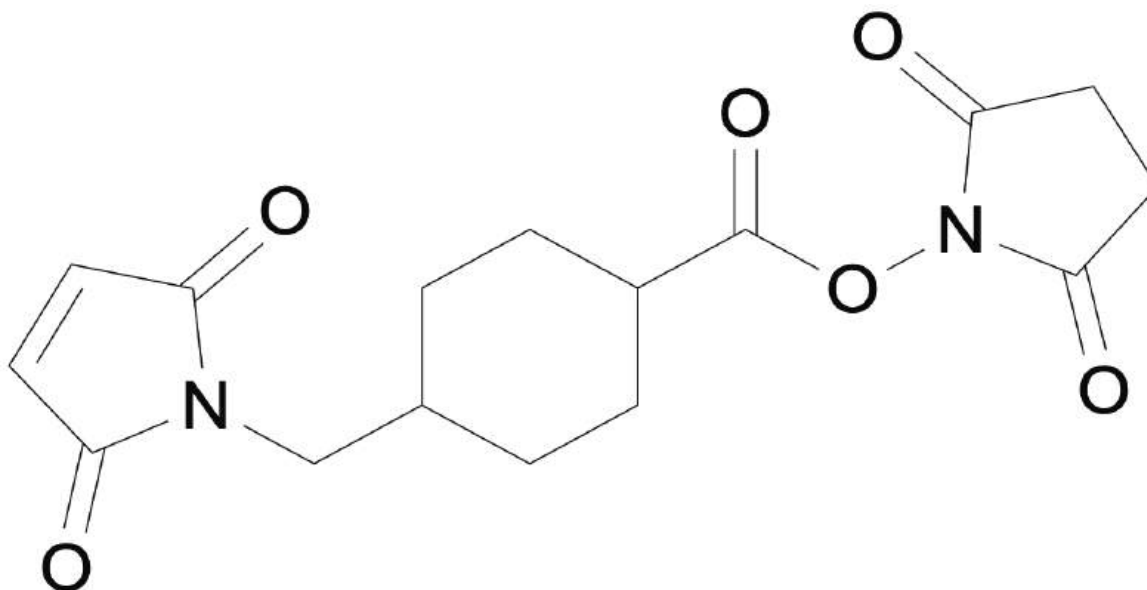


Fig 7. SMCC linker.

The SMCC linker is a common linker used in antibody-drug conjugates (ADCs). It contains two reactive groups: a maleimide group and an N-hydroxysuccinimide (NHS) ester. The NHS ester reacts with primary amines, such as those in lysine residues. At the same time, the maleimide group is normally used for covalent addition to a thiol group of a drug, allowing the SMCC linker to attach drugs to antibodies through these sites. However, many ADCs that utilize this linker technology are unstable as the thiosuccinimide linkage formed is subject to the retro-Michael reaction.

To address the instability issues associated with the SMCC linker, advancements have been made to improve the stability of the resulting ADCs. One such improvement is the use of succinimide hydrolysis, which converts the maleimide-thiol adduct into a more stable form by hydrolyzing the thiosuccinimide linkage (Ravasco et al.). This modification significantly reduces the occurrence of the retro-Michael reaction, thereby enhancing the stability of the conjugate. GQ1001, a next-generation HER2-targeting ADC, effectively incorporates this approach, demonstrating improved stability and efficacy in preclinical studies. GQ1001 is only one of many notable examples of the rapid increase in developments and relevance of the lysine conjugation (Wang et al.).

As of today, 4 FDA-approved ADCs utilize lysine conjugation, of which all of them utilize non-site-specific lysine conjugation. Figure 3 depicts the conjugation methods of clinically active ADCs over the last 10 years, and shows that in these 10 years, there have been no ADCs utilizing non-site-specific lysine conjugation in clinical trials. Instead, ADCs utilizing site-specific lysine conjugation have been emerging, with 100% of the clinically active lysine-conjugated ADCs over the past 10 years with disclosed conjugation being site-specific. This shift in site selectivity marks the rapid pace of development in conjugation chemistry with the transition being driven by such improvements, including enzymatic-directed modification and thiosuccinimide hydrolysis, all aiming for higher precision and consistency in drug delivery.

9. ADCs with Novel Conjugation methods

While lysine and cysteine-maleimide conjugations are well known in the realm of conjugation chemistry, ADCs with original and unconventional conjugation methods are emerging at a rapid pace. These innovative approaches aim to overcome limitations associated with traditional techniques, enhancing selectivity, stability, and efficacy. Out of the 52 ADCs active in clinical trials over the last 10 years, 4 ADCs with unconventional conjugation techniques have been identified, see Fig 2. While these ADCs have not been FDA-approved, they pave the way for future advancements.

9.1. ARX788: pAF-hydroxylamine-PEG4

ARX788 is a cutting-edge ADC that targets HER2-positive cancers, utilizing a unique site-specific conjugation method that involves the incorporation of para-acetylphenylalanine (pAF), a non-natural amino acid. This advanced approach is designed to overcome the limitations of traditional conjugation techniques, providing enhanced stability and

efficacy(Skidmore et al.).

This novel conjugation involves site-specific attachment of drugs to an antibody through a stable oxime bond, which is formed by conjugating hydroxylamine-containing drug-linker to the non-natural amino acid para-acetylphenylalanine (pAF), which is strategically incorporated into the antibody. In this process, pAF is incorporated at two distinct, predetermined sites within the antibody structure, allowing for precise attachment of the drug-linker. Since pAF is introduced at these specific locations, the hydroxylamine-containing drug-linkers can form stable oxime bonds with the pAF residues, resulting in the attachment of two drug-linkers to the antibody. This site-specific approach ensures that the drug-to-antibody ratio (DAR) is controlled, typically resulting in a DAR of 2(Skidmore et al.).

One of the advantages of utilizing this conjugation technique is that it allows site-selective modification, resulting in a homogenous ADC with a consistent DAR. This uniformity is starkly contrasted to the heterogeneous mixtures of ADCs that result from traditional methods, where random attachment to the abundant lysine or cysteine residues can lead to variability in DARs.

Secondly, the stability of the oxime bond is a key feature of this technique, ensuring that the drug remains attached to the antibody during circulation and is only released upon reaching the target tumor cells. In fact, ARX788 exhibits high serum stability in mice and has shown a relatively long ADC half-life of 12.5 days. Additionally, Pharmacokinetic (PK) studies conducted on multiple species verified this highly stable nature(Skidmore et al.).

However, while this new technology seems to be promising pharmacologically, it too has its own shortcomings. Notably, the integration of non-natural amino acids like pAF requires advanced genetic engineering techniques, which can increase the complexity and cost of production. This could pose challenges for large-scale manufacturing and commercialization. Moreover, these implications can also extend beyond cost. Moreover, the oxime ligation requires acidic conditions (pH 4.5), which may introduce even more difficulties in the manufacturing process (Walsh et al.).

9.2 STRO-002 and CC-99712: para-AzidoMethyl-L-Phenylalanine

Similar to ARX788, there have been several other ADCs in clinical trials that have non-natural amino acids as a method of conjugation. Two of them are STRO-002 and CC-99712, which utilize the non-natural amino acid Para-AzidoMethyl-L-Phenylalanine(X. Li et al.). This approach utilizes bioorthogonal chemistry for site-specific drug attachment, leading to more homogeneous ADCs. However, it also comes with certain challenges and potential drawbacks.

The pAMF conjugation method involves incorporating pAMF into the antibody via genetic engineering. The azide group on pAMF reacts with an alkyne-functionalized linker through a strain-promoted azide-alkyne cycloaddition (SPAAC), a type of copper-free click chemistry. This reaction forms a stable triazole bond, ensuring precise drug attachment and maintaining the antibody's integrity(Adhikari et al.).

Using such non-natural amino acids, site selectivity is more achievable. The precise incorporation of pAMF allows for site-specific drug attachment, resulting in homogeneous ADCs with consistent drug-to-antibody ratios. Utilizing this conjugation, STRO-002 has been generated as a homogenous ADC with a DAR of 4(X. Li et al.). However, apart from homogeneity, the triazole bond formed through SPAAC offers great stability. For instance, through in vitro and in vivo stability studies, STRO-002 has demonstrated good stability in preclinical and clinical studies, with data showing prolonged stability and a reduction in premature drug release, In single-dose toxicology studies conducted on rats, SC209(STRO-002's metabolite) was well-tolerated at a maximum dose of 0.6 mg/kg(Solis et al.).

However, utilizing non-natural amino acids in therapeutic applications also introduces several challenges such as increased synthetic complexity, and higher production costs. The incorporation process requires specialized techniques, which complicate large-scale production(Walsh et al.).

9.3 TRPH-222: SMARTag

TRPH-222 is an innovative antibody-drug conjugate (ADC) developed to target CD22-positive B-cell malignancies, such as non-Hodgkin lymphoma. It employs the SMARTag® conjugation technology to achieve site-specific conjugation of the cytotoxic payload, maytansinoid, through a non-cleavable linker, significantly enhancing the stability and efficacy of the ADC.

SMARTag® conjugation is an enzyme-based, site-specific technique that leverages formylglycine-generating enzymes (FGE) to introduce the amino acid C α -formylglycine (FGly) into recombinant proteins post-translationally. This process begins with FGE recognizing a specific five-amino acid sequence—cysteine-x-proline-x-arginine—known as the "aldehyde tag", which is genetically incorporated into the specified site within the antibody's constant region. During protein synthesis, FGE binds to this sequence and enzymatically converts the cysteine residue into formylglycine by oxidizing its thiol group to an aldehyde. The resulting formylglycine residues provide highly precise sites for drug conjugation, ensuring consistent and site-specific attachment of therapeutic agents(Krüger et al.).

The FGE-recognized sequence can be placed anywhere in the sequence of the Antibody. This intentional posing of the sequence allows for modification of the Antibody in predetermined sites, allowing for site-specific conjugation. The amino acid formal glycine contains an aldehyde group that can be chemically coupled in site-specific methodology.

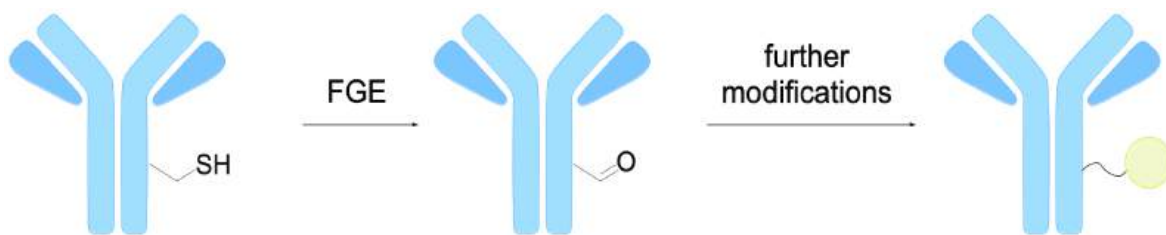


Fig 8. SMARTag® conjugation

This SMARTag® technology therefore enables precise site-specific conjugation, resulting in homogeneous ADCs with consistent DARs(Hernandez-Ilizaliturri et al.). Using this technology, TRPH-222 was generated with the tightly controlled DAR of 2.0. Moreover, in order to generate a higher drug loading, multiple tags can be incorporated into a single antibody.

Stability-wise, this form of conjugation also has its own implications. Proprietary biocompatible smart tag chemistry can be used to generate a stable attachment of toxins, including small molecule drugs. Studies have shown that TRPH-222 maintains stability and efficacy over extended periods, Once-weekly intravenous (IV) administration of TRPH-222 at doses of 1, 3, or 10 mg/kg resulted in a 51% reduction in tumor volume with the 1 mg/kg dose, and a 100% reduction in tumor volume with both the 3 mg/kg and 10 mg/kg doses after 28 days, making it suitable for long-term treatment (Maclaren et al.). While SMARTag® technology offers significant advancements in antibody-drug conjugates (ADCs), it also comes with notable downsides. The production process is expensive, involving engineered antibodies and several complex steps to achieve the final conjugate. Furthermore, scaling up and adapting this technology for production in countries with underdeveloped manufacturing infrastructure may prove challenging.

10. Conclusion

ADCs, by combining the specificity of antibodies with the potency of cytotoxic drugs, offer a targeted approach to cancer treatment. However, one of the factors that affect the success of ADCs is the conjugation strategy used, which determines the DAR, localization of the payload on the antibody, stability, and overall therapeutic effectiveness.

Despite advancements in the field, several challenges remain in optimizing ADC conjugation methods. Future research should prioritize conjugation techniques that enhance homogeneity and stability. Moreover, the development of more robust chemical linkers that resist degradation, such as those that prevent the retro-Michael reaction in maleimide linkers, will be crucial. Additionally, improving the precision of drug attachment through innovative approaches, such as the incorporation of non-natural amino acids, can significantly boost the safety of ADCs.

Innovation in conjugation chemistry is essential for the continued evolution of ADCs. Novel techniques, including enzyme-directed conjugation, should be explored to create more versatile and effective ADCs, ultimately leading to more successful outcomes in clinical practice.

The future of ADCs in cancer therapy is highly promising, particularly as treatments become more personalized to match the molecular profiles of individual patients. This tailored approach enhances treatment efficacy and minimizes adverse effects. However, realizing the full potential of personalized ADCs depends on ongoing advancements in conjugation methods. Continued research and investment in bioconjugation technology will be essential to revolutionize cancer therapy, making ADCs more effective and widely applicable in personalized medicine.

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An Analysis of Formula 1 Race and Qualifying Strategy By Sachin Sidhu

Abstract

This paper was motivated by a missing cohesive breakdown of Formula 1 strategy and the models behind it. Formula 1 technology is innovative and, over time, gets passed down to production cars. Understanding the systems currently used in Formula 1 will therefore help improve knowledge about stock cars in the future. This paper aims to be a guide to the complex nature of Formula 1 strategy. It reviews and summarizes the importance of strategy in Formula 1 racing as well as the various ways it can be modeled and predicted. Starting with an overview of Formula 1 and Formula 1 strategy, this paper then addresses current ways teams operate their strategy departments and finally reviews models developed in past papers that mimic Formula 1 race strategy. Throughout this paper, there is a consistent analysis of the difficulty of the decisions that Formula 1 teams have to make and how these decisions can be made easier. By summarizing the current landscape of Formula 1 strategy, this paper aims not only to inform about the complexity of this topic, but also to build appreciation for the technology that has been developed by Formula 1 teams and their partners to assist with strategic decision making.

Formula 1 Overview

Formula 1 is the highest level of motorsport racing in the world. It is a racing circuit that hosts “Grands Prix” all around the world. In Formula 1 there are 20 cars in each race. There are 10 teams, with 2 drivers per team, who complete 24 races in 21 different countries. Two championship categories exist: the drivers’ championship for racers and the constructors’ championship for teams.



Figure 1. Mercedes Technical detail (Piola, 2024)

Every race follows one of two formats. The first format is three practice sessions, each 1 hour in length, and then there is a qualifying session in which drivers attempt to set the fastest lap time to set the starting grid order for the race. The top 10 drivers from each race get points

for the driver standings. and the sum of each team's two drivers' points become the points gained for the constructors' standings. The second format of racing consists of one practice session followed by a shortened qualifying and then a shortened sprint race, awarding less points. Then the full qualifying happens followed by the full race. The regular format is more common than the sprint format. At the end of the season the driver with the most points wins the drivers' championship while the team with the most points wins the constructors' championship. Those are the basics for a Formula 1 season (What is F1?).



Figure 2. Austrian GP racetrack (Motorsport, 2024)

Formula 1 strategy

Formula 1 racing is about optimizing control. Teams have very few aspects that they can fully control and even less that they can guarantee. Hence, they focus on a few main aspects: the driver, the car being driven, team personnel, and the strategy used before and during a qualifying or race session. Normally during a season, both the driver and the ability of a racecar are fixed variables: the former usually remains the same during the season and the latter, while upgradeable, cannot be changed too drastically as teams must work within a budget cap (What is F1?). Similarly, the personnel makeup of the team (technicians, engineers, strategist, etc) is mostly fixed during the year due to non-competition clauses and Non Disclosure Agreements. As such, the biggest determinant for a team's success is their strategy, broadly comprising three main areas: before a Grand Prix, during a qualifying session and during the race itself. Before a race, important decisions need to be made, including the car setup and predicted strategy. However even before that, there is the key issue of tire selection.

The tire choices include hard, soft or medium compound tires. Harder tires have lower grip but are longer lasting, while soft tires have higher grip but shorter lifespans. Medium tires are in between the two (Asher and Braybrook). Softer tires with higher grip are quicker at peak performance; however, they degrade quickly and need to be changed more frequently which takes time.

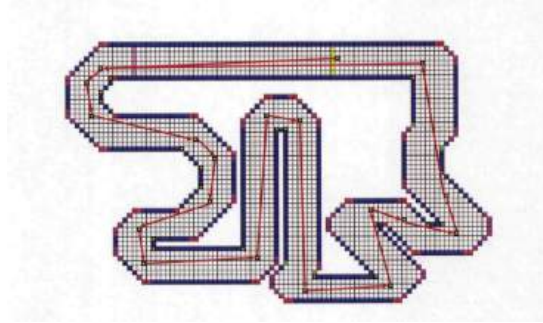


Figure 3. Image of a racetrack model (Hoffman & Margenstern, 2013)

The setup is a pure engineering decision, encompassing mechanical/electrical parameters such as tire pressures, wing angles, engine modes and much more. Predicted strategy is a strategy computed through discussions within the team and simulations through computers. This aspect of pre-race strategy is perhaps the largest part of Formula 1 race strategy in terms of importance. These simulations give engineers an idea of when to pit, a mandatory stop in the race to change tires, or how many times to pit. It can also give an idea of what the race might look like under different conditions, such as weather.

Driver name	Car number	Number of pit stops planned	First stop		Second stop		Third stop		Fourth stop	
			Pit stop lap	Added fuel load (kg)	Pit stop lap	Added fuel load (kg)	Pit stop lap	Added fuel load (kg)	Pit stop lap	Added fuel load (kg)
Albers	1	2	16	59	35	70	-	-	-	-
Freisacher	2	2	15	63	34	79	-	-	-	-
Karthikeyan	3	2	19	59	39	56	-	-	-	-
Monteiro	4	2	18	58	36	70	-	-	-	-
Button	5	1	27	94	-	-	-	-	-	-
⋮	⋮	⋮	⋮	⋮	⋮	⋮	⋮	⋮	⋮	⋮

Figure 4. Table of data typically viewed by a Formula 1 Strategist while planning a Grand Prix. (Bekker & Lotz, 2009)

For pre-race strategy, the key parts are selecting tires and simulating possible strategies. For qualifying, in order to maximize success the team needs to put out their drivers on the right tires at a time when the track is not congested. If those two objectives are met, then the rest is in the hands of the driver. For race day, strategic decisions become increasingly complex. With all the tire decisions made, all the simulations done and the driver's place in the Grand Prix set, there is still so much to be determined to maximize success. Pre-planning involves looking at track quality, driver position, weather, how the tire is predicted to do during the race, pit stop timing and other factors. Even with a pre-strategy set, however, race day execution is still paramount (Bekker and Lotz 953).

Assigned Function	Time to React Seconds	Time to Complete Seconds	Time Taken Seconds	Variability +/- Seconds
Stop car	0	0	0	0
Lift car	0.15	0.2	0.35	0.2
Stabilise car	0.15	0.4	0.55	0.3
Clean front wing	0.15	0.5	0.65	0.5
Clean rear spoiler	0.15	0.5	0.65	0.5
Remove wheel nut	0.15	0.15	0.3	0.2
Remove tyre	0.15	0.35	0.5	0.2
Replace tyre	0.15	0.35	0.5	0.2
Tighten nut	0.15	0.15	0.3	0.2
Lower car	0.15	0.1	0.25	0.2
Check complete	0.15	0.15	0.3	0.2
Spot gap	0.15	0.15	0.3	0.2
Release car	0.15	0.05	0.2	0.2

Figure 5. Image depicting different times required for different types of pitstop changes during a race. (Slater & Hill, 2021)

This strategy is simple to explain but incredibly complex to practice. The race never follows a model perfectly. The team regularly needs to decide whether to change the plan or stick to it; the team could decide to pit early/late, change the type of tire they pit to, instruct the driver to change engineer modes or change almost anything else that was decided before race day. Oftentimes, these calls make or break a race for a team and can be the difference between championship success and failure.

The diagram below summarizes the relationship between concepts in this section. While somewhat broad, it encapsulates the major aspects behind Formula 1 strategy. Something worth noting is the different emphases of pre-race and during race. Pre-race strategy requires extensive prep work, whereas during the race, execution and accuracy are far more important (Bekker and Lotz 956).

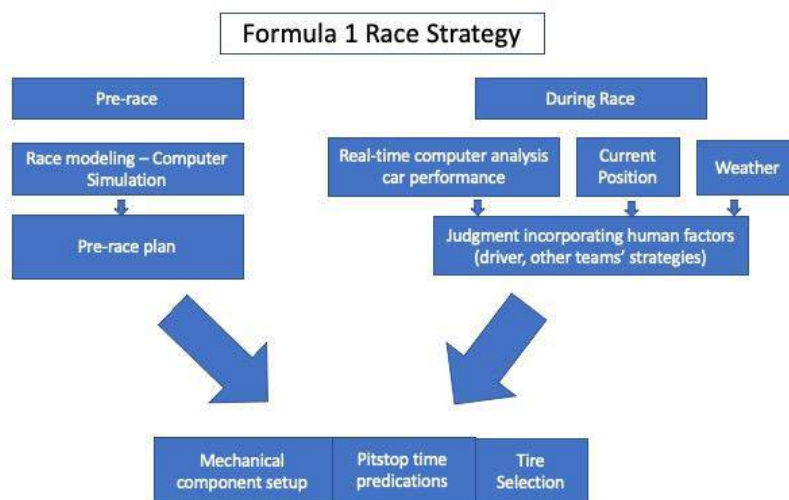


Figure 6. An Image showing a general overview of Formula 1 strategy.

Current state of Formula 1 Race Strategy

When it comes to strategic departments and software all Formula 1 teams will differ from each other. Strategy simulations are often run by software developed by companies outside of Formula 1 and then either sponsored or purchased by teams, such as Red Bull Racing who receive their software from Oracle.

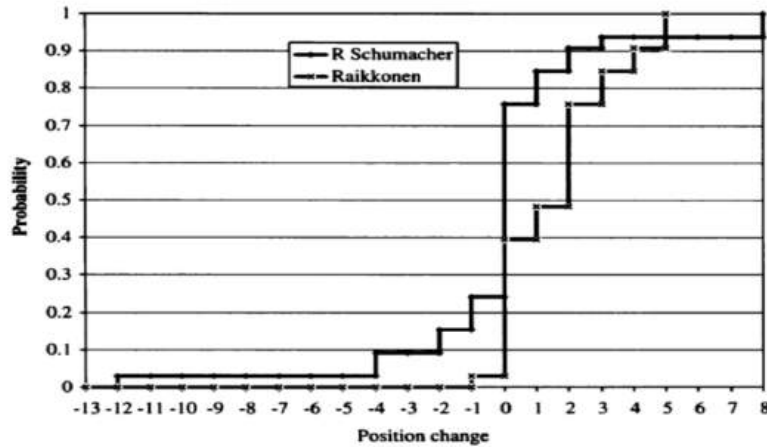


Figure 7. Image showing a prediction for overtaking during a grand prix. (Bekker & Lotz, 2009)

While not a great deal of public information regarding these softwares is available, the basics of how they work can be grasped. These software programs rely heavily on a car's data, specifically data about the cars and the track, including tire pressure, tire surface and core heat, track temperature, engine mode, engine temperature and more. They also rely on race day data, such as total time for a pitstop, other cars' positions and weather. As the race goes on, data is inputted automatically into the software, which will output possibilities and percentages of them occurring. Take, for example, a wet race. Since the possibility of a crash is much more likely, the software may offer strategies of not making a pit stop until an incident happens on the track, as pit stops under a safety car take less time (Heine and Thraves 243).

<i>Sector number</i>	<i>Proportion of lap in sector (%)</i>	<i>Passing manoeuvres permitted?</i>	<i>Proportion of fuel consumption allocated to sector (%)</i>
1	1.25	No	3.9
2	1.25	No	3.9
3	1.25	No	3.9
4	3.5	Yes	1.9
5	2.5	No	1.9
6	2.1	No	2.4
7	2.1	No	2.4
8	2.1	No	2.4
9	2.1	No	2.4
10	4.0	Yes	1.5
⋮	⋮	⋮	⋮

Figure 8. Typical input for a race strategist trying to understand the characteristics of a track. (Bekker & Lotz, 2009)

Ultimately, the strategy decision is between the driver and the team. They will take into account the data and the models as well as intangible aspects: how the driver feels on the day, how well they are driving, and what their goal is for the race. So while the plan can involve many aspects based on computing and data, the ultimate decision will often come down to human instinct.

Strategy Models

Beyond what current Formula 1 teams use for strategy, there has also been public experimentation to try to model Formula 1 strategy. The following example, pictured below, helps to understand the basics of Formula 1 strategy.

Driver name	Car number	Australian Grand Prix		Hungarian Grand Prix		Italian Grand Prix	
		Actual	Estimated	Actual	Estimated	Actual	Estimated
Albers	1	20	20.00	15	15.00	19	18.3
Freisacher	2	19	17.00	18	18.00	18	20
Karthikeyan	3	15	14.74	12	12.00	20	18.7
Monteiro	4	16	16.00	13	13.00	17	16.04
Button	5	11	10.38	5	4.86	8	8.04
Heidfeld	6	19	19.00	6	8.94	7	6.62
Webber	7	5	5.80	7	10.50	14	15.42
Klien	8	7	5.44	19	19.00	13	11.14
Trulli	9	9	8.80	4	3.54	5	4.06
Coulthard	10	4	4.36	20	20.00	15	15.6
Villeneuve	11	13	13.30	16	16.00	11	10.4
Fisichella	12	1	1.00	9	7.40	3	4.34
Massa	13	10	10.54	14	14.00	9	10.38
M Schumacher	14	18	18.00	2	1.92	10	11.44
Sato	15	14	13.02	8	6.14	16	13.42
R Schumacher	16	12	11.92	3	3.42	6	4.26
Alonso	17	3	7.62	11	9.04	2	1.98
Barrichello	18	2	2.06	10	8.92	12	9.42
Raikkonen	19	8	8.06	1	1.32	4	5.72
Montoya	20	6	2.96	17	17.00	1	1.02
<i>P</i> -value		< 0.05		< 0.05		< 0.05	
<i>r</i> _y		0.972		0.970		0.970	
Decision		Reject H ₀		Reject H ₀		Reject H ₀	

Figure 9. A figure showing a model's prediction about races versus the actual result. (Bekker & Lotz, 2009)

Figure 9 shows its output versus an actual race result. Based on inputted data of tires, temperature and other variables discussed earlier, the model gave a predicted placement in the grand prix. The model did this for three grand prix races: Australian, Hungarian and Italian. The actual result is provided in the column next to the predicted result. At the bottom of the page is statistical analysis for each race. In each case the Null hypothesis is rejected, meaning there is statistically significant evidence that this model's predictions are able to reasonably predict race placements (Bekker and Lotz 960).

Most models like this one will only focus on a certain variable affecting a certain event. Take, for example, lap time correlated to timing of a pitstop. A researcher looks at relevant lap time data and builds a picture of when to make a pitstop during a race. When the data shows a strong correlation between lap time and pitstop windows, the researcher can construct a model that can suggest when to make a pitstop based on racepace. That is, a prediction of when the time lost in a pitstop is outweighed by having a faster race car with fresh tires. This is an oversimplified example that shows how real Formula 1 teams use strategy to make decisions.

	<i>Actual race strategy</i>	<i>Simulated race strategies</i>		
		<i>Scenario 1</i>	<i>Scenario 2</i>	<i>Scenario 3</i>
<i>Car 9</i>				
Number of planned pit stops	2	3	2	2
Lap numbers of planned pit stops	18, 36	18, 32, 50	18, 46	18, 46
Position	9	8.10	7.44	7.70
<i>Car 16</i>				
Number of planned pit stops	2	2	1	2
Lap numbers of planned pit stops	19, 40	19, 40	19	19, 46
Position	12	12.08	13.60	9.36
Sum of positions	21	20.18	21.04	17.06

Figure 10. An image showing a predicted and simulated race strategy for two cards. (Bekker & Lotz, 2009)

A model will give suggestions based on inputted data - the more data, the more accurate and reliable the model output/decisions. The software Formula 1 teams use involves incredible amounts of data, making it hard for the public to replicate. However a model that can work with one variable is similar to a model that works with 20, allowing the public to get a better understanding of how strategy models work.

Conclusion

Modeling, planning and executing a Formula 1 race strategy involves many variables. Teams need to take into account factors in their control and not, leading to vast amounts of time and effort being spent on computer models. These models help to support teams when they choose setups, tires and strategy before race, and help the teams adjust their plans during the race. The programs operate based on a variable being inputted then they output a suggestion. These programs have been replicated on a smaller scale by researchers from around the globe, and they can be analyzed and used to fully understand the ones Formula 1 teams use. While the team's software tools utilize the most advanced levels of technology, the future is still unclear. The basis of most of these models is classifiers, which are a key ingredient in Artificial Intelligence. However, the full extent of how AI can affect motor racing is still unclear and should be a large point of research for the future.

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The Educational Background of American Executives: A Detailed Analysis of S&P 500 CEOs in 2024 By Jett Honig

Abstract

This paper examines the educational backgrounds and personal demographic variables of CEOs in the S&P 500. Utilizing a large hand-collected data set on numerous variables regarding the corporations that comprise the S&P 500 and their CEOs, I present data visualizations, summary statistics, and regression analysis to infer patterns amongst the 2024 S&P 500 CEO cohort. I found there was no significant relationship between a specific demographic variable, degree, or attended institution and becoming an S&P 500 CEO, although there are certain ages, demographics, and universities that are represented more heavily than others.

Introduction

The role of Chief Executive Officer is the most sought-after and highest ranking within a corporation. The CEO is responsible for making crucial decisions, ensuring financial stability, mitigating risks, and leading a company through daily challenges. Every product used in people's daily lives has a CEO linked to the brand, and they are accountable for all successes and failures that stem from the good or service. I have a novel set of precise, hand-picked data that steered my research on all five hundred CEOs in the S&P 500. To emphasize the relevance of these 500 individuals, the S&P 500 makes up around 57% of the United States GDP. This statistic emphasizes their essential value as a driving force in our economy. The CEO market is highly competitive. There is a large supply of potential CEOs for few available positions. This paper asks the question: do specific demographic backgrounds and degrees from a particular group of elite universities correlate with the potential of becoming an S&P 500 CEO?

The educational profiles for a CEO position have been extensively researched as educational paths continue to be a prominent discussion due to the controversial perspectives on whether high-level education is necessary. In 2005, Bruce Howard of Wheaton College conducted data analysis and a noteworthy study examining the educational background of corporate America's leaders (3). Dr. Howard utilized numerous visual methods to portray data on many variables: age, undergraduate university, undergraduate degree, graduate school percentage, and graduate degrees. Since the paper was published in 2005, the list of S&P 500 companies has changed tremendously, and the CEO positions at most companies have changed since. Despite these changes, Dr. Howard has created the framework for a highly accurate comparison between 2024's cohort of CEO's in this paper. This research will incorporate similar methods to Dr. Howard's but will look closer at the CEO cohort 20 years later, comparing the results. Comparing past and present data enables us to assess the changes and new trends in the more recent educational paths of executives. Inspired by Dr. Howard, I will model my research paper and update Dr. Howard's report with the 2024 CEO cohort.

Data

Through several hours of hand-picking data across company websites, CEO biographies, LinkedIn pages, corporate directories, and Bloomberg, I collected descriptive and specific data about all five hundred S&P 500 CEOs. Despite the large sample size of CEOs, many of them chose not to disclose particular details on company biographies, leading me to use other sources such as LinkedIn to find facts. Variables like graduate school, graduate school degree, and age were only sometimes available in the fine print on a company's CEO description; it forced me to delve deeper into other sources. I wanted a sample size of precisely 500 CEOs; for example, Paramount Plus has three CEOs, George Cheeks, Chris McCarthy, and Brian Robbins, forcing me to randomly select only one of their three co-CEOs that would be included in the study. The data variables I researched are age, undergraduate college attended, undergraduate major, graduate college attended, and graduate degree. I used histograms, rankings, and pie charts to organize my data in the most precise way.

The data collected for this study uses the S&P 500 stock cohort as of June 23, 2024. Therefore, companies such as Robert Half, Comerica, and Illumina are included in the study. Replacement companies GoDaddy, KKR, and CrowdStrike are absent from the study.

Profiles of the CEOs of S&P 500 Companies in 2024

A. Age

The average age of an S&P 500 CEO is 57 years old. Ages 55 and 59 are the most frequently represented ages in the S&P 500. Both ages 55 and 59 each take up 6% of all CEOs in the S&P 500. 38.6% of the S&P 500 CEOs are in the age range of 50-58. Five CEOs are 39-40 years old, substantially younger than the average. The outlier in the data is Berkshire Hathaway's Warren Buffett; he still holds his position at 93. Figure 1 showcases the age histogram. According to SpencerStuart, the average age of a starting CEO dropped 2.1 years from 55.9 to 53.8 (2). CEOs are getting younger, while most are still effectively gaining momentum in their 50s, similar to the CEO cohort in 2005.

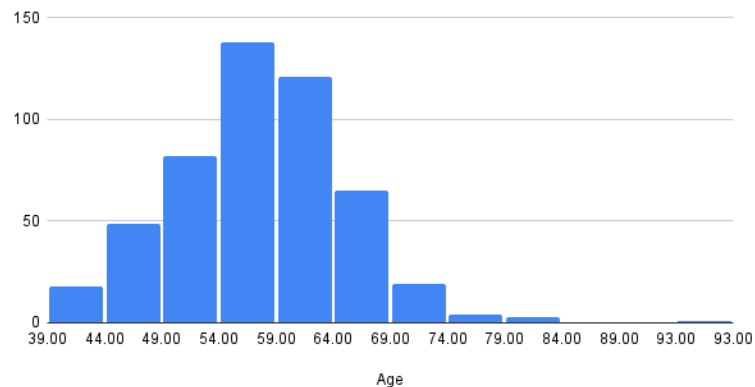


Figure 1: 2024 S&P 500 CEO Age Histogram

The age index was regressed with the market capitalization index. The aim was to see the average increase in market capitalization associated with a CEO who is one year older.

Estimating Equation: the coefficient of interest is β . The interpretation of β is what is the average increase in market capitalization associated with a CEO who is one year older. “MC” represents the market capitalization. “A” represents age. Table 1 represents the results of the regression.

$$MC_i = \alpha + \beta A_i + \epsilon_i$$

Dependent Variable	Independent Variable	R Squared	Estimated Coefficient of Dependent Variable	Standard Error	T Statistic	P Value
Market Capitalization	CEO Age	0.3%	2.386B	1.888	1.264	0.207

Table 1: Regression Results - CEO Age and Market Capitalization

R-squared is the percent of variation in market capitalization that is explained by CEO age. The regression wants to explain that variation. How much does age alone explain the variation in market capitalization? As you can see in the table above, the R-squared is very close to zero, meaning age does not explain very much of the cross-sectional variation in market capitalization because other factors must be driving the variation. The estimated coefficient of the dependent variable is to see for every year older a CEO is, how much the market capitalization is expected to increase or decrease. Standard Error is the standard deviation of the sampling distribution of beta. If the standard error is tiny, we have confidence in the estimated result. T Statistic is a measure of statistical significance. The higher the T value, the greater our confidence in rejecting the null hypothesis. The null hypothesis is that data is equal to 0 so there is no correlation between age and market capitalization. The P-value is the probability that, in repeated sampling, we observe a test statistic at least as large as the one we found, assuming the null hypothesis is true. In this instance, we cannot reject the null hypothesis. The P value is weakly significant.

B. Gender

A study shows that as of May 22, 2024, 32 female CEOs make up 6.4% of the S&P 500 CEOs. This is a 5% increase from 2005 (1).

Education

A. Undergraduate Education

In 2024, 6.2% of the S&P 500 did not graduate with any sort of bachelor's degree. Over a quarter (27.2%) of all CEOs were engineering majors. Since there are many specific types of engineering majors, all are grouped in Figure 2. The engineering majors included are civil, aerospace, computer, chemical, electrical, industrial, mechanical, mineral, marine, petroleum, and metallurgical.

Exactly a third (33.3%) of all CEOs majored in a business-related major. Majors classified as business-related are management, marketing, accounting, finance, and business administration. An economic major is borderline for being included as a business-related major. Therefore, including all economic majors (15.7%) classified as business-related, all business-related majors account for about half (49%) of all 2024 CEOs undergraduate majors. Arts, humanities, and social sciences accounted for 11.2% of all majors. The specific majors in this category are anthropology, architecture, history, communications, English, psychology, journalism, linguistics, philosophy, political science, and government. Figure 2 showcases the spread of undergraduate majors for 2024 S&P 500 CEOs.

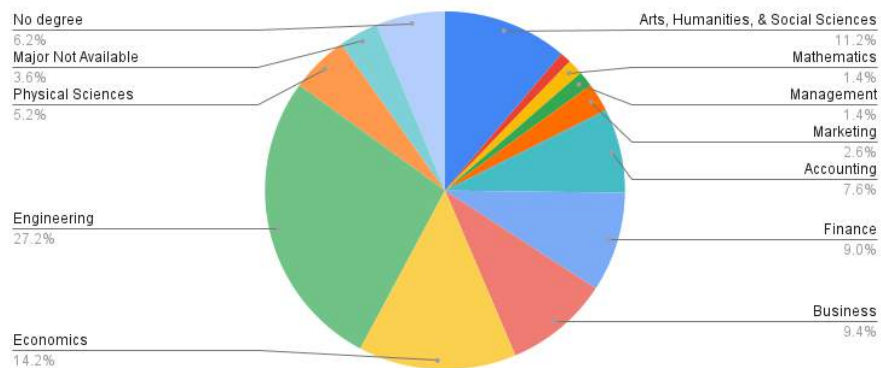


Figure 2: 2024 S&P 500 CEO's Undergraduate Major

Figure 3 summarizes the 2024 S&P 500 CEO's undergraduate major. This excludes all CEOs who did not obtain a bachelor's degree and CEOs whose major was not publicly available.

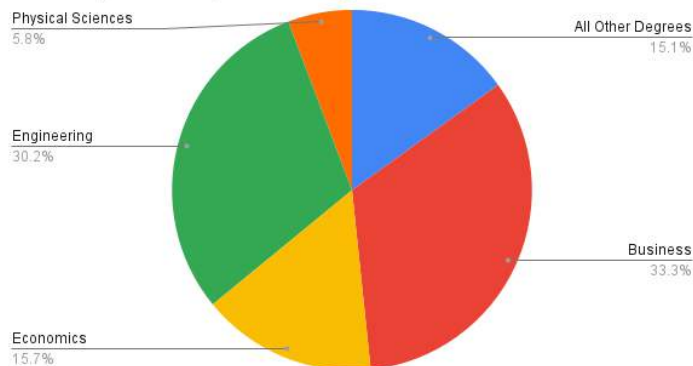


Figure 3: Summary of 2024 S&P 500 CEO's Undergraduate Major

The future generation of CEOs will have many similarities and differences to 2024's S&P 500 CEO cohort. Looking at the United States pool of undergraduate majors conferred in 2022, many things stayed constant for the 2024 CEO cohort. 31.97% of the degrees conferred in 2022 are business-related, highly similar to the S&P 500 CEOs today (33.3%). Engineering majors in 2022 only accounted for 10.48% of all majors. This is 20% lower than the percentage of engineering majors in the 2024 S&P 500 CEO cohort. As an engineering major who has been proven to have had success for many corporate leaders, I am curious about how the CEO cohort in 2044 could look in terms of their educational background and the popularity of engineering majors. Table 2 shows the groups of majors for the national undergraduate pool of 2021-2022 compared to the CEO cohort in 2024 (4).

Undergraduate Degree	Undergraduate Pool 2021-22	CEO's in 2024
Business	31.97%	33.30%
Engineering	10.48%	30.20%
Physical Sciences	11.19%	5.80%
Other	46.36%	30.80%

Table 2: Summary of Undergraduate Degrees of S&P 500 CEOs in 2005 in Relation Pool of Undergraduate Degrees conferred in 2021-22

Rank	Number of Alumni as S&P 500 CEOs	Institution
1	11	University of Pennsylvania
2	10	Cornell University
3	9	Boston College
4	8	Yale University
5	8	Texas A&M University
6	8	Harvard University
7	7	University of Michigan
8	7	University of Florida
9	7	Massachusetts Institute of Technology
10	7	Notre Dame University

Table 3: Top 10 Undergraduate Institutions With Alumni as S&P 500 CEOs in 2024 by Total Number of Alumni As CEOs

The ACC led all conferences with the most alumni (11.1%). The Ivy League followed closely behind (10.4%). Southern Methodist University (0), Stanford University (3), and University of California - Berkeley (4) were accounted for in the ACC. Southern Methodist University officially joined on July 1st, 2024, and Stanford University and the University of California - Berkeley officially joined on August 1st, 2024. The Big Ten does not include recent additions Oregon University (0), University of Washington (1), University of California - Los Angeles (3), and University of Southern California (3). Figure 4 showcases the distribution of S&P 500 undergraduate degrees by athletic conference.

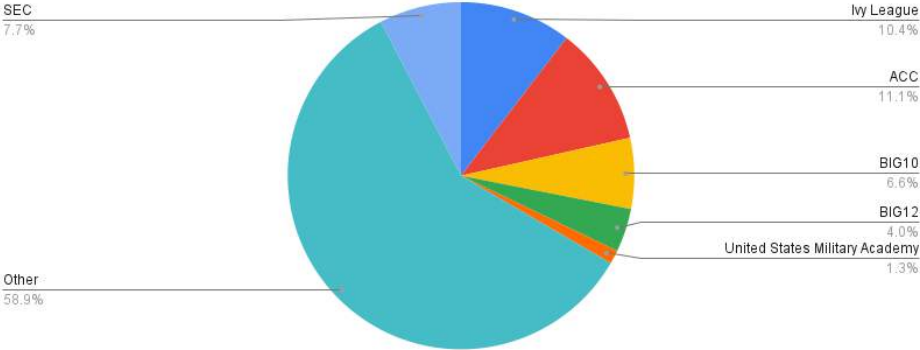


Figure 4: Distribution of Undergraduate Degrees by Athletic Conference

B. Advanced Education

More than half (65.2%) of the 2024 S&P 500 CEO cohort attended graduate schools, and it is evident that having a graduate degree is highly beneficial. However, it has also been shown to be optional because 34.8% of the S&P 500 CEOs did not attend graduate school. Figure 5 shows the percentage of graduate school attendees.

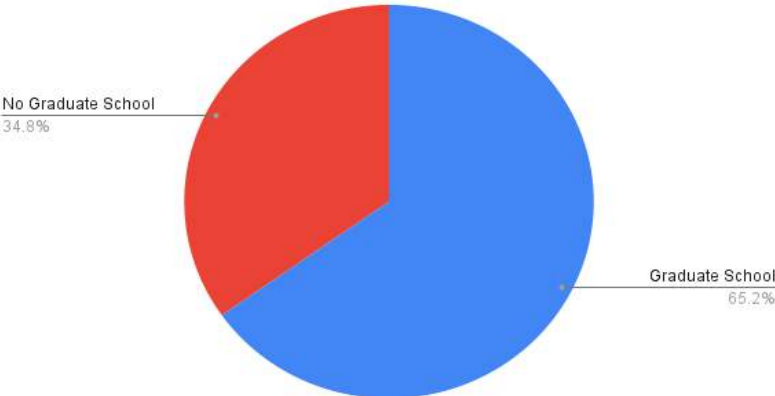


Figure 5: 2024 S&P 500 CEOs Who Attended Graduate School Vs. Not Attended Graduate School

Of the 326 CEOs who attended graduate school, Harvard(41) has the most alumni working as CEOs for an S&P 500 company, followed by Stanford University(18) and

Northwestern University (18). Of Harvard's 41 alums, 36 have an MBA from Harvard Business School. Table 4 ranks the graduate schools with the most alumni serving as an S&P 500 CEO.

Rank	Number of Alumni as S&P 500 CEOs	Institution
1	41	Harvard University
2	18	Stanford University
3	18	Northwestern University
4	16	Columbia University
5	13	University of Chicago
6	11	University of Pennsylvania
7	9	Massachusetts Institute of Technology
8	9	University of Michigan
9	7	New York University
10	6	Boston University

Table 4: Top 10 Graduate Institutions With Alumni as S&P 500 CEOs in 2024 by Total Number of Alumni As CEOs

42% of all 2024 S&P 500 CEOs obtained an MBA, proving it is beneficial in the long run. An MBA can increase your networking ability and worldview and potentially place you into a better job after college. 34.8% of all 2024 S&P 500 CEOs did not obtain an advanced degree. 15.6% of 2024 S&P 500 CEOs earned a master's degree in other fields of study: some other subject fields include engineering, journalism, and computer science. Figure 6 shows the spread of graduate degree profiles.

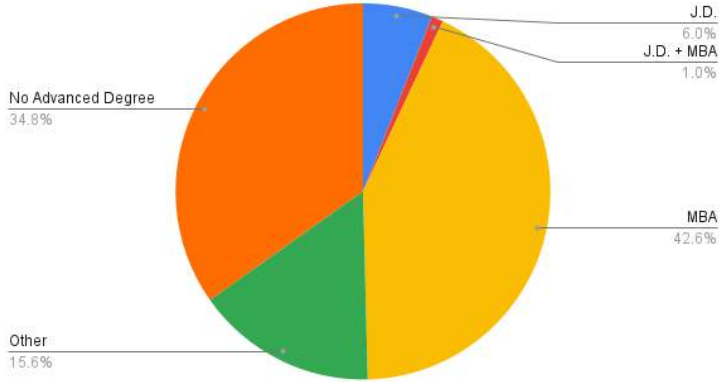


Figure 6: 2024 S&P 500 CEO Graduate Degree Profile

Comparative Analysis - 2005-2024

In roughly two decades, the S&P 500 CEO cohort has changed. Many variables of the CEO's educational background stayed constant, and others fluctuated. The female population serving as CEOs in the S&P 500 increased from 7 to 32. According to SpencerStuart, the average age of a starting CEO dropped 2.1 years from 55.9 to 53.8. The percentage of CEOs who did not receive a bachelor's degree more than doubled from 3% to 6.2%. An undergraduate engineering major has become more popular in the past 20 years: engineering majors increased 8.2% since 2005. Economics majors also increased by 4.2%. In 2005, Harvard had the most alums working as an S&P 500 CEO with 15. In my study, the University of Pennsylvania had the most alums working as an S&P 500 CEO with 11. In 2005, the Ivy League had the most alums working as a CEO in the S&P 500, followed by the Big Ten. The Atlantic Coast Conference (ACC) has the most alumni working as CEOs in the S&P 500, followed by the Ivy League. In 2005, many conferences were composed of different teams, but the majority stayed the same. An advanced degree has become more popular amongst S&P 500 CEOs. The percentage of CEOs who have a graduate degree has increased by 12.3% since 2005.

Conclusion

In this paper, I investigate whether achieving the position of an S&P 500 CEO depends on having a particular demographic background or obtaining a specific undergraduate or graduate degree from a distinct group of universities. Through extensive research, I diligently hand-picked a large raw data set of many variables related to S&P 500 companies and their CEO. The data was meticulously organized into different visualizations, analyzed, and compared to the 2005 study by Dr. Howard. There is no necessary prerequisite that can be obtained to become a CEO; both genders, all ages, college dropouts, and PhDs are all represented in the S&P 500 CEO cohort, proving that CEOs come in all shapes and sizes. There is no clear path to obtaining the position. Building on these results, further research could be conducted on different CEO cohorts (e.g., Russell 1000), looking at other variables not included in this study and using more quantitative metrics to analyze the data.

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and Tanghulu By Jaeheon Shim

Abstract

South Korea has seen a number of foods trend in the past decade or so, a phenomenon this paper coins as a “food fad.” This paper sets out to uncover the factors that lie behind the appearance of food fads in South Korea by focusing on the malatang and tanghulu fad, both of which gained popularity in recent years after trickling into the country from China. The factors this paper identifies as elements that potentially contribute to the development of a food fad are widespread social media usage and the collectivist culture of South Korea. Statistical analysis using data from a survey administered to Koreans residing in Korea between the ages of 13-29 revealed that viewing social media content on malatang/tanghulu and collectivism are correlated with trying those foods for the first time. However, these factors were not indicative of whether Koreans would subsequently consume those foods. This study revealed important insight about the consumer behavior of Koreans when it comes to food.

Keywords: food fad, social media, collectivism, malatang, tanghulu

I. Introduction

While certain gadgets and fashion items are deemed trendy, it may be odd for some people to know that some food items are considered trendy in some parts of the world. It is often the case in South Korea that food items become trending, stay relevant for a couple of years, and then almost disappear in a fad-like fashion. In 2014, honeycomb ice cream swept the nation, and in 2016, Taiwanese castella vendors were extremely popular, with each vanishing within a couple of years of reaching their peaks. In this paper, I will refer to such a phenomenon as a “food fad.”

The most recent and prominent additions to South Korea's history of food fads are malatang and tanghulu. They were not a staple food in South Korea, entering the country only in recent years and quickly gaining popularity. Malatang is a spicy dish from China that serves stir-fried vegetables, meat, and fishcakes in a chili oil broth. Tanghulu is a fruit skewer with the fruit coated with melted sugar. The sugar is given time to harden again before serving. This dessert likewise originates from China. Malatang has seen explosive growth within South Korea in recent years, with the number of franchises in the nation increasing from 4 to 30 between 2018 and 2021 (Lee et al.). Tanghulu sales in South Korea have likewise increased by 1678% in 2023 compared to 2022 (Yoo).

This paper aims to uncover the mechanism behind the continuing cycle of food fads South Korea has seen within the past ten years. Doing so would provide insight into consumer behavior in South Korea, which would prove to be valuable for marketing and sales purposes. Due to the convenience of data collection, this paper will focus on the malatang and tanghulu fads in particular, as they are ongoing. The penetration and widespread use of social media and the collectivist tendencies of South Koreans are some factors that could have contributed to the

ongoing malatang and tanghulu fad in Korea. An examination of each factor will culminate into a survey, and the chi-square test of independence and Pearson's correlation coefficient will be used to analyze the data to see if there is a correlation between any of these factors and the popularity of malatang/tanghulu in South Korea.

II. Literature Review

There is no existing research about food fads in South Korea, so this study conducted a literature review tangentially on related topics. One of the factors that this paper predicts as a cause of food fads is social media. Various studies suggest a relationship between the use of social media and the choices people make on food choices. Images of food tend to make people desire the food at hand ("Pictures of Food"). Considering how social media is a platform where social and visual factors coexist, the idea that food-related posts on social media could influence people's choices on what food to consume is not far-fetched (Brown). Koreans in the age groups 10-19 and 20-29 used social media most frequently in 2023, with 57.2% of the former and 55.5% of the latter checking at least one post per day, suggesting they are the most vulnerable to getting influenced by media platforms ("Korea Media Panel Survey"). The social media use rate fell sharply for other age groups, with only 42.7% of the 30-39 group, 35.3% of the 40-49 group, and 26.9% of the 50-59 group checking at least one post per day ("Korea Media Panel Survey"). Coincidentally, teens and people in their twenties accounted for 33.3% of total malatang consumption and 38.8% of tanghulu consumption in South Korea between December 2023 and February 2024 (Park). People in their forties accounted for 30.3% of total malatang consumption in South Korea in the same period, but this was credited to the fact that a significant portion of spending in this age group is for their children (Park).

Social media allows quicker interaction between people, which in turn leads to a quicker spread of information, effectively shaping the "beliefs, attitudes, and norms" related to eating behaviors among adolescents on a large scale (Chung et al. 6). Food advertisements, in particular, convey the impression that the food in question is widely consumed, evoking a positive emotional response from the viewer (Kucharczuk et al. 9). Adolescents exposed to influencer advertisements of unhealthy snacks showed a notable rise in their consumption of these foods (Brown). They also pick the food that influencers advertise on social media and share their eating habits with their friends, which amplifies the reach of these influencers (Kucharczuk et al. 9). This is likely due to how people often feel acquainted with influencers they see on social media. It could be that watching influencers eat certain foods might subconsciously nudge people to choose the same foods to eat. There was plenty of content of Korean celebrities eating malatang and tanghulu on social media in the past few years, and while these contents were not advertisements, they could have had a similar effect, leading to increased consumption of malatang and tanghulu.

Another factor this paper aims to test is whether the collectivist tendency Koreans show has a connection with the periodic food fads observed in South Korea. Korean culture is highly collectivistic compared to other countries (Moon 3). Within a collectivistic culture, people prefer

to be a part of a group (Chung 61). Accordingly, Korean culture values interdependence, and Koreans place a lot of emphasis on social connections and the relationships they form with others (Ahn 7). Koreans also tend to separate in-group and out-group members (Chung 64). This type of collectivism makes people want to agree with the group and feel a strong social pressure to conform (Tan 36). According to research by Han and Ahn, Koreans often go along with the group to form a consensus rather than express their individual opinions (qt. in Ahn 7). Collectivism in Korean culture plays an important role in why Koreans tend to dress similarly, as they attempt to follow up with the most recent trends and be a part of the majority (Yoo and Ha 26). No research proves a similar observation regarding food choices, but the influence of collectivism likely extends beyond just fashion and possibly into food. The mechanism for the spread, when you put these two factors together, is as follows: 1) some people eat a particular food after viewing related content on social media; 2) these people suggest getting those foods to their groups; 3) everyone in the group either conforms to the social pressure or goes along with the majority for group harmony and tries that food 4) because more people eat this food, more related content pops up on social media; 5) trying the food becomes a trend.

III. Methodology

Since it was difficult to test the above-stated mechanism entirely, this study opted to examine the first three parts of the mechanism. The researcher chose an anonymous electronic survey with thirteen questions for this purpose. The questions intended to find data on the above hypotheses were yes or no questions on whether the participants had seen content related to malatang and tanghulu on social media, whether they value group harmony, whether they are sensitive to peer pressure, and whether they had ever tried malatang or tanghulu. The survey also required participants to answer questions on a 5-point Likert scale or on other scales that asked for the frequency of specific actions or how they felt about different subjects. These questions included how often the students used social media and saw content related to malatang and tanghulu, how much they feel social pressure and value group harmony, and how often they eat malatang or tanghulu.

The researcher chose to use a survey for this study because it allows efficient data collection from a large sample and because the quantitative data collected from it would allow for flexible statistical analysis. With no fixed sample, disseminating an online survey was deemed the best way to reach a broad audience. The survey had to be anonymous because it asked for information people might not wish to honestly answer if their names were revealed, such as their sensitivity to peer pressure and whether group harmony is important to them.

The researcher conducted a sample test run of the survey to see if any questions were confusing. After that, the researcher revised the survey and sent the final version to participants through Google Forms. The survey targeted teenagers and people in their twenties because their age group was perceived to be the heaviest consumers of malatang and tanghulu and the most active social media users. Tweens were excluded from this study to avoid any confusing data, as they might not have the autonomy and means to buy what they wish to eat. Every participant was ethnically Korean residing in South Korea. Convenience sampling was used to collect initial

responses from the researcher's school, church, and other extracurricular organizations, from which snowball sampling was used to receive further responses. All participation was voluntary, and every participant was informed of the anonymity of the survey. Information on the research topic was not shared with the participants to prevent subject bias. Every participant received the same set of questions to enhance the reliability of the study. After three weeks of data collection, the researcher had 83 total responses. Excluding responses from participants who did not fall under the intended age group and those containing inappropriate or irrelevant content, 71 responses remained for analysis.

The primary method of statistical analysis chosen was the chi-square test of independence. The chi-square test was a good fit for this study because it tests whether there is a significant association between two categorical variables. In the chi-square test, there is the null hypothesis, which claims there is no significant association between two variables, and the alternate hypothesis, which states that there is a meaningful correlation between two variables. A calculation is done through the following formula to determine whether to reject or accept the null hypothesis:

$$\text{Chi Square Statistic } X^2 = \sum \frac{(\text{Observed Frequency} - \text{Expected Frequency})^2}{\text{Expected Frequency}}$$

The answers to binary questions were used for the chi-square test. The hypotheses this study tested using the chi-square test of independence were as follows:

- 1) H0: There is no significant association between viewing malatang or tanghulu-related content and trying those foods for the first time.
H1: There is a significant association between viewing malatang or tanghulu-related content and trying those foods for the first time.
- 2) H0: There is no significant association between valuing group harmony and trying malatang and tanghulu for the first time.
H1: There is a significant association between valuing group harmony and trying malatang and tanghulu for the first time.
- 3) H0: There is no significant association between being sensitive to peer pressure and trying malatang and tanghulu for the first time.
H1: There is a significant association between being sensitive to peer pressure and trying malatang and tanghulu for the first time.

Pearson's correlation coefficient was used to see if there were any linear correlations between the following:

- 1) How often do people see malatang and tanghulu on social media, and how often do they consume them?
- 2) How important group harmony is to one, and how often one consumes malatang and tanghulu.

- 3) How sensitive one is to peer pressure, and how often one consumes malatang and tanghulu.

The answers to the Likert scale and frequency questions were used for this part of the study. The answers to frequency questions were grouped into ordinal data to resemble the responses from the Likert scale questions. This process made running Pearson's correlation coefficient on the data simpler.

IV. Findings

A 95% confidence level was used for the chi-square test calculations, typical in social science. The data analysis for the three sets of hypotheses is available below.

- 1) H0: There is no significant association between having seen malatang or tanghulu related content and trying those foods for the first time.

H1: There is a significant association between having seen malatang or tanghulu-related content and trying those foods for the first time.

Table 1. Malatang/Tanghulu Trial and Malatang/Tanghulu Content

Tried Malatang/Tanghulu	Encountered Malatang/Tanghulu Content on Social Media		
	Yes	No	Total
Yes	56	5	61
No	5	5	10
Total	61	10	71

The null hypothesis was rejected, and the alternate hypothesis was accepted, meaning there is a significant association between having seen malatang or tanghulu-related content on social media and trying those foods for the first time.

$$X^2(1, N = 71) = 12.4074, p=0.0043. \text{ Significant at } p < 0.05$$

- 2) H0: There is no significant association between valuing group harmony and trying malatang and tanghulu for the first time.

H1: There is a significant association between valuing group harmony and trying malatang and tanghulu for the first time.

Table 2. Malatang/Tanghulu Trial and Group Harmony

Tried Malatang/Tanghulu	Group Harmony is Important		
	Yes	No	Total
Yes	51	10	61
No	5	5	10
Total	56	15	71

The null hypothesis was rejected, and the alternate hypothesis was accepted, meaning that there is a significant association between considering group harmony as important and trying malatang or tanghulu.

$\chi^2(1, N = 71) = 5.8231, p=0.0158$. Significant at $p < 0.05$.

- 3) H0: There is no significant association between being sensitive to peer pressure and trying malatang and tanghulu for the first time.
 H1: There is a significant association between being sensitive to peer pressure and trying malatang and tanghulu for the first time.

Table 3. Malatang/Tanghulu Trial and Peer Pressure Sensitivity

Tried Malatang/Tanghulu	Sensitive to Peer Pressure		
	Yes	No	Total
Yes	38	23	61
No	4	6	10
Total	42	29	71

The null hypothesis was accepted. No significant association was found between sensitivity to peer pressure and trying malatang or tanghulu.

$\chi^2(1, N = 71) = 1.7675, p=0.1837$. Not significant at $p < 0.05$.

The findings from Pearson's correlation coefficient were less promising than those from the chi-square test of independence. There was a negligible correlation between the variables tested.

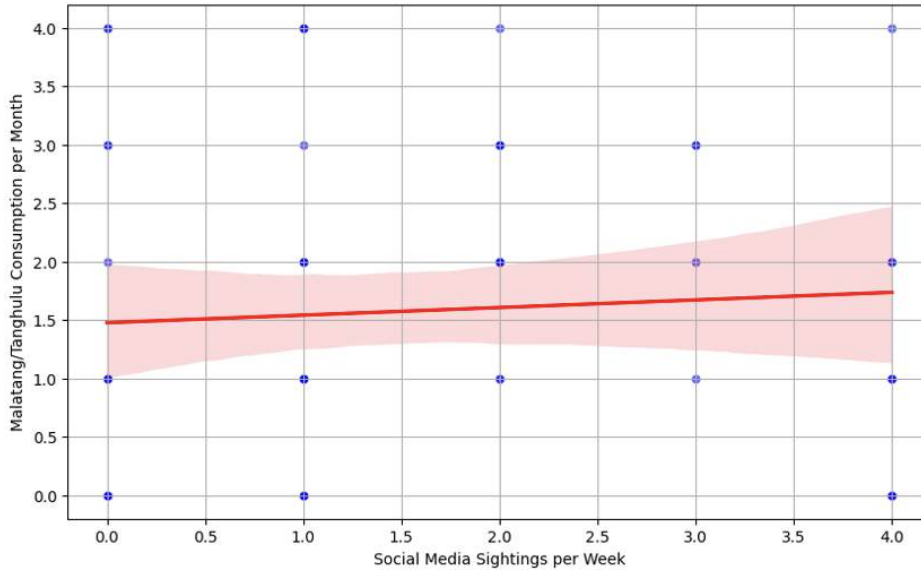


Fig 1: Social Media Sightings of Malatang/Tanghulu per Week vs. Malatang/Tanghulu Consumption per Month

The Pearson correlation coefficient for the two variables above was 0.0653, which signals negligible correlation.

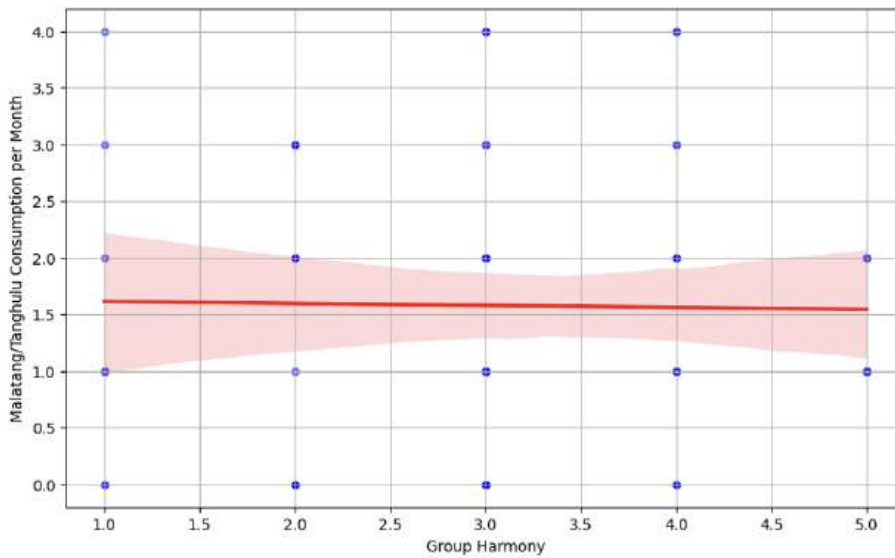


Fig 2: How Strongly One Values Group Harmony vs. Malatang/Tanghulu Consumption per Month

The Pearson correlation coefficient was -0.0154 for this study, indicating a negligible correlation.

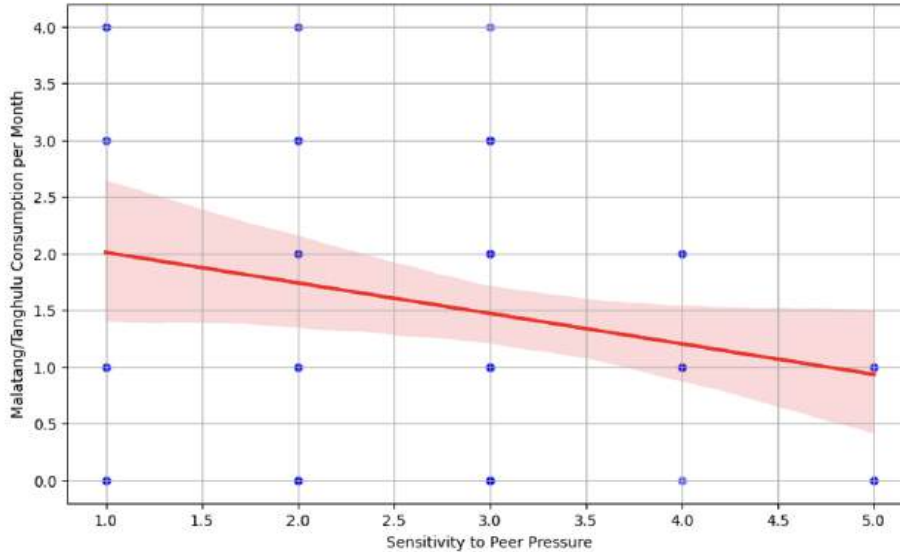


Fig 3: Sensitivity to Peer Pressure vs. Malatang/Tanghulu Consumption per Month

The Pearson correlation coefficient was -0.2327, which is again a negligible correlation.

V. Discussion

The study found that there is a significant correlation between the following variables for Koreans between the ages 13-30: 1) viewing malatang/tanghulu-related content on social media and choosing to try those foods and 2) considering group harmony to be an important value and trying malatang and tanghulu. While this alone is not enough to prove the food fad mechanism as proposed by this paper, this provides enough evidence to suggest that some parts of the mechanism may be true. However, this study did find that there was no significant correlation between considering oneself to be sensitive to peer pressure and choosing to try malatang and tanghulu, which suggests that it may not be pressure from others but rather one's desire to conform to a group that is related to trying these foods. The correlation tests conducted using Pearson's correlation coefficient reveal that there is no statistically significant relationship between the variables studied. How sensitive one is to peer pressure and group harmony, or how exposed one is to social media posts on malatang or tanghulu, did not seem to influence how frequently respondents consume malatang or tanghulu. When put together, it appears that social media posts about malatang and tanghulu and the collectivist tendencies Koreans show are correlated with trying those foods for the first time, but these factors are not indicative of whether these people will continue to consume those foods.

There are limitations to this study that future studies ought to address. First, the sample only consisted of 71 participants, which is not large enough to draw conclusive findings about the Korean population. Replicating the findings of this study with a larger sample would bolster the results found in this paper. In addition, while everyone received the same survey to mitigate issues with reliability, the survey depended on self-reported data, which may be subject to inaccuracies due to a skewed perception of oneself or faulty memory. A more objective way to measure sensitivity to peer pressure or how important group harmony is to a person would

strengthen the findings of this research. Also, there may be factors that contribute to the rise and fall of food fads in South Korea that this study did not count, such as high population density, to name one. Despite these limitations, this study is meaningful in that it is the first to delve directly into the reason behind food fads that frequently appear in South Korea.

VI. Conclusion

This study attempted to uncover the reasons behind the periodic rise of food fads in South Korea observed since the mid-2010s. This study focused exclusively on the ongoing malatang and tanghulu fad to ease the data collection process. Because there was no previous research done in this area, the literature review was carried out in tangential fields, particularly on the effect social media has on people's choices in choosing food to consume and the collectivist tendency Korean society and individuals show.

A survey of thirteen questions was sent out to Koreans between the ages of 13-29, from which 71 responses were fit to be analyzed. Chi-square tests of independence were conducted on binary data, while Pearson's correlation coefficient was used to analyze Likert scale and frequency data. The statistical analysis found a significant correlation between viewing social media content on malatang and tanghulu with trying those foods, and between considering group harmony an important value and attempting those foods. How often people saw malatang/tanghulu content on social media, how highly they valued group harmony, and how sensitive they believed themselves to be to peer pressure were not correlated with how often these people chose to eat malatang or tanghulu. Therefore, this study concluded that while viewing related social media content and valuing group harmony were good indicators of whether one would try malatang or tanghulu, they did not influence how frequently one would subsequently consume these foods.

The findings from this study shed some light on the consumer behavior of South Koreans when it comes to food, and future studies in this area would reveal more insight if they consider other factors that may play a role in the development of a food fad, and also why food fads always end up fading.

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Beyond the Ballot: Exploring the Limits and Aspirations of Democracy in a Polarized World By Hyungjoon Choo

Abstract

Political polarization has intensified in recent years, with divisions growing more intense and deeply entrenched. Many commentators have observed that polarization often stems from democratic excess, where the mechanisms of democracy—such as majority rule—are pushed to extremes, leading to the marginalization of minority voices and the erosion of civil discourse. In other words, the argument is that too much democracy is the cause behind political polarization. But is there such a thing as too much democracy? The paper explores this question by examining two conceptualizations of democracy: procedural and virtue-oriented. Under the *procedural* view which sees democracy primarily as an electoral system, there can be such a thing as “too much” democracy, as it is just one good and can be pursued at the expense of other rights and values. When we adopt the *virtue-oriented* definition, however, we are led to conclude that there is no such thing as “too much” democracy, as it is an ethical orientation that we are always learning to adopt as an equal member of a collective society. From this standpoint, there can never be “too much” democracy; rather, more democratic virtues are needed to navigate our polarized and conflict-ridden world. The paper ultimately concludes that democracy is not merely a political system but an aspirational ideal—a standard by which to evaluate society and a goal toward which we must continually strive.

1. Introduction

1.1 Context

Throughout history, political thinkers have grappled with the potential and pitfalls of democracy. Aristotle argued that rule by the majority could be fair, but only if guided by the ambition and virtue of a select few. The American Founders, too, built a constitutional framework to limit the unchecked will of the people, even as they celebrated self-rule. Indeed, political thinkers have always grappled with the promises and pitfalls of democracy. This discussion continues today, especially since the rise of Donald Trump in 2016. Many thinkers are arguing that the current polarized political landscape may be caused by an abundance of democracy. Political commentator Andrew Sullivan observes that “democracies end when they are too democratic” (Sullivan, 2017). The rise of populist figures worldwide demonstrates the drawbacks of excessive democracy. When the democratic process is overly inclusive, Sullivan argues, it can lead to the election of leaders who will continually exploit populist sentiments and abrade liberal norms. Dalibor Rohac, an American author and policy analyst, makes a similar observation in his essay “Too Much Democracy is Killing Democracy” (Rohac, 2020). Excessive citizen involvement, Rohac argues, is exacerbating partisanship. In a world of “too much democracy,” political parties no longer function as mediators of diverse interest but become battlegrounds for uncompromising groups.

Indeed, the notion that too much democracy can lead to democratic backlash is gaining traction among commentators worldwide. This debate raises a critical question: is democracy good in and of itself, or is democracy something we should be critical of in its wide promotion and adoption? But to answer this question, it is imperative to first ask: what is democracy?

1.2 Democracy, a contested concept

Etymologically, democracy stems from the ancient Greek words “demos” (the people) and “kratos” (to rule). Put together, democracy means “rule by the people” (Larsen, 1973). In its essence, democracy is the power of the people to come together to deliberate the good of society and make decisions that shape their collective fate.

However, democracy is an “essentially contested” concept (Gallie, 1956). Despite the long history of democracy since the Greeks, there is little agreement on its meaning and appropriate scope. Turning to etymology does not fully answer the question of what democracy is, however, as it leaves open the question of what constitutes “the people.” Even Abraham Lincoln’s cogent formulation—democracy as government “of the people, by the people and for the people”—leaves us with a critical question: who exactly comprises “the people.” This is a difficult question to settle, especially in today’s multicultural societies with a continuous inflow and outflow of immigrants (Song, 2012). Who the people are is continuously in flux.

Leaving aside the question of who constitutes the people of a country, there is also the question of how to procedurally arrive at something called the “will of the people.” Is it an aggregated preference of individual citizens, or is there something akin to Rousseau’s “general will,” a volition toward the common good that is above and beyond individual passions and interests (Rousseau, 1762)? If so, how do we come to know what it is?

I do not dare to resolve this long debate that has spanned centuries. Instead, I divide the conceptualizations of democracy into *procedural* and *virtue-oriented* camps and assess whether there can be such a thing as “too much democracy” under these definitions. I recognize that this is a gross reduction of a rich debate in democratic theory and practice, but such a categorization seems inevitable for this short paper.

This paper argues that under the *procedural* definition of democracy as an electoral system, there can be such a thing as “too much” democracy, as it is just one good and can be pursued at the expense of other rights and values. When we adopt the *virtue-oriented* definition, however, we are led to conclude that there is no such thing as “too much” democracy, as it is an ethical orientation that we are always learning to adopt as an equal member of a collective society.

2. Argument

2.1 Assessing Democracy through the Procedural Definition

The procedural definition is nicely summarized by Samuel Huntington who writes that “elections, open, free, and fair, are the essence of democracy, the inescapable sine qua non”

(Huntington, 1980, 9). Democracy, under this minimalist view, is a competitive procedure for selecting leaders by the people they govern. This definition was first critically formulated by Joseph Schumpeter in 1942. In his *Capitalism, Socialism, and Democracy*, Schumpeter pointed to what he conceived to be the limits of the “classical theory of democracy,” which he found to be fraught with normative ideologies. Instead, Schumpeter puts forth “another theory of democracy,” which he defines as the “institutional arrangement for arriving at political decisions in which individuals acquire the power to decide by means of a competitive struggle for the people’s vote” (Schumpeter, 1942, 269). Many researchers have followed Schumpeter’s path. Indeed, as Russell and others at the Center for the Study of Democracy point out, the most widely adopted definitions of democracy focus on the *procedures* of democratic governance (Collier and Levitsky, 1997; Dahl, 1971; Prezeworski et al., 2020; Russell et al., 2007).

Many theorists who embrace such a proceduralist definition point to its analytical clarity. Huntington, for instance, argues that this definition offers “analytical precision and empirical references that make the concept a useful one” (Huntington, 1980, 7). Others, like Fareed Zakaria, point out that “to go beyond this minimal requirement and label a country democratic only if it guarantees a particular catalog of social, political, economic, and religious rights—which will vary with every observer—makes the word “democracy meaningless” (Zakaria, 2003). Indeed, this minimal form has enabled scholars to create scales to conduct global, cross-national quantitative analyses of governments around the world (Landman, 2007). Under this minimalist view, one could maintain that there is such a thing as too much democracy. Democratic elections have been regarded as sacrosanct, but it is not free from errors. Democratic elections can lead to situations wherein the people vote to violate the natural rights of minority individuals. Consider the case of Nazi Germany during World War II, for example. Adolf Hitler was voted into authority through an electoral process. Nevertheless, when Hitler came to power, he not only stripped away the rights of the Jewish people but also violently oppressed them with ruthless cruelty.

Indeed, this is a rather extreme case involving genocide, but there are numerous other examples of democratically elected governments that have failed to uphold the rights of their citizens. Elected leaders—often ones that have been re-elected or reaffirmed through national referenda—are blatantly disregarding the constitutional limits of their power and denying citizens their essential rights. For instance, in an oft-cited example, Zimbabwe’s government leader, elected into office with 52.6% of the vote, is using the law as an instrument of oppression (“Zimbabwe: Human Rights Under Attack,” 2023). In another instance, Turkey’s national election has given rise to a government blatantly exploiting the rights of citizens (“Turkey: UN Report,” 2018). These examples across time and space serve to demonstrate the striking possible outcomes of the democratic process.

At times, the democratic process can even create situations that undermine the democratic process itself. That is, many citizens in so-called democracies have voted to exclude certain minority groups from voting, linking the right to vote to qualifications like wealth or race. Consider Jim Crow America and the policy of Apartheid in South Africa. In both cases,

individuals who had been voted into office denied the voices of others. Throughout history, the “will of the people” defined as the majority vote has sanctioned the denial of others’ votes.

Fareed Zakaria calls these systems “illiberal democracies.” *In the Future of Freedom: Illiberal Democracy at Home and Abroad*, Zakaria argues that the unchecked proliferation of democratic processes has given rise to “illiberal democracies,” tyrannical regimes that maintain the facade of democracy but lack the foundations of a truly liberal democracy. What Zakaria points out is that democracy should never be pursued at the expense of liberty—understood as the “rule of law, a separation of powers, and the protection of basic liberties of speech, assembly, religion, and property” (Zakaria, 2003). Democracy without these norms can be self-defeating. Or as Tibor Machan puts it, “unlimited democracy can undo democracy itself” (Machan, 2002, xx). In short, we may say that there can be such a thing as too much democracy if the overemphasis on electoral procedures is giving rise to results that violate natural rights and liberties.

2.2 Assessing Democracy through the Expansive Definition

However, if by democracy we mean its electoral system, then we must further question *why* that specific institution is considered essential to democracy. In other words, what do democracy’s central institutions seek to achieve? I argue that underlying the institution of “free, fair elections” is the principle of equality. It is the belief that we are all born equal, that no human is innately a slave or a master. And if individuals are equal, then they should not be arbitrarily subject to the will of another. In this section, I argue that when understood more broadly as a set of aspirations toward political equality, there can never be such a thing as “too much democracy,” as this is an aspiration and an *ethos* we are always trying to fulfill.

Democracy is not just a set of institutions—a process of making group decisions—but rather a set of beliefs about political equality. This premise of equality is at the heart of democracy. As Robert Talisse puts it in *Sustaining Democracy*, “the very idea of an election embeds the idea that everyone who is affected by a collective decision is entitled to an equal share of the decision-making power” (Talisse, 2021). In other words, the premise of equality necessitates that governance operates with the consent of the governed. When we are all equals, each individual adult should have an equal say in the decision-making of policies that affects them.

Alternatively put, democracy is anchored in the principle of individual sovereignty. In a democratic system, individuals collectively hold the sovereign right of political rule. Sovereignty in a democracy is a shared and public right, not reserved for particular groups of people, and certainly not derived from hereditary privileges. As bearers of sovereignty, individuals are co-ruling as political equals.

The democratic notion of political equality and individual sovereignty implies certain rules of conduct. Many political thinkers have called these rules the democratic ethos (Dewey, 2016; Tully et al., 2022; White, 2016). The democratic ethos is a set of attitudes that show a willingness to treat other citizens as equals. Talisse points to three important attitudes: the virtue

of public-mindedness, reciprocity, and transparency (Talisso, 2021, 31-32). In other words, the democratic ethos calls for an orientation towards the common good, the consideration of multiple perspectives, and the willingness to give political rationales that are comprehensible. Democracy, in short, is not just elections but the very ethos that sustains the rule of political equals.

Thus, to answer the question of “is there such a thing as too much democracy?”, the value-oriented camp would respond with a strong *no*. Democracy means not just institutions but the values that sustain it. And we can never have enough of them. These are no easy values to uphold and maintain in practice—especially because the self-government of equals involves intense disagreements about what we take to be the most fundamental values, like freedom and dignity. Democracy is no easy task, and there can never be too much of these values.

3. Conclusion

The long debate over whether there is such a thing as too much democracy hinges on how one sets the scope of democracy. In the often-cited procedural definition of democracy, we may say that there is such a thing as “too much” democracy if an over-reliance or undue faith in the procedure itself undermines liberal norms and intensifies partisanship. On the other hand, the expansive definition views democracy as a commitment to political equality, a commitment that requires a strong action-driven ethos. These ethical orientations can never be in excess. There can never be “too much” of the norms that will allow us to live together as political equals. We need much more of it in our polarized, conflict-ridden contemporary world. Democracy is not just a political system but an ideal, a *yardstick* by which to judge our current society, and an *aspiration* to continuously strive towards.

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Explainable Deep Learning for the Prediction and Multi-Classification of Musculoskeletal Fractures: A Comparative Study By Deepak Pathak

Abstract

Traditional methods of diagnosing fractures largely depend on radiographic imaging and clinical expertise, which can be time-consuming and sometimes unreliable. However, a recent surge in the use of machine learning in medicine has improved the diagnosis of bone fractures. This study presents an interpretable approach that compares different deep learning models to identify whether there is a fracture and then classifies it as one of the various musculoskeletal fracture types: comminuted, greenstick, oblique, spiral, and transverse. This study involved two steps: binary prediction and multiclassification of musculoskeletal fractures.

The study involving the binary prediction of musculoskeletal fractures investigated an array of deep learning techniques, such as logistic regression, vanilla neural networks, and convolutional neural networks, using the Bone Fracture Dataset from Kaggle. This dataset, curated by Osama Jalil, contains X-ray images labeled for the presence or absence of fractures, providing a comprehensive resource for training and evaluating machine learning models [1]. Normalization and augmentation were used to enhance the model performance and robustness.

The second step of the study involving the multi-classification of musculoskeletal fractures investigated a more powerful set of object detection algorithms, such as SqueezeNet, MobileNet v3, Resnet 50, and YOLOv9, and aimed to be explainable (indicating where the fracture is believed to be) to assist physicians in confirming the model's prediction. This was made possible through saliency maps and bounding box technology. The dataset used for this step was the Bone OD Dataset from Roboflow, which included comprehensively labeled and annotated X-rays of various musculoskeletal fractures, was essential for training and evaluating the models [2]. However, the dataset was imbalanced and as a result, required augmentation; the models were also unable to pick up on the edges of the bone, leading to the use of filters and edge detection techniques. This study aimed to maximize the validation accuracy of the detection, classification, and localization of musculoskeletal fractures by comparing various powerful deep learning models.

Introduction

A musculoskeletal fracture, also commonly known as a bone fracture, happen due to accidents, sports-related injuries, and conditions that weaken bones like osteoporosis. Musculoskeletal fractures occur frequently because of aging populations and more active lifestyles, as indicated by the yearly average of 6.3 million fractures in the United States [3]. The accurate and quick diagnosis of these fractures is essential to create appropriate treatment plans, and enhance recovery.

There are several types of musculoskeletal fractures (comminuted, greenstick, oblique, spiral, transverse), each with distinct characteristics that require unique treatments. Comminuted fractures involve bone shattering in at least two places and are caused by severe traumas like car

accidents; since major damage is done to bones in comminuted fractures, surgery is usually required [4]. Often occurring in children due to the elasticity of their bones, a greenstick fracture is an incomplete fracture where the bone bends rather than breaking completely [5].

Additionally, an oblique fracture, which is characterized by a diagonal break across the bone, usually results from angulation and axial twisting [6]. A spiral fracture wraps around the bone's shaft, indicative of a twisting force applied to the bone. Finally, a transverse fracture is a straight break across the bone's shaft, typically due to a force applied perpendicular to the bone's axis [7].

Conventional diagnostic techniques depend on medical professionals' knowledge and radiographic imaging. These techniques, however, take a lot of effort and are prone to human mistakes. New developments in machine learning have opened up new avenues for improving the diagnostic procedure. Clinics can benefit from deep learning algorithms that are trained on massive X-ray image datasets since they can quickly and accurately identify fractures; this is because of the innate ability of these models to accurately detect patterns and abnormalities in medical images using complex layers. Through a comparative and explainable method, this study uses deep learning technology to improve the initial detection and then classification accuracy of musculoskeletal fractures, as well as demonstrate the effect that techniques such as augmentation can have on validation accuracy.

Materials and Data Processing (Binary Prediction of Musculoskeletal Fracture)

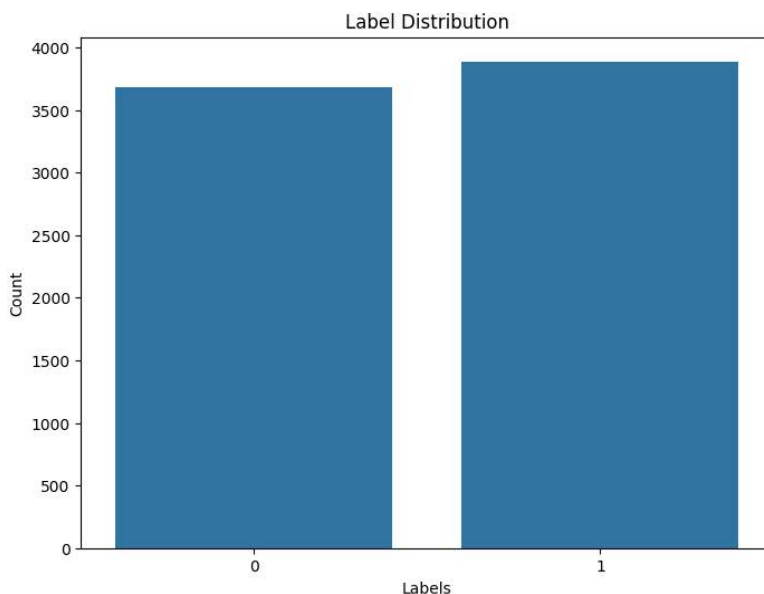
The analysis and usage of the Bone Fracture Dataset by Jalil, Osama for the binary prediction of the fracture demanded strict data preprocessing to prepare the features for strong, and properly-fitted machine learning modeling. This dataset comprises of around 9,000 X-ray images, divided into training and testing folders, with sub-folders for fractured and not fractured images. The original dataset contained no preprocessing, so normalization techniques were manually applied to enhance the performance of the machine-learning models. Although the convolutional neural network (CNN) was initially vanilla, normalization and augmentation were later added to another CNN to demonstrate the effect that these techniques can have on training and validation accuracy.

Normalization is the process of scaling individual data points to ensure that they have a mean of zero and a standard deviation of one. This step is crucial for image data as it helps improve the convergence speed of neural networks, and also ensures that all features contribute equally to the model. In this case, normalization involved scaling the pixel values of the X-ray images from the original range (which was 0 to 255) to a range of 0 to 1.

The labels for the images were one-hot encoded, with `fractured = 0` and `not_fractured = 1`. One-hot encoding converts categorical labels into a binary format that machine learning models can process; this is especially important for neural networks, which require numerical inputs to compute gradients during training. The dataset was split into training and testing sets with their corresponding labels, and `x_train`, `x_test`, `y_train`, and `y_test` were created; The training data (`x_train`) had a shape of (7570, 150, 150, 3) indicating 7570 images with dimensions of 150 x 150 pixels and 3 color channels (RGB). To optimize learning, the 3-color

channel images (RGB) were converted to a single color channel (grayscale) to reduce the number of parameters, and enhance particular patterns for medical images, the relevant information is contained in the intensity of the pixels rather than their color and thus by converting to grayscale, the model focuses on the important features/variations), assist with computational efficiency, and ensure that X-rays are consistent when inputted. The labels for the training data (y_{train}) had a shape of (7570,1), indicating a label for each training image; the testing data (x_{test} and y_{test}) had similar dimensions but contained fewer images due to an 80/20 train-test split.

Figure 1: Label Distribution of Binary Prediction Dataset



The training dataset was balanced with an equal number of fractured and not-fractured images. This balance is essential because it ensures that the model does not become biased towards one class due to an unequal representation of classes in the training data, and may predict that class excessively (and this negatively impacts the validation accuracy).

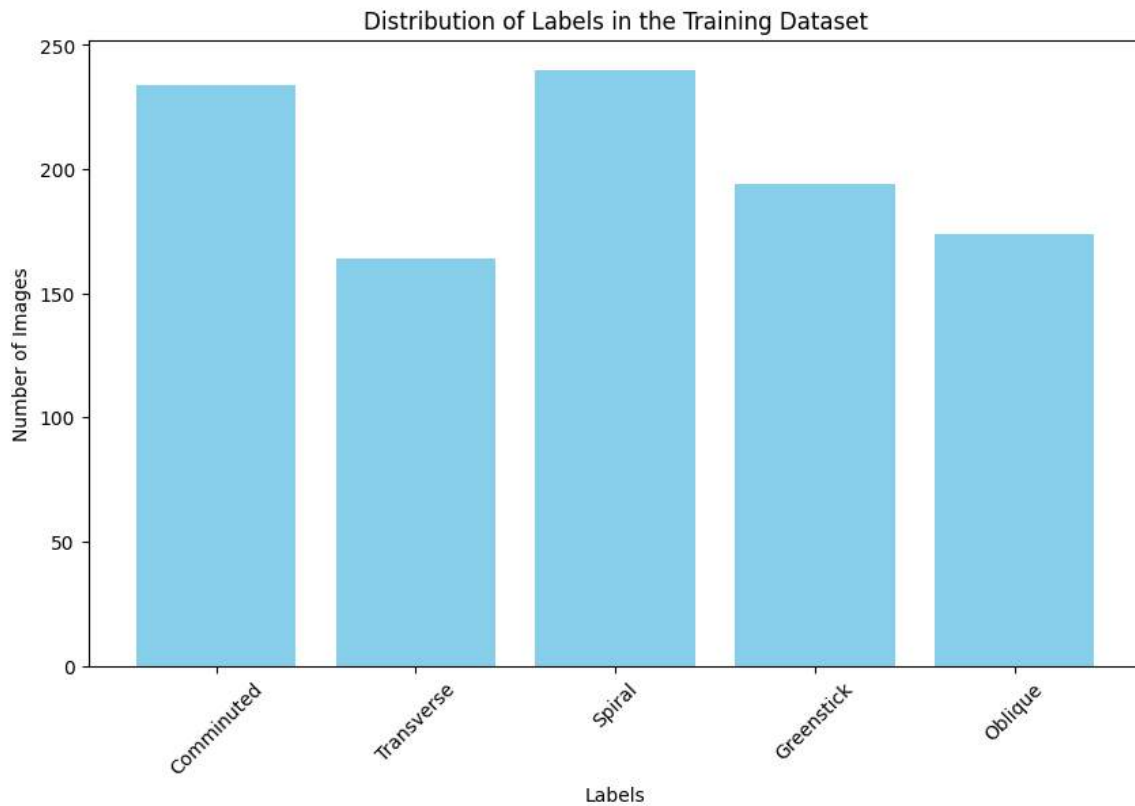
Materials and Data Processing (Multi-Classification of Musculoskeletal Fracture)

The Bone OD Dataset from Roboflow was used to classify musculoskeletal fractures: its 8th version includes 991 X-ray pictures, with 882 in the training set, 97 in the validation set, and 12 in the test set. The images in the dataset were resized to a standard size of 640 x 640 pixels to maintain consistency during training, labeled, and classified into five categories: comminuted, greenstick, oblique, spiral, and transverse; the dataset was also accurately annotated to aid in object detection.

The Bone OD Dataset had a class imbalance in terms of fracture types, which introduced bias to the model to predict some classes more than others. To address this, a combination of data augmentation and redistribution of images between data sets was done. 10 random images of transverse fractures along with their corresponding annotation files were moved from the

validation set to the training set, and 1 transverse image was moved from the testing set to the training set. Similarly, 11 oblique-fracture images were moved from the testing set to the training set, and 10 greenstick-fracture images were transferred from the validation set to the training set. Additionally, these moved images were then subjected to a series of augmentation techniques, which included random horizontal flips (with a 50% probability), rotations between -10 and +10 degrees, scaling between 0.9 and 1.1, horizontal and vertical translations of up to 10%, and brightness adjustments by a random factor between 0.8 and 1.2. Because of these augmentations, the dataset was made more balanced, which is essential for minimizing model bias and enhancing generalization across all classes. The annotations were also updated accordingly to maintain accuracy after augmentations.

Figure 2: Label Distribution of Multi-Classification Dataset Prior to Transfers/Augmentations

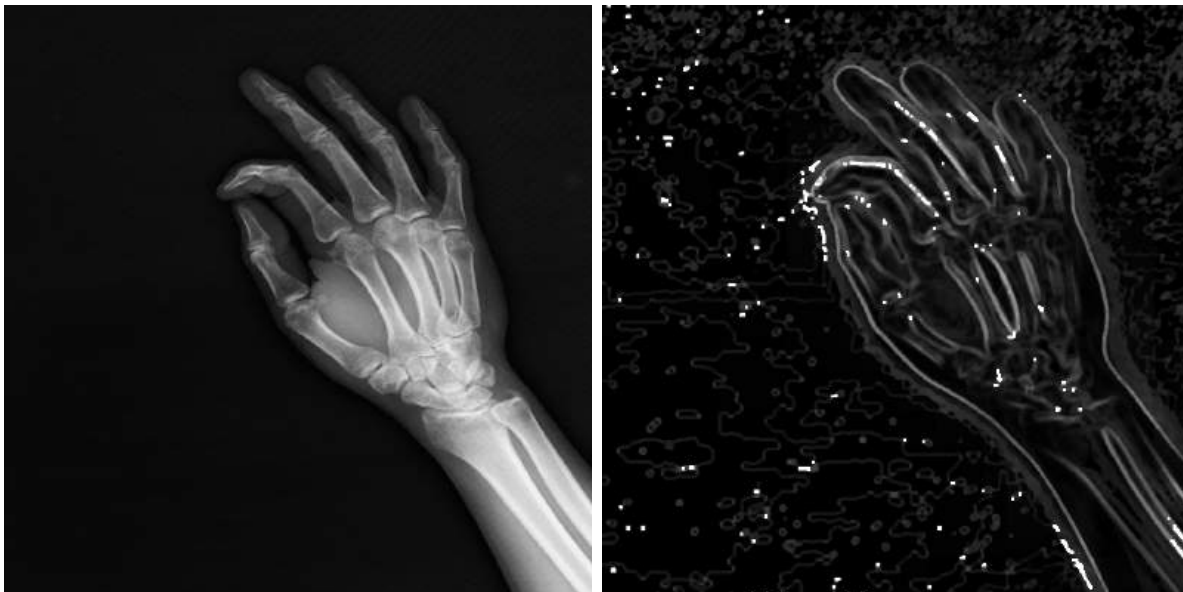


Initially, the study used the unbalanced dataset to begin multi-classification using object detection algorithms. However, it became clear after evaluating the models that they were predicting comminuted and spiral fractures at a much greater frequency than other labels, negatively affecting validation accuracy.

Several preprocessing approaches, including filters and edge detection algorithms, were used to improve the quality of the X-ray images and distinguish the musculoskeletal fractures. First, the images were transformed to grayscale to decrease complexity while keeping the critical information needed for fracture diagnosis. A Gaussian filter was used to decrease noise, which is necessary in medical imaging to prevent misinterpretation. This was followed by a median filter to remove the salt-and-pepper noise that is prevalent in X-ray pictures. To preserve edges while further lowering noise, a bilateral filter was utilized, ensuring that critical features such as fracture edges were not blurred.

Next, histogram equalization was used to increase the contrast of the pictures, highlighting small changes in bone structure. This was followed by adaptive histogram equalization, which modifies the contrast locally to improve fracture visibility. Sobel edge detection was then employed to highlight the edges within the images, capturing gradient changes that are indicative of fractures. To complement Sobel, Canny edge detection was applied, which is effective in detecting a wide range of edges, providing a more comprehensive outline of potential fracture sites. The outputs of Sobel and Canny edge detection were combined, and then morphological techniques were used to refine the edges by closing small gaps and eliminating noise. The modified images were then saved, overwriting the original files to ensure that the dataset was ready for the next steps in the modeling process.

Figure 3: The Effect of Filters and Edge Detection Techniques



This set of preprocessing techniques was crucial in preparing the X-ray images for analysis since it improved fracture visibility and reduced the likelihood of false positives during classification.

Methods (Binary Prediction of Musculoskeletal Fracture)

The logistic regression model was implemented using ‘LogisticRegression()’ from the sklearn library. This model requires the input images to be flattened into one-dimensional vectors containing pixel values because logistic regression operates on linear feature spaces. In the study, this was achieved by reshaping the image data from its original multi-dimensional form to a single continuous vector for each image. Logistic regression works by finding a linear boundary that separates the two classes (fractured vs. not fractured) in the feature space. It calculates the probability of an instance belonging to a particular class and uses a threshold (which for this study was 0.5) to make a binary decision. While logistic regression is simple and interpretable, it is not the best fit for complex image data. Medical images, like X-rays, require very sophisticated pattern recognition capabilities that logistic regression may not accurately capture. The model was then trained using maximum likelihood estimation, adjusting the weights to minimize the binary cross-entropy loss function. The performance of the model was then evaluated using accuracy, classification reports, and confusion matrices to understand its predictive capabilities (which were also used for all the following models).

A vanilla neural network was implemented using the ‘Sequential()’ model from the sklearn library for binary prediction of musculoskeletal fractures. This model also required the input images to be flattened into one-dimensional vectors. Vanilla neural networks consist of fully connected layers where each neuron is connected to every neuron in the subsequent layer. They can capture non-linear relationships in the data through activation functions like ReLU and Sigmoid; to ensure that no overfitting occurs, dropout layers were added throughout the model’s architecture. While more flexible and powerful than logistic regression for non-linear data, vanilla neural networks may still not be optimal for image data, especially when spatial hierarchies are important. Thus, they did not provide high accuracy for the binary prediction in comparison with more sophisticated models such as convolutional neural networks.

The convolutional neural network models (both the vanilla and augmented versions) were built using TensorFlow, allowing for the easy customization of architectures by adding layers and adjusting hyperparameters to best promote accuracy. The model required the input images to retain their original shape (150, 150, 1) to capture spatial relationships through kernels. The convolutional neural network models used convolutional layers to detect features through filters, followed by pooling layers to reduce dimensionality and retain important features, as well as dropout layers to ensure there was no overfitting. Hyperparameter adjustments were made to optimize the number of kernels, kernel sizes, and dropout rates to maximize the accuracy of these models. The augmented convolutional neural network leveraged Tensorflow’s ‘BatchNormalization()’ to ensure consistent activations, and Tensorflow’s ‘ImageDataGenerator’ to augment the data with a rotation range of 40, a zoom range of 0.2, horizontal and vertical flips. Augmented convolutional neural networks are highly effective for medical images, as they can leverage the increased data variability, and capture more intricate patterns due to their ability to

create feature maps. The augmented convolutional neural network model was trained on the most epochs, promoting more learning; the other models would have reached a higher validation accuracy than they had for this study if trained on a similar amount of epochs.

Figure 4: Architectures of Models For Binary Prediction

Feature	Logistic Regression	Vanilla Neural Network	Vanilla CNN	Augmented + Normalized CNN
Model	Logistic Regression	Neural Network	Convolutional Neural Network	Convolutional Neural Network
Optimization Type	L-BFGS	Adam	Adam	Adam
Normalization	StandardScaler	StandardScaler	None	Batch Normalization
Loss Function	Binary Cross-Entropy	Binary Cross-Entropy	Binary Cross-Entropy	Binary Cross-Entropy
Input Shape	(22500,)	(22500,)	(150,150,1)	(150,150,1)
Layers	Single Linear Transformation	Dense (512), Dropout (0.5), Dense(256), Dropout (0.5), Dense(1)	Conv2D (64), MaxPooling2D, Conv2D (64), MaxPooling2D, Dropout (0.3), Conv2D (128), MaxPooling2D, Dropout (0.2), Flatten, Dense (512), Dropout (0.5), Dense (1)	Conv2D (64), BatchNormalization, MaxPooling2D, Conv2D (64), BatchNormalization, MaxPooling2D, Dropout (0.3), Conv2D (128), BatchNormalization, MaxPooling2D, Dropout (0.2), Flatten, Dense (216), BatchNormalization, Dropout (0.5), Dense (1)
Activation	Sigmoid	ReLU for hidden, sigmoid for output	ReLU for hidden, sigmoid for output	ReLU for hidden, sigmoid for output
Augmentation	None	None	None	Rotation, Zoom, Flip, Shift
Additional	One-hot encoding, Requires Flattening	One-hot encoding, Requires Flattening	Requires one-hot encoding, conversion from RGB to grayscale	Requires one-hot encoding, conversion from RGB to grayscale, Batch Normalization

Methods (Multi-Classification of Musculoskeletal Fracture)

The PyTorch library was used to fit, train, and validate the object detection models (SqueezeNet, MobileNet v3, ResNet50, YOLOv9). These models were made to fit this specific dataset, which had to undergo prior augmentation and had been organized into .jpg images and .xml annotation files. The 'custom_data.yaml' file was created to integrate this data into the training. By being pre-trained on ImageNet, the models took advantage of transfer learning. This file contained the necessary paths to the images and label folders for both the training and validation sets. The dataset was classified into five categories (comminuted, greenstick, oblique, spiral, and transverse). For object detection tasks, however, an additional class labeled '___background___' was included. The extra class is extremely important for object detection models since it represents areas in the image that don't belong to any of the target classes, helping the model tell between the object and the background of the image. A 'train.py' script was utilized, with the 'custom_data.yaml' file being used, and the number of epochs and batch size being specified.

The SqueezeNet model was implemented inside a Faster R-CNN framework using the PyTorch library, chosen because of its simple architecture that balances computational efficiency and accuracy [8]. The last convolutional layer of 'SqueezeNet1_0', which outputs 512 channels, was used as input for the region proposal network (RPN) within the Faster R-CNN model. Then, the region proposal network was configured with an anchor generator that used five different sizes and three different aspect ratios, resulting in a total of 15 anchors. This setup was essential as it captured the varying scales and shapes of fractures in the X-ray images. In addition, the 'MultiScaleRoIAlign' operation was used to pool the feature maps to a fixed size, preserving the spatial alignment of features.

MobileNet v3, made for mobile and embedded vision applications, is known for its simple architecture which is necessary for handling large-scale medical imaging tasks like bone fracture detection [9]. The model's architecture efficiently processes the images while maintaining performance through its convolutions that split depth and spatial operations. The final layer of the MobileNet v3 backbone outputs feature maps that are fed to the region proposal network (RPN) in the Faster R-CNN model.

The ResNet 50 model, which has a deeper architecture than both the SqueezeNet and MobileNet v3, balances both depth and performance through residual learning [10]. ResNet 50, with its 50 layers, is particularly effective for recognizing complex patterns, which makes it a strong candidate for medical imaging tasks like bone fracture detection. This backbone architecture is designed to tackle the vanishing gradient problem, enabling the model to learn more effectively from large datasets by allowing gradients to flow more easily through the network. Similar to the MobileNetv3, The output from the ResNet 50 backbone was passed into the region proposal network (RPN) within the Faster R-CNN model.

The YOLOv9 model was implemented using the Ultralytics YOLO framework, which is different from the other models as the model doesn't use Pascal VOC data [11]. Instead, the

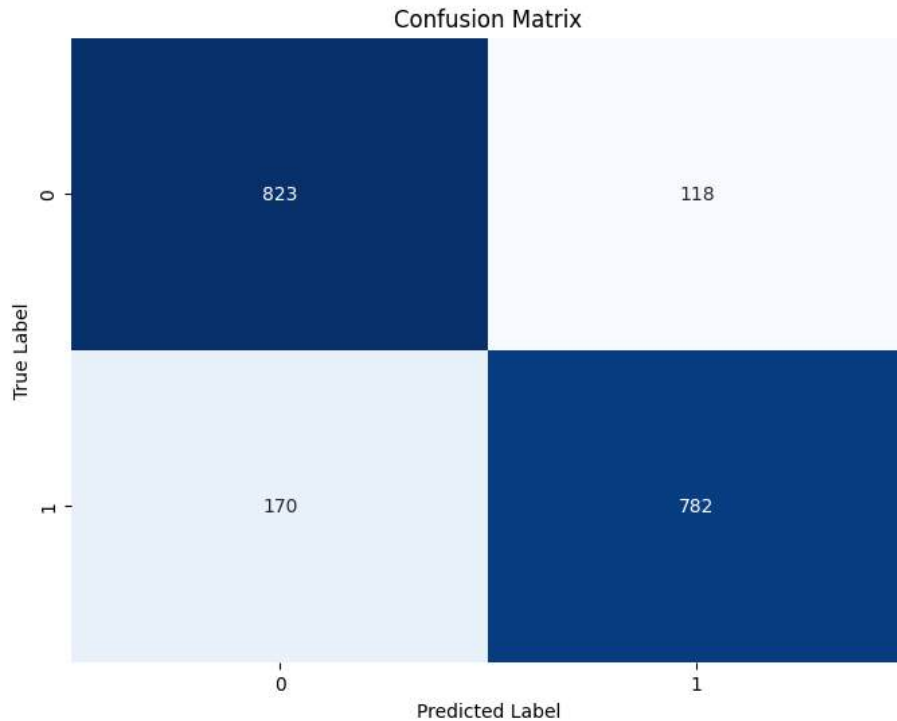
YOLOv9 model separated the data into image folders and label folders, with the label folders containing '.txt' files containing the number representing the class and the bounding box annotation. The YOLOv9 model utilized the COCO dataset for pre-training, reflecting its robust capabilities in object detection tasks. Ultralytics YOLO facilitated the integration of pre-trained weights, which were crucial for achieving effective bone fracture classification. Customization was achieved through a tailored 'data.yaml' file, enabling the model to be adapted specifically for this classification task. The training process involved running the model for an initial 100 epochs, after which the best-performing weights were saved. The model was then retrained for an additional 100 epochs using these saved weights, and this cycle was repeated to iteratively optimize validation accuracy and minimize losses. The flexibility to update the weights directory path allowed for effective management of model training and fine-tuning, ensuring improved performance on the bone fracture classification task.

Results (Binary Prediction of Musculoskeletal Fracture)

The logistic regression model achieved a test accuracy of 84.7%, demonstrating a strong performance in distinguishing between fractures and non-fractures. The precision for fractures (Class 1) was 0.87, demonstrating that 87% of the instances predicted as fractures are correctly identified as such; this suggests that when the model predicts a fracture, it is highly reliable. The recall for fractures was 0.82, which means the model successfully identifies 82% of actual fractures. Furthermore, the F1-score for fractures is 0.84, which is the harmonic mean of precision and recall, reflecting a balanced performance between these two metrics. For non-fractures (Class 0), the precision was 0.83, showing that 83% of the predictions for non-fractures are accurate, and the recall was 0.87, indicating the model correctly identifies 87% of true non-fracture cases. The F1-score for non-fractures was 0.85, also highlighting a good balance between precision and recall.

The model's relatively high precision and recall values suggest that it is effective at both identifying and verifying fractures and non-fractures. Its high precision and recall indicate that the model has successfully captured the underlying patterns in the data, likely benefiting from well-chosen features and effective training on a balanced dataset. However, the presence of 170 false negatives (as demonstrated by the confusion matrix) indicates that the model occasionally fails to detect true fractures, which could be critical in medical contexts where missed detections might impact patient outcomes.

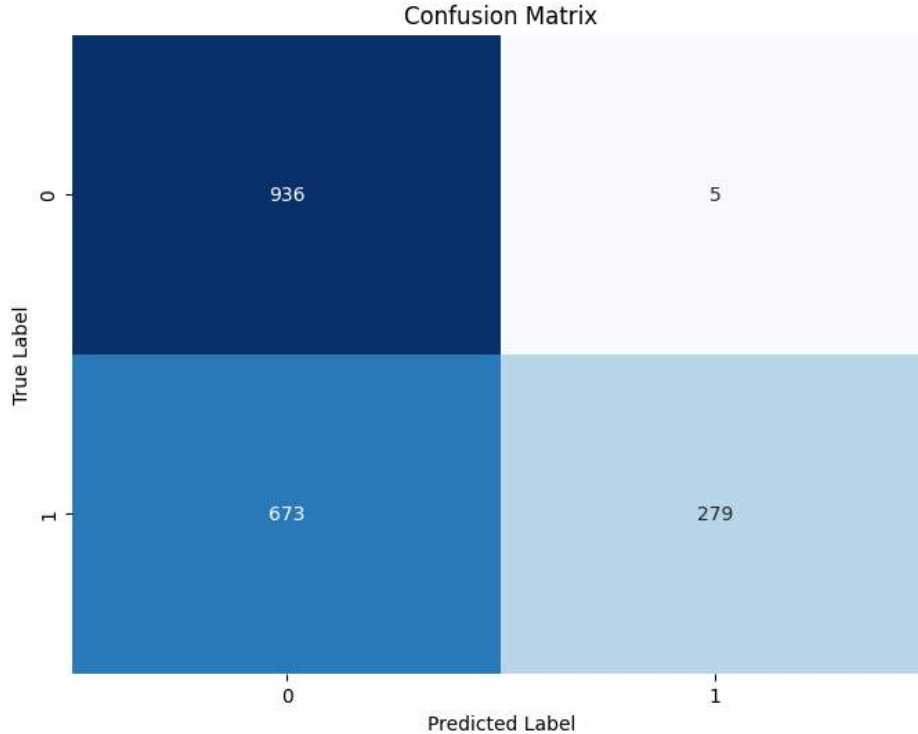
Figure 6: Confusion Matrix of Logistic Regression Model



The results for the vanilla neural network in the binary prediction of bone fractures indicate mixed performance. The model achieved a test accuracy of 64.2%, but this metric alone does not fully capture the model's performance. The precision, recall and F1 score values reveal a significant imbalance in the model's ability to classify the two classes. For class 0 (non-fracture), the precision was 0.58, the recall was 0.99, and the F1 score was 0.73. This high recall suggests that the model was very good at identifying non-fracture cases but is less reliable in its precision. For class 1 (fracture), the precision was very high at 0.98, but the recall was only 0.29, resulting in a low F1 score of 0.45. This disparity indicates that while the model was very accurate when it came to predicting a fracture, it failed to identify a substantial number of actual fractures, showing a clear imbalance in classification performance.

The confusion matrix supports these findings: the high number of false negatives indicates that the model struggles to detect fractures, despite its high precision for positive cases. The low recall for fractures, paired with a relatively high number of false negatives, suggests overfitting or that the model has difficulty generalizing from the training data to accurately predict fractures in unseen data. Overall, while the model exhibits strong precision for fractures, its poor recall and accuracy demonstrate a need for further refinement, potentially through more advanced architectures meant for image data.

Figure 7: Confusion Matrix of Vanilla Neural Network

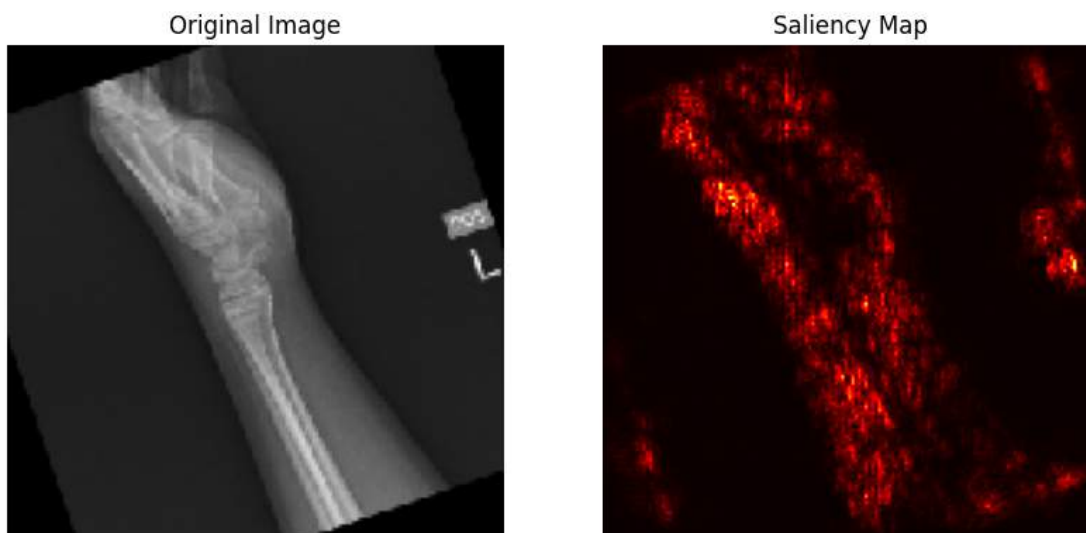


The confusion matrix gave deeper insights into the prediction patterns of the vanilla neural network. The model almost always predicts that the image is a non-fracture (label 0), indicating a bias due to a lack of data and a simple architecture.

The vanilla CNN model, which was trained without any augmentations or normalization techniques, performed remarkably well, achieving a test accuracy of 97%. The precision, recall, and F1 scores for both classes were consistently high at 0.97, indicating that the model effectively identified and classified the data with minimal errors. This high level of performance can be attributed to the model's architecture and its ability to capture essential features from the dataset. However, the absence of data augmentation and normalization may limit the model's robustness, potentially making it sensitive to variations in the input data that it was not exposed to during training.

The augmented CNN model, which incorporated data augmentation and normalization techniques, showed an improvement in performance with a test accuracy of 99.21%, a low loss of 0.0677, and a validation loss of 0.0180. The slight discrepancy between the training accuracy (97.65%) and validation accuracy (99.21%) suggests that the model is well-generalized without significant overfitting, as the validation performance is superior. The low loss values indicate that the model effectively minimized errors during training, and the incorporation of augmentations likely helped the model to become more resilient to variations, leading to better generalization and improved performance on unseen data.

Figure 8: Saliency Map of Convolutional Neural Network: Interpretation of Fracture



The saliency map of the bone fracture X-ray highlights the areas that the CNN considered most significant for its prediction, showing which parts of the image contributed most to the model's identification of the fracture.

Results (Multi-Classification of Musculoskeletal Fracture)

The results of the multi-classification task using SqueezeNet, MobileNet v3, ResNet50, and the YOLOv9 models were clear from using metrics such as 'train class loss', 'train box regression loss', and 'train object loss', which assess the models' abilities to classify the fractures, then accurately localize them, and identify regions of interest. Additionally, mean Average Precision (mAP) at IoU thresholds of 0.5 and 0.5:0.95 were calculated to determine the models' overall detection accuracy across varying degrees of localization precision. These metrics provided a comprehensive evaluation of each model's performance in classifying and localizing musculoskeletal fractures.

Furthermore, by effectively comparing the training losses to the mean average precision across varying IoU thresholds, patterns and limitations of the models can be derived. Furthermore, analyzing metrics such as average recall (AR) gives further insights into what bone fracture classes the models can accurately classify.

Figure 9: Performance Metrics of Models for Musculoskeletal Fracture for Multi-Classification

Model	mAP@0.5	mAP@0.5:0.95	Train Class Loss	Train Box Reg Loss	Train Obj Loss
SqueezeNet	0.321	0.11	0.059	0.057	0.040
MobileNet v3	0.374	0.133	0.12	0.00257	0.00248
ResNet 50	0.380	0.191	0.021	0.043	0.00062
YOLO v9	0.479	0.178	0.59	0.68	N/A

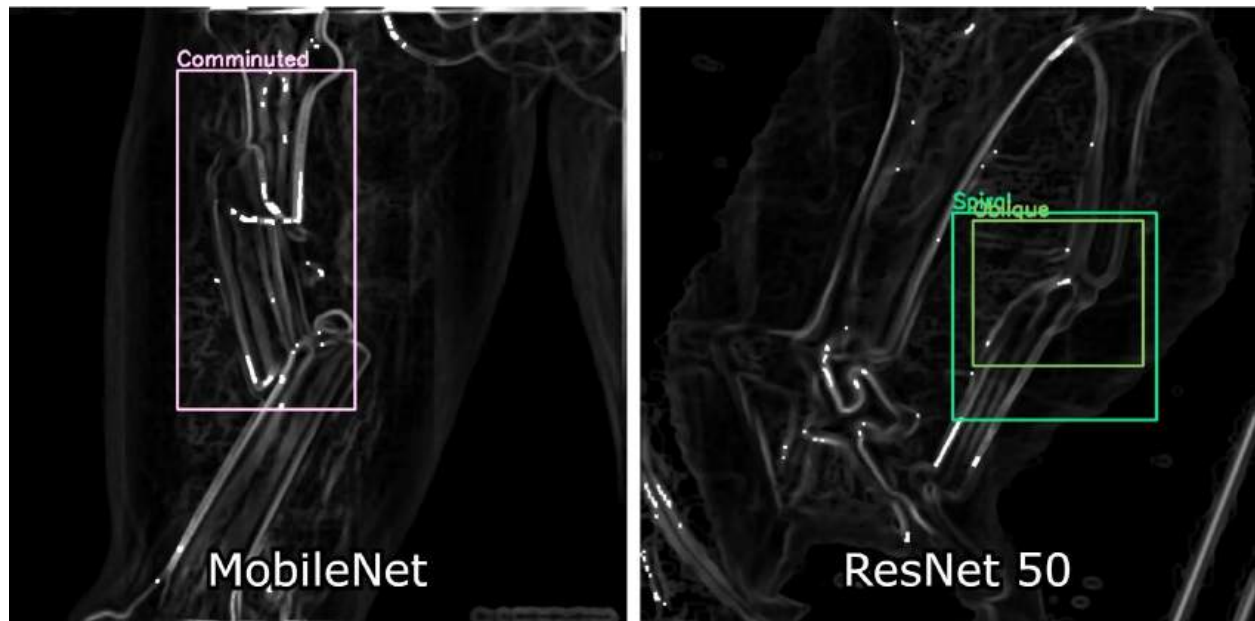
The SqueezeNet model yielded a mean average precision (mAP) of 32.1% at an IoU threshold of 0.5, showing moderate accuracy in the detection and classification of a musculoskeletal fracture; The mAP@0.5:0.95 was 11%, however, suggesting that the model found it difficult to precisely localize the fracture across varying IoU thresholds. By comparing the moderate accuracy of the mAP@0.5:0.95 and mAP@0.5 to the low training accuracy, it became apparent that overfitting may have occurred in the SqueezeNet model; the overfitting and the inability to precisely localize the fracture can be attributed to the small amount of data provided during the training phase. The SqueezeNet has the most simple architecture of all the models for the multi-classification task, which explains why it has the lowest mAP@0.5 and mAP@0.5:0.95.

The MobileNet v3 model yielded a mean average precision (mAP) of 37.4% at an IoU threshold of 0.5, showing a greater ability to detect and classify musculoskeletal fractures than the SqueezeNet. Additionally, the mAP@0.5:0.95 was 13.3%, which demonstrated a slight improvement in precisely localizing the fracture across varying IoU thresholds. Similar to SqueezeNet, the MobileNet v3 had a low training accuracy relative to the mean average precision; this can be attributed to overfitting caused by a lack of sufficient data. However, the training class loss of the MobileNet v3 was 0.12, indicating that there was less overfitting when it came to determining the class of the musculoskeletal fracture than the SqueezeNet model. The average recall (AR) results for the MobileNet model gave further insight into the varying levels of detection for specific bone fracture classes; comminuted fractures, with an average recall of 0.186, were identified at a higher capability than the other fracture classes. On the other hand, oblique fractures were the most difficult to identify, having an average recall of 0.074.

The ResNet 50 model had a mean average precision (mAP) of 38.0% at an IoU threshold of 0.5, demonstrating a slightly increased accuracy in detecting and classifying musculoskeletal fractures than the MobileNet v3 model. The mAP@0.5:0.95 was 19.1%, demonstrating the highest accuracy between the deep learning models when it came to precisely localizing the fractures across varying IoU thresholds. The ResNet 50 model was most likely overfitted, as demonstrated by the discrepancy between the low training loss and the mean average precision. Despite the limitations of the dataset following augmentation regarding a lack of sufficient data,

the ResNet 50 was still able to achieve a respectable mean average precision; with a larger dataset, the ResNet 50 could achieve a much greater accuracy when it comes to the classification and localization of musculoskeletal fractures.

Figure 10: MobileNet vs ResNet 50: Bounding Box Highlighting Fracture Class and Region

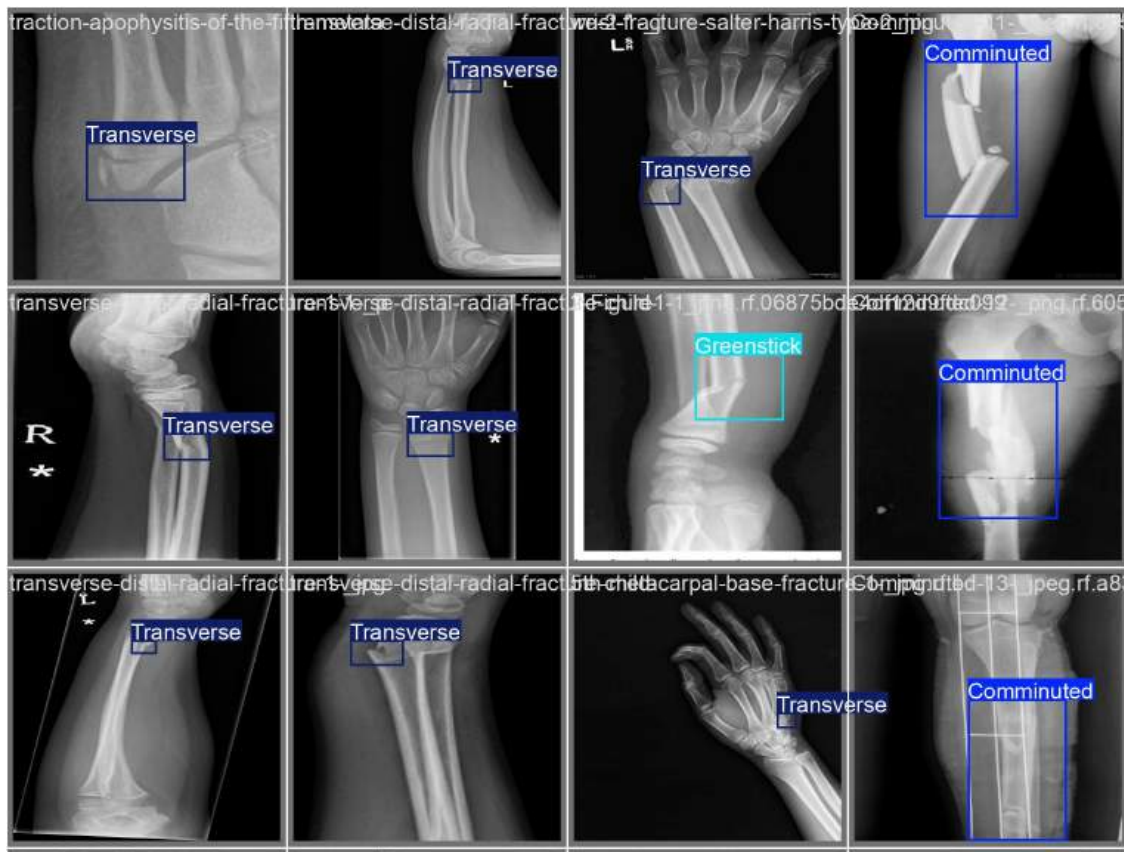


Each model identifies and localizes the fracture regions within the X-ray images, showcasing the differences in detection accuracy and precision between the two architectures.

The YOLOv9 model had a mean average precision (mAP) of 47.9%, demonstrating the highest accuracy between the deep learning models when it came to the detection and classification of the fracture. The mAP@0.5:0.95 was 17.8%, however, showing a lower accuracy in precisely localizing the fracture across varying IoU thresholds than the ResNet 50. Unlike the other models, the YOLOv9 model had training losses that reflected the mean average precisions; these results demonstrate that overfitting, while still being a limitation, was able to learn and perform more accurately than the previous models. Therefore, the YOLOv9 had the most potential in terms of accurately detecting, classifying, and localizing musculoskeletal fractures and can be improved with more iterations and increased data quantity and quality.

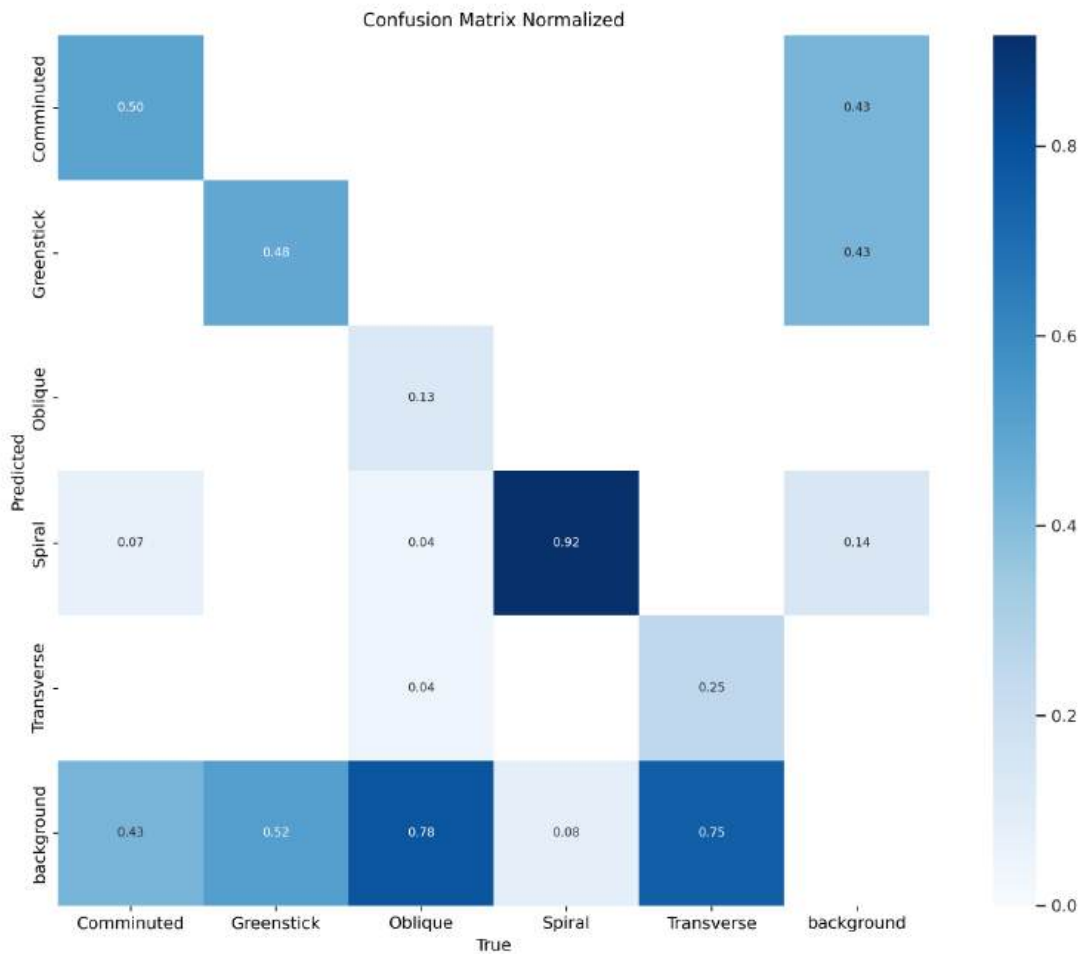
The YOLOv9 model achieved a precision of 0.59, indicating that 59% of the predicted fractures were correctly identified as fractures; this suggests that the model was reasonably accurate at avoiding false positives. Furthermore, the model had a recall of 0.47, showing the model can identify nearly half of the actual fractures present in the dataset. While there is a need for improvement, the performance of the model suggests that the model can learn relevant features and can be further optimized, and is simply limited by the amount of data it is being trained on.

Figure 11: YOLOv9: Musculoskeletal Fracture Classification and Localization



This figure demonstrates the predictions made by the YOLOv9 model for classifying different types of musculoskeletal fractures in X-ray images. Each X-ray image in the figure has a bounding box around the area where the model detected a fracture, and the predicted classification is labeled within the box; the bounding box allows for the prediction to be explainable, as it gives the overall location where the model believes there is a fracture. This is important as it allows medical professionals to verify the predictions of the model. Of the 12 X-ray images provided, the fracture types that were identified the most were “Transverse” and “Comminuted”.

Figure 12: YOLOv9: Confusion Matrix With Normalized Values



The confusion matrix of the YOLOv9 gives insight into the prediction patterns of the model. The model struggled most when it came to differentiating the background and oblique fractures, indicated by a value of 0.78; it also struggled with differentiating transverse fractures with the background, possibly a result of transverse fractures being much more complex visually than the other fractures. The matrix demonstrates that the model was able to tell that a fracture was comminuted 50% of the time, while oblique fractures were only correctly identified 13% of the time. While the YOLOv9 model struggled with oblique and transverse fractures the most, it excelled when it came to predicting and classifying spiral fractures; this is indicated by a high value of 0.92. By analyzing the confusion matrix, it became apparent that the model could correctly identify spiral fractures, and had moderate accuracy for both comminuted and greenstick fractures. To improve this model, it is essential to have more data for the classes that are being predicted the least, such as oblique and transverse fractures.

Discussion and Analysis

This study demonstrates that advanced machine learning algorithms, can effectively predict, classify, and locate musculoskeletal fractures in X-ray imaging. For the binary prediction of the musculoskeletal fractures, the augmented convolutional neural network achieved the highest accuracy of 99.21%; it outperformed the 84% accuracy of logistic regression, 64.2% accuracy of the vanilla neural network, and 97% accuracy of the vanilla convolutional neural network. In the context of the study, this result was expected as convolutional neural networks are configured for image data, while the other models require flattening and are unable to pick up on patterns.

The lack of high-quality data did not limit the models meant for the prediction of musculoskeletal fractures as much as the other models that had the responsibility of both classifying and localizing the fractures. This is evidenced by the fact that overfitting did not occur with the logistic regression, vanilla neural network, or convolutional networks: this conclusion was made due to the fact that the training loss and accuracy were similar to the validation loss and accuracy, demonstrating that the model was truly picking up on patterns and learning. By using a saliency map, it became clear which parts of an input image are most influential in a model's decision-making process; the map showed that the model's decision was more influenced by the outline of the fracture, and not necessarily the edges. This prompted the use of filters and edge detection techniques that would enhance the model's ability to recognize the crucial fracture details.

Additionally, the dataset used for the multi-classification and localization of musculoskeletal fractures was initially imbalanced; this negatively affected the performance of the models. To demonstrate this, this study trained a ResNet50 on the unbalanced data and achieved an average precision (AP) of 0.089 and an average recall (AR) of 0.174. After balancing the dataset, these metrics improved substantially, with the average precision rising to 0.111 and the average recall jumping to 0.207. This illustrated the importance of balancing datasets when trying to achieve reliable multi-classification results.

The results of the multi-classification and localization of musculoskeletal fractures varied: while the ResNet 50 had the greatest mAP at IoU thresholds of 0.5:0.95, the YOLOv9 had the greatest mAP at the IoU threshold of 0.5. In other words, the ResNet 50 had the greatest accuracy when it came to accurately determining the location of the fracture, while the YOLOv9 model had the greatest accuracy when it came to detecting and classifying the fracture.

By leveraging saliency maps and bounding box technology, the models used in bone fracture classification became more explainable, allowing medical professionals to verify predictions. Saliency maps help visualize which areas of an image the model focuses on while bounding boxes highlight the exact location of detected fractures. This explainability is crucial for doctors, as it enables them to cross-check the model's predictions against their assessments, ensuring that the correct fracture is identified and the appropriate treatment is administered.

The growing use of machine learning in medical imaging, notably for the classification and localization of musculoskeletal fractures, represents a substantial advancement over previous

CAD systems. Historically, CAD systems depended on traditional machine learning algorithms that extracted features and recognized illness patterns using domain-specific image analysis techniques. However, this approach was frequently constrained by the developers' capacity to translate complicated illness patterns into a finite collection of characteristics, resulting in difficulties with generalizability and a high false positive rate when sensitivity was prioritized [12].

In contrast, deep learning has revolutionized this field by automating feature extraction and learning from large datasets. Deep learning models can process vast amounts of imaging data, discovering feature representations that are more selective and invariant than hand-engineered features. This capability not only improves the accuracy of detecting and classifying fractures but also enhances the robustness of models against variations in medical imaging data. However, while deep learning offers significant advantages, including high selectivity and the ability to learn from extensive datasets, it also presents challenges such as the need for large, diverse datasets and the risk of overfitting. Despite these limitations, the application of deep learning in medical imaging holds great promise for improving diagnostic accuracy and supporting clinical decision-making in musculoskeletal fracture classification and beyond.

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Exploring Hemophilia: A Literature Review of Evolving Treatments and Therapeutic Advancements By Eleanor Day and Gianna Darmo

Abstract

Hemophilia, which is a genetically inherited blood clotting disorder, has two main types. Hemophilia A, which is more common, and Hemophilia B currently have been different experimental treatments which aim on reducing the severity of the disease. One of these treatments, gene therapy, could prove a definitive cure to the disease. Gene therapy includes removing the hemophilia-causing regions of one's DNA, and then inserting the normal sequence. This paper will discuss the background and current treatments of Hemophilia, along with a discussion of the use of the rapidly growing technology associated with the gene therapy and its potential implications and costs.

Background

Severity

Hemophilia A and B are blood clotting disorders that account for about a 15 year median lifespan decrease in severe cases (Darby et al. 2007). According to a UK based retrospective study, severe hemophilia mortality rate increased by a factor of 2.69 (in a male population) as age increased, compared to an increase by a factor of 1.19 for mild to moderate hemophilia (Darby et al. 2007). There are many factors that determine how severe a case of hemophilia is, but in general, the disorder's severity is determined by plasma levels, where <1% of mutated coagulation factors corresponds to a normal phenotype, 1-5% is moderate, and 5-40% is considered severe (Castaman and Mantino, 2019). Mild, moderate, and half the cases of severe hemophilia A are caused by heterogeneous mutations on the F8 gene while 45-50% of severe hemophilia A cases are caused by an intron 22 inversion mutation followed by the intron 1 inversion mutation (S Keeney. et al. 2005). There are more than 1200 F9 gene variants that can cause hemophilia B, consisting of approximately 71.9% point mutations which correspond to a milder phenotype and 28.1% frameshift which can cause a more extreme phenotype (Journal of Transfusion Medicine 2022, vol. 15, no. 2).

Prevalence

Hemophilia A is more common than Hemophilia B, occurring in 1/5000 male births compared to 1/30,000 male births in Hemophilia B (Castaman and Mantino, 2019). In high income countries, hemophilia A occurs in $12.6 \pm 6/100,000$ males, compared to $6.6 \pm 4.8/100,000$ males for the other countries (Stonebraker et al. 2009). In comparison, the prevalence rate for Hemophilia B was generally lower, occurring in $2.69 \pm 1.61/100,000$ males in high-income countries vs $1.20 \pm 1.33/100,000$ males in lower-income countries (Stonebraker et al. 2011). Evidently, Hemophilia A appears to be slightly more common than Hemophilia B, though both are still fairly rare.

Mortality Rates

In post 2010 studies, the leading cause of death for hemophiliacs is unknown/other, but is followed by hemorrhages or thrombosis in most cases (J Thromb Haemost, 2021). However, these studies found many different percentages of blood-related deaths, varying from 5.9% in a US-based study to 52.6% in a Korea-based study (J Thromb Haemost, 2021). Another study from Brazil found that hemophilia directly caused about 45.09% of deaths among male hemophiliacs, and it was an associated cause of death in about 54.91% of cases (Santo, 2021). In this sample, about 50% of deaths occurred before 35 where hemophilia was the direct cause, and about 50% of death occurred before 41 in cases where hemophilia was an associated cause. The significantly reduced life expectancy indicates the extreme need for a clear understanding of the types of hemophilias, and what causes them. This paper will examine the history, biological aspects, along with the current treatments and prospective treatments for both Hemophilia A and Hemophilia B.

Genetic Inheritance

About 70% of both types hemophilia is X-linked and recessive, which leads them to be generally more prevalent in males than females (López-Arroyo JL, Pérez-Zúñiga JM, Merino-Pasaye LE, Saavedra-González A, Alcivar-Cedeño LM, Álvarez-Vera JL, et al 2021). However, the other 30% of cases are caused by spontaneous *de novo* mutations, which do not follow a simple inheritance pattern and cannot be simply traced (López-Arroyo JL, Pérez-Zúñiga JM, Merino-Pasaye LE, Saavedra-González A, Alcivar-Cedeño LM, Álvarez-Vera JL, et al 2021). In X-linked recessive inheritance, the probability of the offspring inheriting the disease is fairly simple to calculate. Since the disease is located on the X chromosome and is recessive, females need both a hemophiliac father and a carrier/affected mother. However, males only need an affected or carrier mother, as they receive a Y chromosome from their father. Therefore, females are more likely to be carriers of the disease, while males can never be carriers and are more likely to be affected.

Types of Mutations

There are many different types of mutations that can cause varying phenotypes in both Hemophilia A and B. For example, in Hemophilia B, there are over 1000 pathogenic mutations of the FIX gene that can cause disease (Miller, 2021).

In general, the mutations to the coagulation factors slow or prevent the typical coagulation cascade in blood clotting. For non-synonymous mutations, the primary structure of the factor is affected which leads to a more mild phenotype (Simhadri V, Hamasaki-Katagiri N, Lin B et al. 2007). Synonymous mutations affect the translation of the factors by slowing them down and altering its quaternary structure, resulting in decreased extracellular protein levels and a more severe phenotype (Simhadri V, Hamasaki-Katagiri N, Lin B et al. 2007). In addition, both FVIII and FIX are vitamin K dependent glycoproteins that are synthesized in the liver and then

further modified before being secreted into the blood, where they can be activated in blood clotting (Shen G, Gao M, Cao Q et al. 2022).

History and Progression

Hemophilia has a long history, reflecting advancements in treatment and understanding scientifically. Spanning over centuries, this disorder has driven improvements in diagnosis and management through clinical practice and scientific research.

Ancient Time Period

The disease was first recognized in ancient times. Early texts, including passages from the Greeks and Romans, discuss potential hemophilia symptoms even if the accounts likely did not have a complete understanding of the disorder. At this time, the disease was associated with excessive or spontaneous bleeding in specific families, implicating that a hereditary role existed in its transmission (Goudemand et. al, 2009).

19th Century

The word “hemophilia” was first used in the 19th century, derived from the words “haima” and “philia” meaning “blood” and “affinity” in Greek (respectively). The word reflected the nature of the bleeding disorder, as hemophilia is associated with excessive bleeding. In the 19th century, more documentation of the disease was found within royals (like the House of Windsor). This earned hemophilia the nickname of the “Royal Disease” because of its prevalence in royal families. While the tendency of hemophilia to run in bloodlines at this time did allow for the hereditary aspect of the disorder to be implied, the cause for hemophilia was not discovered at this time (Horton, 1998).

Early 20th Century

In the 20th century, advanced understanding of hemophilia was found with the inclusion of genetic research. Hemophilia was found to be a sex-linked genetic disorder and inherited in a recessive X-linked pattern. This meant that men (who only had one X chromosome) were more likely to be affected by hemophilia than females (who have two X chromosomes). Further, females were found capable of being carriers, being able to carry both a dominant and recessive gene for the disorder without exhibiting symptoms (Mannucci, 2008). The insight gained in this time period allowed for the future development of treatment and diagnostic tools for hemophilia.

Mid-20th Century

In the mid-20th century, improvements were made in hemophilia diagnosis and treatment. Blood transfusions and early clotting factor concentrates, introduced in the 1960s/1970s, helped improve the quality of life for hemophilia patients, helping manage the frequency and severity of bleeding episodes (Mannucci, 2008). However, these measures for management presented risks for infection like hepatitis C and HIV (from blood contamination). Another advancement in

hemophilia treatment, recombinant factor concentrates, presented a way to manage hemophilia without the risk of infectious diseases (Franchini et. al, 2011).

Genetic Research

Over time, advancements in hemophilia treatment and management have been focused on personalized treatments. Genetic research has led to the use of gene therapy in treatment to target the genetic defects that result in hemophilia symptoms. Gene therapy has shown promise in long-term treatment of hemophilia (Angrini et al., 10).

As time has progressed, advancements in the diagnosis, treatment, and management of hemophilia have increased. The continuation of research is necessary to help aim for a future where the disorder can be best treated.

Treatment Options and Future Growth

Traditional Treatment for Hemophilia

In both types of Hemophilia, treatment for the lack of the necessary blood clotting factors is simply injecting the necessary factor. To summarize, factor VIII is injected for hemophilia A patients and factor VIX is injected for hemophilia B patients. However, intravenous factor replacement has an array of downsides, including intravenous administration, frequent injections due the factor's short half lives, and the potential for development of inhibitors which will essentially render the factors useless (Okaygoun, Oliveira, Soman, & Williams, 2021). These inhibitors, also called neutralizing antibodies, are produced by the immune system in approximately 30% of hemophilia A patients and 3% of hemophilia B patients (Okaygoun, Oliveira, Soman, & Williams, 2021). Evidently, the higher chance of producing inhibitors in hemophilia A makes traditional treatment much less efficacious in many patients, leading to an increased emphasis of novel treatments. While the presence of inhibitors can deter treatment, immune tolerance induction (ITI) can help the body adjust to the factors and allow them to be used effectively. However, ITI requires extremely high doses of the replacement factors which can take a physical toll on the patient, along with costing the patient upwards of 1 million dollars depending on their prognosis (Thorat, Neumann, & Chambers, 2018).

Besides the risk of inhibitor development, traditional hemophilia treatment can also negatively impact a patient's quality of life. Intravenous injections can cause vein damage and scarring, and is painful, all of which can lower compliance to the treatments. Also, other methods of prophylaxis such as avoiding high-risk activities can be restricting for patients and have no guarantee of preventing long-term sequelae. Hemophilia is an expensive disease due to constant injections, but can become even more costly due to 1. A more severe phenotype 2. The presence of inhibitors. Firstly, for hemophilia A patients, the average cost for severe phenotypes was \$85,346, moderate was \$9,344, and mild was \$872. This pattern persists for hemophilia B, where severe was associated with a mean cost of \$123,716 and mild was associated with a mean cost of \$11,239 (likely due to the general rarity of mild hemophilia B). In general, the actual factors

represent approximately 50-90% of the hospital costs in hemophilia (Rocha, Carvalho, Lopes, & Araújo, 2015). Next, the presence of inhibitors is associated with a 4.8x increase in hemophilia care cost (Armstrong, Malone, Krishnan, & Wessler, 2014).

Novel Treatments for Hemophilia

As discussed in the previous paragraph, intravenous factor replacement can be an effective treatment of hemophilia, but still has many downsides that novel treatments aim to solve. These treatments include, but are not limited to: extended half life products (EHL), bypassing agents, anti tissue factor pathway inhibitors (Anti-TFPI), and small interfering RNA (siRNA).

First, extended half-life products are derived from recombinant FVIII (rFVIII) and recombinant FIX (rFIX), which are alterations of the naturally occurring factors, in order to increase the natural half life of the factors in the body (Okayoun, Oliveira, Soman, & Williams, 2021). These products do not change the typical injection method (remains intravenous) since the actual factors serve an identical purpose to the traditional methods, but they do provide the benefit of not requiring injections as frequently due the longer period of efficacy that the recombinant factors possess. In addition, EHL cannot account for the potential of inhibitors forming, so they are ineffective in many hemophilia patients, specifically hemophilia A patients. EHL products also carry a greater cost for patients. According to a retrospective study in Europe examining hemophilia B patients, the average cost for SHL (standard half-life) factor replacement was €186,528 compared to €290,620 for EHL products (Burke T, Asghar S, O'Hara J et al, 2021). While the increased cost could be a symptom of the novelty of EHL products, it is still associated with a significantly higher cost for only slightly less inconvenience for the patients.

While EHL products aren't effective in patients who develop inhibitors, a different therapy, bypassing agents, can be extremely helpful. There are two types of bypassing agents available in the treatment of hemophilia, which include recombinant factor VIIa (rFVIIa) and activated prothrombin complex concentrates (aPCC). Both serve the same function of bypassing the need for either factor (FVIII or FVIX), but they have different compositions. rFVIIa is a synthetic protein that helps the blood clot, while aPCC is a mix of clotting factors (Okayoun, Oliveira, Soman, & Williams, 2021). An example of a cutting edge bypassing agent is emicizumab, which mimics FVIIa by bringing together factor IXa and factor X to facilitate blood clotting (Shapiro A, Mitchell I, Nasr S, 2018). In addition, emicizumab can be injected subcutaneously (so it is significantly less painful, simpler, and more convenient for patients), has a longer half-life so it requires fewer injections, and like all bypassing agents, is not affected by inhibitors (Nogami, 2021). While bypassing agents represent a crucial solution to hemophilia patients who produce inhibitors, its usefulness is reflected in its high cost compared to traditional methods. According to a US study in 2014, treatment for patients with inhibitors through bypassing agents cost about \$613,000 (Thorat, Neumann, & Chambers, 2018). Undoubtedly, bypassing agents represent a not only more convenient but crucial advancement in hemophilia

treatment, but they carry a significantly increased cost compared to alternative treatments and therefore may not be feasible for some patients.

Another treatment option is antitissue factor pathway inhibitors. TFPI is a naturally occurring anticoagulant which slows down the initial phases of coagulation through a negative feedback loop that regulates the production of FXa. Anti-TFPI regulates TFPI in order to extend the initial phases of coagulation to produce more FXa. FXa is a thrombin precursor, and therefore is crucial in helping the body form stable blood clots. Examples of commercially available anti-TFPIs include concizumab, BAY 1093884, marstacimab, and MG1113. While anti-TFPIs represent a powerful preventative treatment for clotting issues, they cannot treat active bleeding episodes so patients must rely on other treatments for even acute bleeding. In addition, anti-TFPIs are not affected by inhibitors, but they do not have an extended half life and must be subcutaneously injected very frequently and even daily in some cases (Butterfield, Hege, Herzog, & Kaczmarek, 2020). Unfortunately, anti-TFPI treatments are still largely experimental and are undergoing clinical trials, so there are no current cost averages available for comparisons.

The last main treatment option includes subcutaneous double-stranded small interfering RNA (siRNA). Essentially, siRNA prevents the translation of anti-thrombin, which can help encourage thrombin development and consequently healthy blood clotting. In general, a 75% or greater reduction of antithrombin levels corresponds to thrombin levels of non-hemophiliac. The main siRNA treatment is Fitusiran, which is dosed subcutaneously once monthly. While the medication was largely effective, occasional breakthrough bleeding required other treatment (such as bypassing agents) and unfortunately resulted in a fatal cerebral venous sinus thrombosis which halted trials and has slowed down the drug's progression to the commercial space (Butterfield, Hege, Herzog, & Kaczmarek, 2020).

Gene Therapy Treatment

Gene therapy has shown promise as a potential treatment for hemophilia. This treatment could potentially be used as a cure long-term by replacing the defective gene associated with hemophilia with a functional one. This would allow for endogenous production of critical proteins, letting the body function healthily.

In the past, treatment for hemophilia has concerned regular infusions and treatments to regulate levels of FVIII and FIX. While there have been advancements in these treatments, challenges present themselves like high costs and the need for administration of treatment often (Nathwani 19). Further, gene therapy presents a potentially more secure, affordable, and long-term treatment.

Gene therapy for hemophilia provides appeal in its potential ability to maintain FVIII or FIX levels long-term. This would allow for a more stable level of clotting with only one treatment (Nathwani 19). While there are multiple benefits, there are still challenges to gene therapy. Some trials of gene therapy treatments for hemophilia faced problems with sustained expression and therapeutic levels (Batty 19; Nathwani 19). Gene therapy trials using AAV

vectors demonstrate the greatest promise, but still face issues like immune responses and transgene expression variability. The first study of AAV2 vectors presented transient FIX expression because of immune responses. This illustrated the necessity for refinement in vector design and delivery of gene therapy. Additionally, high costs associated with gene therapy prevent it from becoming accessible to many patients (Nathwani 19). Potential inaccessibility in low-resource areas could create an inequity in providing the best care of hemophilia to all patients. Further, more research and efforts must be done to ensure the safety of gene therapy use for hemophilia and the affordability of treatments.

Guidelines and Future Directions

Hemophilia treatment is and will continue to constantly evolve, which has prompted the World Federation of Hemophilia to create a series of the guidelines to structure patients' care.

Principle 1: National Coordination and Delivery of Hemophilia Care

A well-organized program aids in getting hemophiliacs people more reliable care. For this reason, hemophilia care centers should be part of responsible and accredited hospitals committed to patient care. These centers can work together and receive government funding. Lastly, these centers are part of a national registry.

Principle 2: Access to Safe Treatment

People with hemophilia need safe and effective treatments to prevent and manage bleeding. A majority of centers have access to traditional treatments, but many do not have access to novel treatments or gene therapies.

Principle 3: Laboratory and Genetic Services

Hemophilia care centers require lab services that can test blood and genetics or hemophilia and its treatment.

Principle 4: Education and Training in Hemophilia Care

Since hemophilia is a fairly rare disease, hospitals need trained specialists to help administer treatment to patients and ensure doctors are being constantly trained on novel treatments.

Principle 5: Clinical and Epidemiological Research

Many centers participate in clinical trials to help further hemophilia research, but the sheer lack of patients makes trials more difficult. Finding participants for gene therapy treatments is particularly difficult.

Principle 6: Acute and Emergency Care for Bleeds

Due to the danger of any bleeding for hemophiliacs, patients need rapid access to emergency care at hospitals during dire situations.

Conclusion

Hemophilia is prevalent globally and has affected both men and women, but primarily men. Throughout the history of the disorder, advancements have been made to improve its treatment, management, and diagnosis. Gene therapy has shown promise in the management of hemophilia long-term. Scientific research must be continued in order to produce more effective and affordable treatments for hemophilia.

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Antisense Oligonucleotide (ASO) Therapeutics, Optimal to Treat Pancreatic Cancer in Modern Age By Ryan Schoenstedt

Abstract

Pancreatic cancer (PC) is a rapidly increasing health concern, characterized by late-stage diagnosis and poor survival rates. Antisense oligonucleotides (ASOs) have emerged as a promising therapeutic approach for targeting oncogenic RNA in cancer cells. These small, nucleotide-based molecules bind and degrade the RNA critical for cancer cell survival, thereby inhibiting the protein production necessary for tumor growth. A unique attribute of ASOs is that they are highly customizable, enabling precise targeting of specific proteins involved in PC development. Despite significant progress, further research and funding are necessary to fully harness the therapeutic potential of ASOs in combating pancreatic cancer. This review explores the current advancements in ASO research, highlighting mechanisms of action, and the potential they hold for improving PC treatment through continued funding and research.

Pancreatic Cancer Statistics

Pancreatic cancer (PC) remains a significant health challenge, with approximately 30,000 new cases diagnosed annually in the United States (Yeo et al.). Responsible for 4.5% of cancer-related deaths globally, PC diagnosis is slightly more common in men (5.5 per 100,000) compared to women (4.0 per 100,000). The likelihood of PC increases with age (Prashanth et al.), typically diagnosed in people with a median age of 70 years (Aier et al.) and at a late stage, with approximately 80% of patients with metastatic disease upon presentation (Yeo et al.). Aggressive removal of the tumor has a median survival rate of 17-23 months, and the five-year survival rate of 8.5%. Once metastasis occurs, survival rates drop to 5% (Aier et al.).

PC progresses through four stages defined by specific criteria and reduced survival rates. In Stage I, the tumor is confined to the pancreas and measures 2 to 4 cm in diameter. Stage II involves the spread of the tumor to local lymph nodes and an increase in size beyond 4 cm (Prashanth et al.). The one-year survival rate for these early stages is approximately 41% (Tas et al.). In Stage III, the tumor extends to nearby blood vessels (Prashanth et al.), with a one-year survival rate dropping to 13% (Tas et al.). Stage IV is characterized by distant metastasis (Prashanth et al.), resulting in the one-year survival rate of 7% (Tas et al.).

Internal and external risk factors contribute to the development of PC. Smoking is a major factor, having an odds ratio of 1.74 for current smokers compared with never smokers. Carcinogens in tobacco smoke are released in the pancreas, inhibiting the protective enzymes that filter and detoxify, leading to mutations in those critical enzymes. Diabetes also elevates the risk of PC, with around 1% of cases attributed to this condition. Additionally, genetic mutations, particularly in the BRCA2 gene, significantly heighten the risk (Lowenfels and Maisonneuve). BRCA2 mutations impede the production of tumor suppressor proteins, thereby increasing susceptibility to PC (Venkitaraman).

When a patient is diagnosed with PC, the primary tumor and metastatic lesion are imaged. This guides the treatment plan, either resulting in surgery or chemotherapy. The first option, surgery, is usually a viable option for around 20% of patients. The low percentage can be attributed to PC's high metastasis rate, rendering surgery an ineffective treatment. For chemotherapy, two combinatorial therapeutics are the current standard of care for PC (Conroy et al.). The first treatment is Folfirinox and Gemcitabine. The drugs compliment each other, providing non-overlapping effects, making them a suitable pair. In clinical trials, there was a higher median survival with the combination of treatments, at 6.24 months, compared to 5.91 when only using gemcitabine. The treatment combination has a high incidence of grade 3 to 4 adverse events like neutropenia (Saung and Zheng) (reduction in number of white blood cells (Thomas)) and thrombocytopenia ((Saung and Zheng) (low platelet count (Stasi)). The other combination of successful therapeutics is Nab-Paclitaxel and Gemcitabine. Similar to Folfirinox and Gemcitabine, median survival increased with combined treatment, at 8.5 months, up from 6.7 months when only administering gemcitabine. This particular combination was especially effective for patients with advanced PC and the elderly. Higher levels of neutropenia and fatigue were the main side effects ((Saung and Zheng). Although the current standards of care increase the median weeks of survival, there is still more research that needs to be done. There is currently a lack of targeted therapies in the field, which is why scientists are beginning to search for other therapies as the future of PC treatment.

Antisense oligonucleotides (ASOs) present a promising avenue for targeted therapy, with the potential to disrupt oncogenic processes in PC cells. Continued research and investment are necessary to advance these targeted therapies and improve survival rates for PC patients.

From Conception to Clinical Approval

Due to PC's low survival rate, researchers are developing innovative therapies targeting oncogenic RNA in patient cells. This is a high priority for scientists due to how critical RNA is for gene expression. Once RNA is mutated, gene expression is altered. This creates a phenotypic change in the cell that eventually leads to cancer (Goodall and Wickramasinghe).

Antisense oligonucleotides (ASOs) (Holgersen et al.), chemically modified short chains of synthetic RNA or DNA (Schiffelers et al.), represent a promising therapeutic approach. When ASOs bind to RNA, the ASO changes the RNA in two ways. First, RNA cleavage and degradation leads to steric blocking, which does not directly degrade RNA (Bennett). Instead, regulatory proteins are blocked which change the RNA secondary structure (Holgersen et al.). In both major modifications, the length of the ASO grants it a high level of specificity providing scientists flexibility to specialize treatments to the patient (Bennett).

The field of ASOs is relatively new, originating with the first practical application in Novosibirsk, Russia, in 1967. Scientists attached chemical groups to oligonucleotides, changing their function. These scientists found that adding chemical groups direct the oligonucleotide to a complementary nucleic acid strand, changing the characteristic and function. Seven years later, in 1974, the phosphite coupling approach was created which allowed a new class of ASOs to be

created, the phosphoramidates. This development highlighted ASOs' potential as a modular and targeted therapeutic. Then, in 1978, ASOs were found to inhibit RSV replication in a chicken embryo with modifications at the 3' and 5' ends. This was the first successful in vivo experiment, demonstrating the practicality of ASOs in live organisms. Unfortunately, in the 1980's, progress stalled due to three reasons. There was a belief that the hydrophilic sugar-phosphate backbone prevented ASOs from entering eukaryotic cells. Another was that the synthesis for these ASOs was very time-consuming and expensive due to the lack of available technology necessary for automating ASO production. The final reason was the limited knowledge on the genomic sequences for humans and other organisms. Since ASOs need to bind to specific RNA molecules, not knowing the genetic sequence making up that RNA inherently weakens the ASO's practicality.

The slow progress of ASOs' research and advancement continued until the late 1990s, when the first antisense drug was created: fomivirsen. This drug treated cytomegalovirus (CMV) retinitis (Oberemok et al.), a sight-threatening form of CMV (Eid et al.) in AIDS patients. Fomivirsen was effective because ASOs could target the mRNA that encoded the CMV retinitis protein required for viral replication, inhibiting the virus. Although fomivirsen was replaced by newer drugs in the early 2000s, it was pivotal in demonstrating that ASOs can be effective on the cellular and patient levels.

Around 18 years later, in 2016, two more antisense drugs were approved by the FDA. Approved in September, Eteplirsen treated Duchenne muscular dystrophy (DMD) (Oberemok et al.), a severe condition that degrades muscles causing weakness and atrophy (Allen and Whitehead). Eteplirsen uses RNase H to degrade exon 51 on the DMD RNA. In doing so, the exon is skipped during RNA splicing, correcting the disorder. A more recently approved antisense drug, Spinraza, has been approved for spinal muscular atrophy (SMA) (Oberemok et al.), a disorder caused by degeneration of neurons in the spinal cord leading to muscular atrophy and weakness (Wirth et al.). This disorder is caused by mutations to the SMN2 gene, changing the function of the overall survival motor neuron (SMN) protein. Spinraza blocks intron 7 at the internal splicing site, the reason for the mutated SMN protein. By blocking intron 7, exon 7 can be expressed thus correcting the reading frame and stabilizing the SMN protein (Oberemok et al.).

Although there is great evidence of the efficacy of ASOs on noncancer-related diseases, scientists are continuously looking for better ways to apply ASO technology to treat PC.

Connecting ASO technology to Known Protein Changes in Cancer

KRAS

PC exhibits numerous oncogenic markers suitable for ASO-targeted therapy. KRAS gene mutations, present in approximately 90% of PC cases, represent a key target (Dai et al.). KRAS regulates cell division by relaying external signals to the nucleus of cells. When this gene becomes mutated, KRAS molecules prevent the hydrolysis of GTP, permanently activating the

RAS molecule. Due to the continual activation of the RAS molecule, the cell continues to divide and becomes oncogenic (Jančík et al.). ASOs targeting KRAS point mutations could inhibit cell growth by preventing continuous RAS molecule stimulation (Nakada et al.). This was demonstrated in 2001 when a 17-mer phosphorothioate ASO inhibited PC invasiveness by targeting the KRAS gene. The study was expanded upon in 2013, where it was shown that ASOs could limit PC cells from moving to S phase, inducing apoptosis in many cases (Dai et al.). Given that chemotherapy and other classical cancer therapeutics have significant off-target effects, ASOs have demonstrated their ability to preferentially target the mutated KRAS cells.

Heat Shock Protein 27 (Hsp27)

Heat Shock Protein 27 (Hsp27) is another protein of interest due to its frequent mutations in PC. This protein is induced by stress signals and regulates apoptotic effectors. When mutated, Hsp27 reduces the toxic effect of the oxidized proteins responsible for cellular degradation during apoptosis. Hsp27 can protect the cell from apoptosis by restoring protein homeostasis and the actin-cytoskeleton (Matsui et al.), allowing cell signaling to continue (Moujaber and Stochaj). When Hsp27 is suppressed, the cell becomes sensitive to chemotherapy, as demonstrated by Apatorsen, a second-generation ASO, in prostate cancer. The drug inhibits Hsp27 expression, and similar to what is expected, the rate of tumor growth decreased. A majority of patients saw a decline in Hsp27 expression after several doses. In 2016, Apatorsen began a randomized stage II trial and represents the potential for ASOs in cancer (Chi et al. 1). In bladder cancer, similar results were observed. A second-generation ASO, when working with chemotherapy, has been shown to delay tumor growth due to the repression of Hsp27 (Hadaschik et al.). Although these represent breakthroughs in other cancers, similar therapeutics could likely prove efficacious in PC through more research.

Bcl-2 & Bcl-xL

A family of proteins, Bcl-2, is of key interest to scientists. This family of proteins regulates apoptosis, a necessary role to maintain homeostasis. When mutated, the proteins inside of the family can change functions and become oncogenic (Zhang et al.). A key protein in this family is Bcl-xL, which inhibits apoptosis and can prevent cell death. Increased expression has been seen to cause cancer, mainly in gastric, ovarian, and pancreatic malignancies (Xu et al.). It has been found that anti-Bcl-xL ASOs inhibit PC cell growth and increase apoptosis rate. The modified ASO used showed a significant decrease in cell growth compared with the unconjugated ASO (Dai et al.). Furthermore, anti-Bcl-xL ASOs in conjunction with Gemcitabine (Xu et al.), a drug that decreases DNA synthesis (Noble and Goa), has been shown to lower cell viability. This means that this concurrent treatment decreased the amount of alive PC cells, indicating a potential therapy (Xu et al.). Finally, Oblimersen, an ASO drug for chronic lymphocytic leukemia and multiple myeloma, has had a strong response in the phase I/II trials. This drug binds to the first 6 codons of Bcl-2 mRNA, silencing the protein. About 20% decrease in Bcl-2 protein levels was observed in the phase I trial. In that same trial, 14 out of 29 patients

had a complete remission. Oblimersen is currently in a phase III trial in combination with other chemotherapeutic drugs (Gagliardi and Ashizawa). Similar to other proteins listed, further studies of Bcl-2 and Bcl-xL in the context of PC could lead to novel therapeutics as these genes play documented roles in PC biology.

miRNA

MicroRNA (miRNA) are yet another target of interest for scientists. These small non-coding RNA molecules regulate protein expression. It is estimated that 30% of genes are regulated by miRNA. miRNA-21, a specific type of miRNA, can be overexpressed in PC. When miRNA-21 becomes mutated, the RNA promotes tumor development by negatively inhibiting tumor suppressor genes (Hu et al.), thus initiating cancer (Li et al.). In addition, miRNA-21 has been linked to increased chemotherapy resistance in PC cells (Pan et al.). A combination of one antimicroRNA-21 ASO and Gemcitabine has been shown to downregulate oncogenic miRNA-21 in PC. This leads to an upregulation in tumor-suppressor genes, limiting cells from multiplying. A combination of these two drugs yields a higher rate of apoptosis than just the ASO or Gemcitabine in isolation, suggesting that a blend of treatments can be effective (Dai et al.). Future researchers might investigate using ASOs to modulate miRNA-21 levels to make cells more sensitive to chemotherapy. In doing so, any changes in miRNA-21 expression could become a marker for treatment success, providing more data for oncologists and researchers (Pan et al.).

TGF-beta2 Receptors

A final target of interest for ASO therapy is the TGF-beta2 receptors, responsible for cell proliferation and migration (Schlingensiepen, Fischer-Blass, et al.). TGF-Beta 2 is a tumor suppressor, but once mutated, becomes highly protumorigenic (D'Cruz et al.). There are three isoforms in mammalian cells: TGF-beta1, TGF-beta2 and TGF-beta3. Although all three are upregulated in cancer, TGF-beta2 is correlated with advanced tumor stages in PC. Also, PC patients with mutations in TGF-beta2 have the shortest postoperative survival periods of any other isoforms (Schlingensiepen, Fischer-Blass, et al.). An ASO therapy in development is Trabedersen or AP 12009 (D'Cruz et al.), a 1st generation ASO (Schlingensiepen, Fischer-Blass, et al.) that binds to a complementary sequence of mRNA from the TGF-beta2 gene (Schlingensiepen, Jaschinski, et al.). In doing so, Trabedersen stimulates inflammatory cytokines (proteins regulating immunity levels), strengthening the immune system (D'Cruz et al.) and reversing the immunosuppressive effects of TGF-Beta2. Another effect of treatment is that cell-dependent cytotoxicity of lymphokine activated killer (LAK) cells becomes activated, allowing LAK cells to target and kill PC cells (Schlingensiepen, Fischer-Blass, et al.). In phase I/II trials, Trabedersen has reduced TGF-beta2 secretion in PC cells. The safety profile has been strong, although final results have not been shared (D'Cruz et al.).

Overall, ASO therapies present a promising avenue for increasing the standard of care for PC. The ability of ASOs to specifically target oncogenic mutations, such as those found in KRAS, Heat Shock Protein 27, Bcl-2 family proteins, miRNA-21, and TGF-beta2 receptors, highlight their potential. Continued research and clinical trials are essential to fully realizing the therapeutic potential of ASOs in combating PC and improving patient outcomes.

From Injection to Degradation

ASOs can be administered via intravenous infusion or subcutaneous injection, subsequently entering the body via the bloodstream. The highest level of distribution is in the liver, kidney, bone marrow, adipocytes, and lymph nodes. ASOs that are uncharged and have a smaller affinity to plasma proteins are uptaken into cells at a slow rate. Since these ASOs stay in the blood, they are more likely to end up in the liver, kidney, or other clearance organs. However, ASOs have a difficult time achieving results in the central nervous system. This is due to their inability to cross the blood-brain barrier. To counter this, ASOs are usually delivered into the spinal fluid directly. The ASOs need to be modified single-strand oligonucleotides delivered by intrathecal injections for the best treatment. Active uptake into neurons via this process has shown to be successful.

ASOs exhibit long half-lives, usually 2 to 4 weeks (Geary et al.). This is in sharp contrast to other types of RNA therapies like positive siRNA/polymers which have a half-life of around five minutes (Baigude and Rana). In the half-life of the ASO, they must undergo two parts of cellular uptake. The first step is adsorption where the ASOs adhere to the surface of the cell. This is a rapid process that requires no energy input and can be saturated. Once bound to the surface of the cell, many different pathways are possible for the ASOs. Some of these pathways are productive, leading to a pharmacological effect and hopefully the intended therapeutic outcome. Alternatively, other uptake mechanisms lead to no therapeutic effect, which researchers aim to avoid when designing ASOs. As for the productive mechanisms, examples are uptake via clathrin- and caveolin-dependent mechanisms (Crooke, Wang, et al.). These mechanisms rely on different endocytosis pathways and unique vesicles to uptake the ASO molecule into the cell (Abrami et al.; Minshall et al.; McMahon and Boucrot). It is important to note that with such diversity of uptake mechanisms, ASOs have the potential to use different pathways should one be blocked.

Many different cell surface proteins are helpful in ASO uptake. For example, receptor SCARA1 modifies ASOs with peptide-conjugated phosphorodiamidate morpholino (PPMO) or phosphorothioate tricyclo DNA (tcDNA). SCARA1, with these modifications, helps ASOs to become nanoparticles that can enter the cell. Furthermore, changes to the structure of ASOs alter the relationship with the proteins involved in uptake. For example, the conjugation of N-acetyl galactosamine to PS-ASOs allows the new conjugate to bind to the asialoglycoprotein receptor (ASGPR). This would increase the delivery of ASO into hepatocytes (Crooke, Wang, et al.) (liver cells that break down lipids and carbohydrates) which has broad therapeutic applications (Grompe).

Once internalized in the cell, the ASOs have to reach their RNA target (Crooke, Wang, et al.). The ASOs target RNA in the nucleus and/or cytoplasm (Geary et al.). The ASOs must travel through the cellular space to hybridize at the target's cognate site. The ASOs must interact with many proteins like AGO2 which binds to RNA and aligns that RNA strand with a complementary nucleic acid (the ASO) (Janowski et al.). The Ago 2 protein increases the kinetics of ASO binding. Furthermore, ASOs must recruit enzymes to accomplish their goal. One of the most readily used is RNase H1, a relatively free enzyme located in cellular areas where ASOs typically are: nucleus, cytoplasm, and mitochondria. ASOs recruit RNase H1 to degrade DNA-RNA duplexes (Crooke). This is not an abnormal process as homeostasis is regulated by the formation and deletion of DNA/RNA hybrids (Liu et al.). After ASO-RNA hybridization, different proteins can be recruited to the new ASO/RNA heteroduplex. These proteins could modify the RNase H1 cleavage which would modulate the final effect on the RNA. It has been shown that Ku70, P54nrb, and HspA8 can bind to the heteroduplex and inhibit RNase H1 recruitment (Liang et al.), which remains a significant area of research for investigators to explore.

Chemical Modifications of ASOs

ASOs are easily modulated due to their chemistry. Naturally, there have been numerous changes over the years that modify the structure of the ASO into three broad types: first, second, and third generation (Quemener et al.). The first synthesized modified oligonucleotides were methyl-phosphates. These are non-charged oligomers where the non-binding oxygen atom is replaced with a methyl group at each phosphorus. Due to methyl-phosphates being uncharged, they have limited solubility and cellular uptake. An upside with these being uncharged, though, is that it limits charge-charge repulsions. That allows ASOs to bond with RNA/DNA duplexes at a higher rate (Dias and Stein). Unfortunately, these modified ASOs are incompatible with the cleavage of RNase H (Quemener et al.). This feature limits the cleavage ability of methyl-phosphate ASOs.

The most attractive first generation ASO are the phosphorothioates which are relatively stable and highly soluble. This solved the problem with methyl-phosphates because the phosphorothioates can activate RNase H (Dias and Stein). The problem with this modification is that the phosphorothioates are very toxic and non-specific (Quemener et al.). However, scientists have found that the phosphorothioates can cause effects downstream from the initial site. These downstream effects occur because of the limited specificity of the phosphorothioates, allowing a high affinity for various cellular proteins (helicases, polymerases, and nucleases) (Crooke, Vickers, et al.). Consequently, there could be one phosphorothioate molecule that triggers a cascade leading to the downregulation of other proteins. As opposed to only targeting one protein, these phosphorothioates offer the opportunity for therapeutics to target multiple at the same time which explains why these have gotten very close to clinical application.

Wanting to improve upon the first generation of ASOs, the second generation ASOs aimed to be more resistant to degradation by cellular nucleases and have increased hybridization

to their target (Dias and Stein). A common modification is the addition of an alkyl group at the 2' end of the ribose. This creates ASOs that are less toxic and can have a higher affinity for their target. However, these types of ASOs are unable to recruit RNase H leading scientists to look for other modifications. The last generation of ASOs tries to improve binding affinity and resistance to nucleases. A common modification is to replace the ribose with a morpholine moiety and the phosphodiester bond with a phosphorodiamidate bond. This creates phosphorodiamidate morpholino oligomers (PMOs) (Quemener et al.) which are very stable yet do not have downstream effects like the phosphorothioates (Dias and Stein). To reiterate, second and third-generation ASOs do not recruit RNase H1 (Quemener et al.) which forces scientists to determine other mechanisms of action to elicit a treatment response.

Significance and Mechanism of RNase H1

RNase H1 is a pivotal enzyme in the therapeutic action of antisense oligonucleotides (ASOs) due to its role in degrading RNA within DNA/RNA hybrids. Composed of a single peptide chain, RNase H1 specifically targets RNA strands that are hybridized with DNA, binding at the N terminus of the RNA (Vickers and Crooke). The enzyme comprises three key domains: hybrid binding, catalytic, and connecting, with the hybrid binding domain being extensively studied for its ability to recognize and bind DNA/RNA hybrids (Reyes et al.). Upon binding to a DNA/RNA hybrid, RNase H1 initiates the cleavage of the RNA strand starting at the 5' end (Vickers and Crooke). The enzyme hydrolyzes the phosphodiester bonds within the RNA (Li and Wartell), progressively degrading the RNA strand over a stretch of 7-10 nucleotides, which corresponds to approximately one helical turn of the hybrid (Vickers and Crooke).

It is important to note that RNase H1's function is not only pivotal for ASO efficacy but also for normal cellular homeostasis. In unicellular organisms, the absence of RNase H1 slows growth rates, while in multicellular organisms, its deletion is lethal due to failures in mitochondrial DNA replication. Overall, RNase H1's ability to degrade RNA within DNA/RNA hybrids is crucial for both maintaining cellular homeostasis and the therapeutic efficacy of ASOs (Nowotny et al.).

Conclusion

Pancreatic cancer is one of the most deadly cancers worldwide, with a median five-year survival rate of only 8.5% (Aier et al.). Current treatment options, including surgery and chemotherapy, offer limited efficacy due to the high rate of metastasis and significant adverse effects (Conroy et al.). Antisense oligonucleotides (ASOs) present a promising alternative by specifically targeting and degrading oncogenic RNA, thereby inhibiting the production of proteins crucial for cancer cell survival (Quemener et al.). ASOs have already shown success in treating non-cancer-related diseases, with FDA-approved drugs like Eteplirsen and Spinraza demonstrating their potential (Oberemok et al.). In the context of pancreatic cancer, ASOs can be tailored to target specific mutations and proteins, such as KRAS, Heat Shock Protein 27, Bcl-2 family proteins, miRNA-21, and TGF-beta2 receptors. These targets are integral to the

development and progression of pancreatic cancer, making ASOs a viable option for targeted therapy.

The versatility of ASOs, along with their ability to recruit RNase H1 to degrade RNA within DNA/RNA hybrids, underscores their therapeutic potential (Vickers and Crooke). By reducing the levels of target RNA, ASOs can effectively decrease the expression of oncogenic proteins, offering a more precise and less toxic treatment option compared to traditional chemotherapy. Despite the promising advancements, further research and funding are essential to fully harness the potential of ASO therapies for pancreatic cancer. Continued clinical trials and studies will help to refine these treatments, improve their efficacy, and ultimately, increase the survival rates and quality of life for pancreatic cancer patients.

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Development of the Prototype of Carbon Capture Method Using Chlorella

By Youngseok Song

Abstract

This research aimed to discuss the issues with the increasing amount of greenhouse gasses in the atmosphere and the evaluation of currently adapted methods to tackle such issues. The Carbon Capture Storage (CCS) method is adapted in many places, however its cost and efficiency limitations were investigated. To suggest a new method which could be more efficient as well as promoting participation at individual level, this research developed a prototype which uses chlorella to capture CO₂ in the atmosphere. Although the prototype does have some limitations such as requiring input energy which is solar power, the prototype does hold great potential to be adapted and utilized at the individual level.

Introduction

One of the biggest challenges that the Earth faces today is climate change, which is caused by high emissions of greenhouse gasses. The greenhouse gasses in the ground level absorbs all the heat and forcibly cools the stratosphere, which damages the ozone layer. Ozone layer loss means more UV rays entering Earth's geosphere, which essentially makes Earth even hotter because Earth's atmosphere already has a high concentration of greenhouse gasses. This leads to melting of ice, rising sea levels, and various climate hazards. Scientists have known about the potential dangers of increased greenhouse gas levels in the atmosphere all the way from 1896 by a Swedish scientist Svante Arrhenius (Crawford).

As shown in Appendix A, greenhouse gasses have been in a worryingly increasing trend over the past few decades (Appendix A). The carbon dioxide (CO₂) levels in our atmosphere have especially been increasing steadily. Unfortunately, despite all this knowledge, modern science is yet to find a successful method to tackle this challenge that is both effective and efficient.

To tackle the problem mentioned above, this research aims to determine if chlorella could be a potential solution to carbon capturing that is both effective and costly. To do so, this research discusses what chlorella could do in comparison to the method that is adapted for now as well as building a prototype to stimulate how it could work.

Background Studies

Before discussing chlorella and its use for the carbon capturing method, carbon capturing and how it is used today should be discussed first. Carbon capture is definitely one of the most effective technologies to tackle the level of carbon dioxide in the atmosphere. It usually involves capturing CO₂ emissions at their source, such as power plants and factories, before they are released into the air. The captured CO₂ is then transported, often via pipelines, to storage sites. It is injected deep underground into geological formations, such as depleted oil and gas fields or saline aquifers. Here, the CO₂ would be stored for an indefinite amount of time. In addition, all

types of carbon capture technology fall under three categories; post-combustion carbon capture, pre-combustion carbon capture, and oxy-fuel combustion systems. First, post-combustion carbon capture grabs CO₂ from the exhaust gasses after the fuel is burned. This method uses chemicals to absorb CO₂ from the smoke, making it easier to trap and store. Second, pre-combustion carbon capture involves removing CO₂ before burning the fuel. Here, the fuel is converted into a mix of hydrogen and CO₂, and the CO₂ is then separated out. Finally, oxy-fuel combustion systems burn the fuel in pure oxygen instead of regular air. This creates exhaust that's mostly CO₂ and water vapor, simplifying the CO₂ capture process (Gonzales).

Unfortunately, although the carbon capture technology seems promising in theory, that is far from the reality. The two main issues that the current carbon capture technology face today are cost and efficiency. Currently, there are 42 Carbon Capture and Storage (CCS) projects around the world operating. According to their designs, they are intended to capture and store approximately 49 million tonnes of carbon dioxide annually. Sadly, the world produces 37 billion tonnes of carbon dioxide per year. This would make the intended storage amount only around 0.1 percent of the annual carbon dioxide emissions (Abreu). Furthermore, these technologies can only be implemented in wealthy countries that can afford to build carbon capturing facilities. CCS technology is really expensive and costs a lot to implement. Capturing CO₂ can range from 27–48 Canadian dollars per ton for concentrated streams and 50–150 dollars per ton for diluted streams, which is quite high. That would cost billions of dollars annually. In Canada, the costs often end up on the pricier side due to the complexity and need for custom designs. Despite being around for over 50 years, CCS hasn't gotten much cheaper, especially when compared to how quickly solar and wind have dropped in cost. The oil and gas sector still heavily depends on government funding to make CCS work financially. Unlike renewable energy, which needed subsidies mainly at the start, CCS continues to need a lot of financial help to be viable (Cameron). Even after all the funding put into the CCS technology, it is incapable of grossing any proper results as explained before. The core of the problem with the CCS technology lies in the fact that only the government and unenthusiastic corporations are taking actions to clean up after the mess made by the general public. When every single individual in the world is emitting larger amounts of CO₂ daily, only a small group of activists are taking action to promote CCS technology. Instead of cliques taking huge actions, the mass taking small actions would build up to a much more satisfying result.

This is why this research developed a prototype which uses chlorella as a carbon capturing method, as it can enable individuals to participate altogether. Not only is chlorella a resilient plant that can reproduce at a rapid rate, but the microalgae can absorb CO₂ from the atmosphere with great efficiency. *Chlorella vulgaris*, a specific type of chlorella has been found to be most effective in the removal. It could absorb 0.412 grams of CO₂ per liter of TPP (Thermal Power Plant) exhaust gasses and that it removed up to 8.125 liters of CO₂ per day (Politaeva). In addition, Chlorella is one of the easiest organisms to take care of with not much attention needed by the owners. The prototype which this research developed will provide an

environment where chlorella can thrive without much intervention, which is explained throughout the next section of the research.

Mechanics

The mechanics of the prototype revolve mainly around the use of photosynthesis. In each of the terrariums, adapted design mimics an actual aquatic ecosystem to ensure chlorella's survival and vitality. This meant that the terrarium must provide the fundamental conditions for photosynthesis. The four biggest conditions would be chlorophyll, sunlight, CO₂, and water. Chlorella already contains chlorophyll and is placed in water, meaning that it only has to provide sources of sunlight and CO₂. Nonetheless, continuous light supplies and air flotation provided through electricity would have been inappropriate as using electricity generally leads to more CO₂ production due to its production procedure. Therefore, the prototype adapted a solar panel which can provide continuous energy sources to provide lights and ventilation through fans.

All the terrariums are equipped with an LED light source. This component is crucial to our design because it provides the light necessary for photosynthesis. Connected to Arduino, the LED turns in dark environments to provide the chlorella with light. There's also a green LED light that comes on when the light level is adequate. Using this set up, the design ensures that the chlorella receives the right amount of sunlight to thrive.

As discussed earlier, the solar panel also powers the fan, which is connected to digital pin 7. Good air circulation is essential to make sure that chlorella receives an even distribution of CO₂. Without proper airflow, the carbon dioxide levels around the chlorella can become uneven, reducing its efficiency in capturing carbon. Using the fan, the model provides full ventilation that brings in the necessary carbon dioxide into the terrarium (Appendix B and C). This would then be used by the chlorella to perform photosynthesis. Furthermore, the fan helps regulate the temperature within the terrarium. When placed in underutilized spaces like the basement, the temperatures may become too hot or cold since those spaces don't receive as much ventilation or sunlight. By keeping the air moving and the temperature consistent, the fan promotes chlorella's healthy growth.

The Arduino system includes other parts as well. The model includes an Arduino system using an Arduino microcontroller with sensors that measure the key components of photosynthesis. A light sensor (connected to analog pin A0) monitors the ambient light levels of the terrarium. The light sensor is connected to an analog input pin on the Arduino and continuously reads the intensity of light in the environment (Appendix D). Additionally, if the light levels exceed a certain threshold, the system activates the fan to cool down. The fan is activated when the light level exceeds a threshold of 200 units. Moreover, a green LED, connected to digital pin 9, is used to indicate optimal lighting conditions. Another sensor in the system is the temperature sensor. The design integrates a DS18B20 temperature sensor to the Arduino system by connecting it to digital pin 6. It updates the owner of the environment's temperature in real-time readings.

Similar to the light sensor, the fan would turn on if the temperature rises to a certain degree. The last sensor in our model is the CO₂ sensor. The CO₂ sensor detects the level of carbon dioxide inside the tank and displays it just like the other sensors. It will help ensure that the chlorella colony is receiving an adequate amount of CO₂ to thrive. On top of all of this, to make sure everything runs smoothly, this prototype implemented a monitoring design. The information gained from the various sensors is then displayed on a Liquid-crystal Display (LCD) screen, constantly updating as the data fluctuates. The LCD is connected via I2C communication (SDA to A4 and SCL to A5). This simple setup informs the user of any potential harm being done to the design (Appendix E), and finally, the final version of the prototype can be found in appendix F (Appendix F).

Results

As of now, the model has not been running for a long enough time for it to gather enough data to present an accurate set of results. However, there are multiple studies that illustrate the effectiveness of chlorella in capturing CO₂.

According to a study conducted by the Department of Chemical Engineering, University of Sistan and Baluchestan, Zahedan, chlorella was very well adapted to removing carbon dioxide from atmospheres with high levels of CO₂. In their studies, the researchers conducted an experiment where *Chlorella vulgaris* would grow and capture CO₂ in a special tank called an airlift bioreactor. The temperature inside the tank was kept at 30°C, which is about as warm as a summer day. This temperature would be similar to the temperatures our terrariums would be placed in during summertime. The main goal was to find out if blowing a lot of gas into the tank would help the algae grow better and they did so by pumping high amounts of gas into the airlift bioreactor. The results showed that *Chlorella vulgaris* still grew well in the gas-concentrated conditions, and was able to remove nearly 80% of the CO₂ (Sadeghizadeh).

This research can be used to predict the efficiency for this specific design as well. In the research, scientists used a chlorella colony of 5 million algae cells per milliliter of the water and used 20 liters of the medium. The prototype of this research utilized a standard sized fish tank, which would be approximately 50 liters. If the chlorella of the tank has a concentration similar to that of the study's, this prototype would be able to remove CO₂ from the atmosphere just as much, if not more efficiently.

Why chlorella?

As has been stated before, there have been many attempts at removing CO₂ from the atmosphere such as the CCS technology. However, time and time again, the most sustainable and cost-effective solution has been proven to be biological CO₂ fixations. It is a naturally occurring process in which autotrophic organisms convert CO₂ into organic matter (Barahoei). Using chlorella to remove CO₂ from the atmosphere would be a type of biological CO₂ fixation. To pursue a more sustainable solution, this project implemented a microalgae like chlorella to perform the fixation.

The two most compelling advantages that the choice of chlorella provides for this project are its resilient nature and eco-friendliness. Chlorella is incredibly hardy and adaptable which is ideal for a variety of environmental conditions. Thus, chlorella can thrive in environments such as low-light or variable temperature settings which is often the case for underutilized spaces in buildings. Whether installed in a dimly lit office basement or a sunlit corner of a home, chlorella can continue to perform photosynthesis effectively.

Moreover, chlorella's eco-friendly characteristics align perfectly with the goals of this project. Chlorella reduces carbon footprints without the need for harmful chemicals or synthetic materials. Photosynthesis also releases oxygen, thereby improving air quality. However, the part that makes chlorella really eco-friendly is the fact that it doesn't only have to serve as a medium for photosynthesis. Chlorella can be harvested and used in various ways, such as in food products or fertilizers.

Limitations

Considering all the advantages chlorella holds in CO₂ removal, it may seem as if it is flawless. Unfortunately, there are certain drawbacks to keeping the prototype completely eco-friendly. Because the components of the arduino system are powered by the solar panel, it can only operate during daytime when it is fueled by solar energy. Without a constant supply of sunlight, it would not be able to function. This is not simply a problem of day and night, but also a problem of geographical location and weather. During rainy seasons or in places where the sunlights are not supplied enough, it will not function properly due to lack of solar energy. Nonetheless, this problem could be tackled with an addition of a battery to the prototype. If the solar panel could charge up the battery, this prototype would be able to function during the night time or even in places with no sunlight as long as its user is willing to move the prototype when charging is needed.

Another limitation of the design is the need for space. Even though the prototype can be placed on top of each other to minimize the amount of space it takes up, there is a limit to how many 50 liter tanks can be placed on top of each other in a chosen space. In normal households, it is unlikely that more than one tank will be implemented. Also, if the tanks are built upon each other, it may make it harder for its user to move the prototype from one place to another when needed.

Finally, although the prototype is mostly designed to be self-sustainable, it may require maintenance checks from time to time. Much like a regular fish tank, the chlorella colonies are going to require cleaning and the components are going to have to be repaired or replaced. Just like any other technology, it is going to require attention from the users to operate for a long time.

All in all, as a prototype design, the chlorella model still has ways to go before it can be fully implemented into daily lives of people, addressing these limitations is crucial for improving the design and ensuring its practical implementation and long-term sustainability.

Conclusion

This study aimed to develop an innovative and space-efficient design that relies on chlorella's resilience and photosynthesis to remove CO₂ from the atmosphere, so that this could promote participation from the individual levels instead of solely depending on the government and country to solve the CO₂ emission issues. The final design of the prototype was a series of vertical aquatic terrariums each containing chlorella and can be placed in underutilized spaces in houses or office spaces. Furthermore, there are not many space limitations as the tanks which contain algae could be designed according to the users. The convenience of the product would appeal to the mass public, as it does not require much human attention nor work as well as any additional input power sources. It is expected that if this can be promoted to a mass number of individuals, it may be more efficient than solely relying on a single CCS facility to capture the CO₂ in the atmosphere.

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Impact of Neutral and Negative Word Choices in News Headlines on Thai People's Perception of Environment News Stories By Dalida Milly Janevathanavitya

Abstract

In today's fast-paced media landscape, headlines serve as a contact point between audiences and news stories. The language used in these headlines is not merely to summarize, but to influence how the readers interpret the article as a whole. To determine the level of impact headlines have on the perception of news stories, two surveys were designed and sent to two randomized groups of 56 people from Bangkok, Thailand. One survey showed a headline with neutral language and the other showed a headline with negative language, and both asked the respondents to predict information presented in the article. To reduce bias from politics and social issues, the news topic selected was environment news.

Results of this research demonstrate that negative word choices lead to more polarized perceptions compared to neutral ones. Usually, negative language triggers more people to choose relatively negative answers. However, on some occasions, it results in polarization, with most people choosing very negative or very positive answers. Moreover, predictions made by respondents who read headlines with neutral language were closer to real-world values than those made by respondents who read headlines with negative language. This suggests that negative word choices can distort perceptions and highlights the importance of careful consideration before drawing conclusions from headlines.

Keywords: word choice, news headlines, perception

Introduction

Diction manipulation is everywhere, from billboards on the roads to advertisements on milk cartons. Notably, even factual content such as news stories dedicated to public information distribution are designed to evoke emotions. With abundant information available due to the rise of digital media, a reporter's task is not only to provide informative updates but also to make them eye-catching, at times at the cause of shifting the story's tone.

Understanding the impact of diction manipulation is essential in promoting journalistic integrity and fostering an informed public. The media has a powerful role in shaping public opinion (Happer and Philo, 2013), and it does so through language. Previous experiments have shown that even subtle changes in language, such as a single word in a metaphor, can significantly alter the interpretation of stories (Thibodeau and Boroditsky, 2011). This research builds on that foundation by specifically examining how neutral versus negative word choices in headlines sway readers' perceptions of news stories.

To effectively analyze how language affects perception, this study minimizes pre-existing biases by choosing a less polarizing news topic: environment. By using weather patterns as the

focal point, this research hopes that the impact on perception is primarily due to the differences in language used in the headlines.

Literature Review

Several theories are crucial to understanding the impact of word choice in news reporting, including schema theory. Schema theory proposes that readers interpret information based on pre-existing mental frameworks or schemas — shaped through personal experiences, social interactions, and cultural influences (Bartlett, 1932). These schemas can be triggered by certain words or phrases. For example, in a narrative where characters are described using words associated with heroism or villainy, readers may activate corresponding schemas that affect their judgments of characters' actions later on. This is often applied to news reporting, where players are connected to characteristics through word choice.

In addition, other biases can arise upon encountering each word. Semantic network theory suggests that minds have interconnected structures to help people make connections quickly and use prior knowledge to facilitate comprehension (M.R. Quillian, 1967)—which encourages the linkage of words and ideas. Quillian states that each concept: *a car, to drive, keys,* are nodes made up of around 10-100 neurons in a network in our minds. A node, when activated, can activate related nodes, making inferences. Essentially, each word people read will activate connected ideas, which can drive perception one way or another.

Specifically, negative words can impact perception more than positive words. Negativity bias explains that negative experiences evoke stronger reactions and are processed more thoroughly than positive ones (Rozin and Royzman, 2001). In other words, a single negative event can outweigh several positive events in shaping a person's overall perception and memory. This bias has evolutionary roots, as paying more attention to negative stimuli (eg. tigers, wildfires) historically increased chances of survival. As the media realizes the power of negativity, it puts out overly negative news coverage that sometimes does not match the reality of the world (Soroka et al., 2015).

Aware that language influences perception, many searched for methods to shape understanding by taking advantage of word choice. Today, professionals in mass communications often utilize this deliberate wording design to package and present information to the public to achieve a desired viewpoint. In particular, for environment news, the media tends to oversimplify, misrepresent, or dramatize research results (Lewandowsky et al., 2012) through diction. This may be because scientific details can be difficult to digest or uninteresting for news readers. For example, when writing about a study forecasting future global extinctions as a result of climate change, news reports made the consequences seem more severe and the timescale shorter than actually projected by the study (Ladle Jepson and Whittaker, 2005).

Within news reporting, headlines are effective devices to alter the impression, understanding, and memory of the news content. Research has shown that misleading headlines, including those of factual articles, can influence the recall and interpretation of the article's

content (Ecker et al., 2014). Incorrect perception due to headlines can cause people to have a permanently biased understanding of the news. Studies have shown that corrections rarely eliminate reliance on misinformation, even when people acknowledge and remember the correction. Multiple explanations exist for the continued influence effect. One is the formation of mental models, where people prefer a coherent but incorrect model over an incomplete one (Johnson and Seifert, 1994). Another is retrieval failure, involving source confusion or failure of strategic monitoring processes, which can cause misinformation to be remembered as credible (Ayers and Reder, 1998). Lastly, people resist being told what to think which makes retractions ineffective (Lewandowsky et al., 2012).

Research Methodology

Designing the Surveys

To determine the differences in people's perception of news stories when headlines use neutral versus negative language, two surveys were designed. The headline in Survey 1 used neutral language (eg. reports, unusual) whilst the one in Survey 2 used negative language (eg. warns, catastrophe). The news headlines within each survey were created for the experiment to avoid prior knowledge or bias from real news.

The headline in Survey 1: Scientists Report Climate Impacts as Unusual Weather Patterns Arise
The headline in Survey 2: Scientists Warn of Imminent Climate Catastrophe as Extreme Weather Escalates

Both surveys asked the same three questions:

1. From this headline, estimate the earth's annual temperature increase mentioned in the article.
2. From this headline, estimate the annual percentage increase of natural disasters mentioned in the article.
3. From this headline, estimate the number of annual deaths due to natural disasters mentioned in the article.

All answers were multiple choice with an option for "other, please specify." All responses were collected through Google Forms.

Sampling

In total, 112 people in Bangkok, Thailand were surveyed. 56 responded to Survey 1 and 56 responded to Survey 2. The respondents included individuals from a wide range of educational qualifications, incomes, and ages, to satisfy the purpose of this research. As for age, in Survey 1, 9.1% of respondents are in middle or high school (approximately 13-18 years old),

1.8% are in university (approximately 19-22 years old), 67.3% are working age (approximately 23-60 years old), and 21.8% are retired (above 60 years old). In Survey 2, the numbers were 16.1%, 5.4%, 71.4%, and 7.1%, correspondingly. As for gender, in Survey 1, 67.9% of respondents were male, and 32.1% were female. In Survey 2, 55.4% of respondents were male, 42.9% were female, and 1.8% answered other/prefer not to say.

Survey Findings

Q1	0.05°C	0.10°C	0.15°C	0.20°C	Total
Survey 1: neutral language	10	15	8	23	56
Survey 2: negative language	3	12	8	33	56
Total	13	27	16	56	112

Figure 1: Table displaying responses to Q1 from Survey 1 and Survey 2 respondents

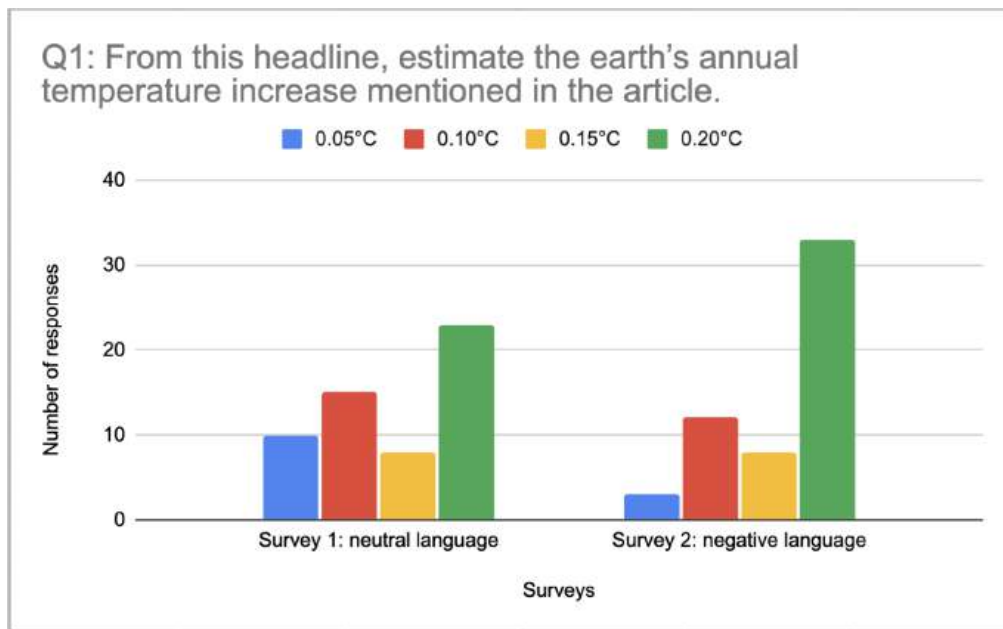


Figure 2: Graph comparing responses to Q1 from Survey 1 and Survey 2 respondents

In Q1, the results of Survey 1 and Survey 2 had similar trends but displayed a notable difference between the respondents' perceptions. The most popular prediction is the same in both surveys, which is 0.20°C or the highest temperature choice. However, the answers were more extremely skewed towards higher temperatures in Survey 2, as 58.9% of Survey 2 respondents chose 0.20°C, but only 41.1% of Survey 1 respondents did. It is also apparent that the lowest value is extremely unpopular in Survey 2, with only 5.4% choosing it. The average of Survey 1 responses was 0.14°C and the average of Survey 2 responses was 0.16°C, which has a percentage difference of 13.3%.

Q2	5-10%	11-15%	16-20%	21-25%	Total
Survey 1: neutral language	18	21	11	6	56
Survey 2: negative language	13	12	16	15	56

Figure 3: Table displaying responses to Q2 from Survey 1 and Survey 2 respondents

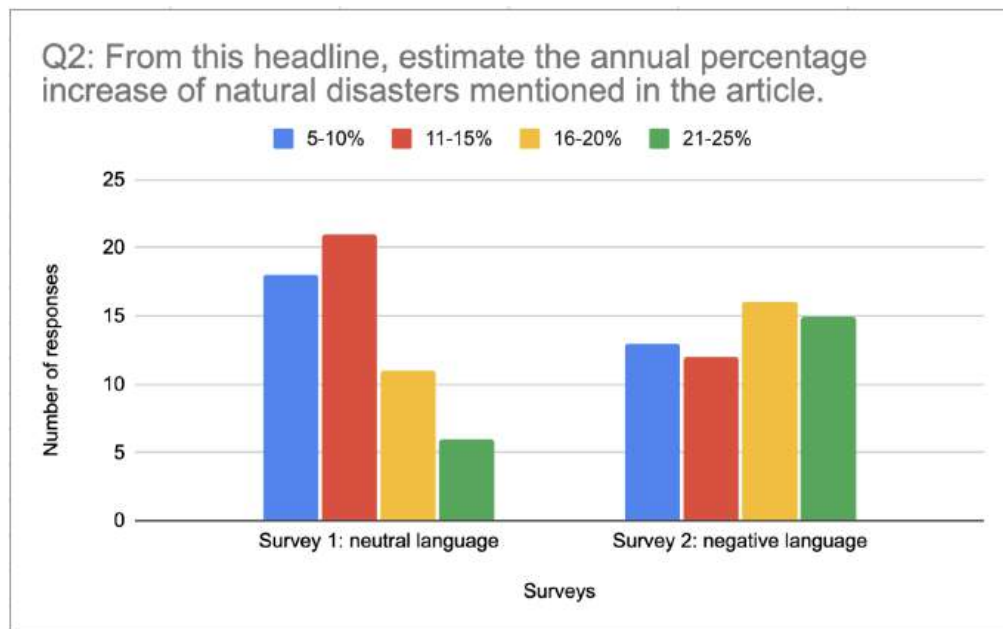


Figure 4: Graph comparing responses to Q2 from Survey 1 and Survey 2 respondents

In Q2, Survey 1's answers were skewed towards lower percentage ranges, whilst Survey 2's answers were more evenly distributed. Survey 1's most popular choice was the lower-leaning range of 11-15%, whilst Survey 2's most popular choice was the higher-leaning range of 16-20%. The average of Survey 1 responses was 13.3% and the average of Survey 2 responses was 15.8% (each average is derived from averaging middle values of each percentage range), which has a percentage difference of 17.5%.

Q3	10,000-30,000	30,001-50,000	50,001-70,000	70,001-90,000	Total
Survey 1: neutral language	19	23	7	7	56
Survey 2: negative language	21	13	7	15	56

Figure 5: Table displaying responses to Q3 from Survey 1 and Survey 2 respondents

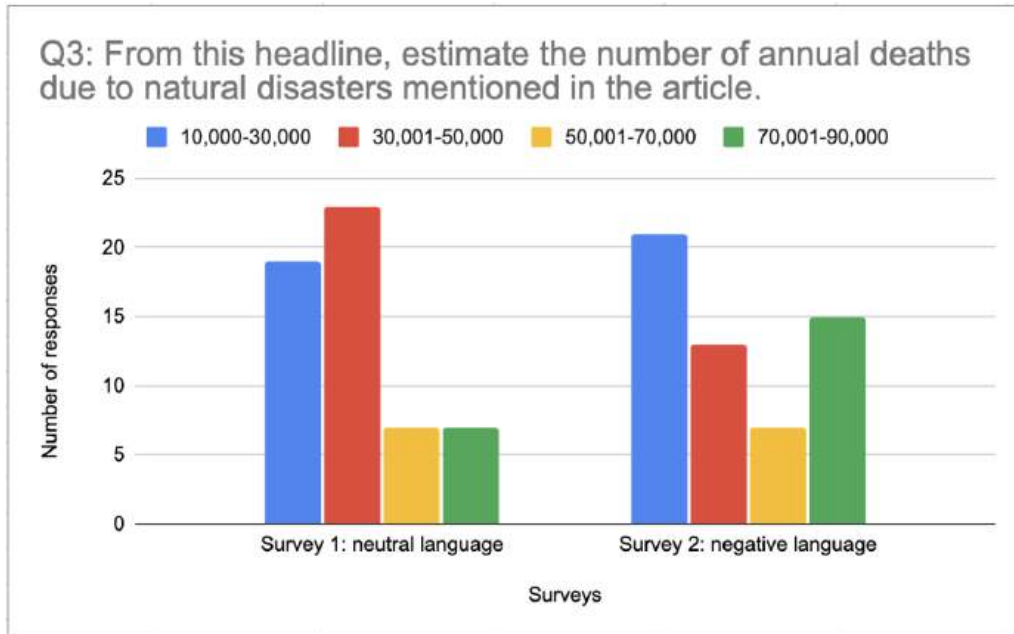


Figure 6: Graph comparing responses to Q3 from Survey 1 and Survey 2 respondents

Interestingly, in Q3, slightly more Survey 2 respondents predicted the lowest range of the number of annual deaths due to natural disasters compared to Survey 1 respondents. Whilst 33.9% of Survey 1 respondents chose the lowest range, 10,000-30,000 deaths, 37.5% of Survey 2 respondents did. In other words, unlike many may assume, the respondents who read negative language were more likely to predict the lowest range of deaths than the respondents who read neutral language in this particular question of this survey.

Resultantly, Survey 2 responses were extreme on both sides whilst Survey 1 responses were skewed towards lower ranges. Notably, 42 respondents of Survey 1 predicted lower-leaning ranges whilst only 34 respondents of Survey 2 did. This could suggest that most respondents of both surveys predicted lower-leaning annual deaths, but negative language also triggered some respondents of Survey 2 to choose the highest range, making the data polarized. Overall, the average of Survey 1 responses was 40,714 and the average of Survey 2 responses was 45,714 (each average is derived from averaging middle values of each death range), which has a percentage difference of 11.6%.

Analysis of Findings

All in all, negative language in headlines evokes substantially more negative perceptions of news articles in comparison to neutral language. In all cases, negative words in headlines triggered more people to make high predictions of the severity of an event compared to neutral words.

As the results of Q1 and Q3 from both surveys display similar trends, it can be presumed that a person's answer is predominantly dependent on their prior knowledge or opinion of the

topic. Perhaps most people heard climate change was bringing extreme weather and assumed the value of the increase would be very high, without considering that small values may be significant in the atmospheric scale. Nonetheless, an independent samples t-test revealed that the responses from Survey 1 significantly differed from those of Survey 2, $t(110) = 2.360$, $p = 0.020$, $d = 0.370$. Also, a chi-square analysis of Q2 responses found a statistically significant difference, $\chi^2(3, N = 112) = 8.044$, $p = 0.045$, $V = 0.268$. Hence, regardless of whether their fundamental beliefs were changed, their immediate decision-making was affected by language.

Even though Q3 did not show statistical significance according to the chi-square analysis, $\chi^2(3, N = 112) = 5.787$, $p = 0.122$, $V = 0.227$, it did show a nuanced insight. While negative language typically leads people to make more negative predictions, in this case, it also unexpectedly leads some people to make more positive predictions. This suggests that negative language can sometimes prompt a counter-reaction, causing people to consider less dire outcomes. This could be due to the backfire effect, which is when people become more entrenched in their beliefs after they have seen new information that firmly contradicts them (Swire-Thompson et al., 2020). Two types of backfire effects could potentially explain the phenomenon: the familiarity backfire effect and the worldview backfire effect. The worldview backfire effect happens when someone feels compelled to defend their worldview after receiving information that contradicts their beliefs. On the other hand, the familiarity backfire effect happens when false information is reiterated within the retraction, strengthening the belief. However, it is to be noted that the backfire effect has been difficult for researchers to find and replicate. Perhaps the results of this experiment could hint at the existence of the backfire effect in certain conditions. In summary, neutral language led to more moderate estimates, whereas negative language caused polarization, with respondents divided between very low and very high predictions. This highlights the complexity of human behavior and the importance of understanding how different framing effects can influence perceptions in varied ways.

Another interesting angle to observe is the survey results compared to real-world data. In general, respondents who read neutral language predicted values closer to real-world data. For Q1, the real-world answer is approximately 0.08°C (calculated from the average global temperature in 2020, $1.20 \pm 0.1^\circ\text{C}$ above the pre-industrial level (WMO, 2021), and the average global temperature in 2023, $1.45 \pm 0.12^\circ\text{C}$ above the pre-industrial level (WMO, 2024)). The average of Survey 1 responses was closer to 0.08°C at 0.14°C compared to the average of Survey 2 responses at 0.16°C . Furthermore, 0.08°C is closest to 0.10°C , which was chosen by three more respondents who read neutral language. Slightly better results are revealed in Q2. For Q2, the answer was 9.1% (calculated from the increase to “28 weather and climate disasters in 2023” from “the previous record of 22 in 2020” (Smith, 2024)). The average of Survey 1 responses was 13.3%, which is closer to 9.1% than the average of Survey 2 responses at 15.8%. Moreover, 9.09% was within the range of 5-10%, which was chosen by five more respondents who read neutral language. Additionally, the most popular choice in Survey 1 is 11-15%, closer to 5-10% than the most popular choice in Survey 2, 16-20%. The clearest results lay in Q3. For Q3, the

answer was 40,000-50,000 per year (Ritchie and Rosado, 2024). The average of Survey 2 responses was 45,714, right around the middle between the real-world range whilst the average of Survey 1 responses was 40,714. Here, the most popular answer for respondents who read neutral language, 30,001-50,000, matched with real-world data, but the most popular answer for respondents who read negative language did not. It could be interpreted that the similarities between survey values and real-world values correlated with the question's relevance to the headline. Question 1 was about temperature, which is most directly linked to the headline. Question 3, however, was about deaths, which requires double-layer thinking when related to the headline. From this research, it is clear that neutral language encourages more accurate perceptions of news stories.

Limitations

This study has several limitations that must be acknowledged. One limitation concerns the interpretation of responses gathered through the survey. The use of multiple-choice questions, even with the option for “other, please specify,” may not fully capture the depth of participants' perceptions. Incorporating more detailed open-ended questions in future studies could capture the nuances of their interpretation of the headlines and offer richer insights into how language framing affects perceptions.

Another significant limitation is the potential influence of participants' prior knowledge and opinions about the environment. This could confound the effects of word choice, making it challenging to isolate the impact of neutral versus negative language on their perceptions. To mitigate this limitation, future research could assess participants' baseline knowledge and attitudes, allowing for a more controlled analysis. Additionally, expanding the scope to include a broader range of topics could help isolate the effects of language framing more effectively.

Conclusions and Recommendations

This study reveals that word choice significantly influences public perception of climate-related news and likely other types of news. Generally, negative headlines led to higher predictions of event severity, whereas neutral headlines resulted in more moderate estimates. Occasionally, though, negative language results in polarization, with most people choosing extremely negative or extremely positive answers, indicating a complex interaction between language and perception. In addition, comparisons with real-world data showed that predictions based on neutral language were closer to actual values, underscoring the effectiveness of neutral language in promoting realistic perceptions.

These findings suggest that media outlets should adopt guidelines for balanced headline writing to avoid sensationalism and encourage accurate public understanding. Journalism training programs could include modules on the impact of language framing to sensitize future journalists to these effects. This approach can help mitigate the polarization and fear induced by negatively

framed headlines. Furthermore, educating the public on language framing and promoting media literacy can empower individuals to critically evaluate news stories.

Future research should explore diverse contexts to generalize these findings. For example, studies could use a wider range of headlines to test the effect of word choice on the perception of various topics of news stories. Cross-cultural studies could provide insights into how people from different countries interpret and respond to language framing in the news. Additionally, examining the impact of positive headlines compared to neutral and negative ones could offer a more comprehensive understanding of language effects. Sharing these findings can foster a more informed and less polarized public, ultimately enhancing the quality of news consumption and discourse.

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Understanding the Role of Alpha-Synuclein in Parkinson's Disease and Dementia with Lewy Bodies By Yara Walker

Abstract

Parkinson's disease (PD) is the second most pervasive neurodegenerative disease after Alzheimer's disease (AD) and the most widespread synucleinopathy, characterised by the clustering of alpha-synuclein (a-syn) in intracellular inclusion bodies known as Lewy bodies (LB). Much like PD, Dementia with Lewy Bodies (DLB) is a prevalent neurodegenerative dementia marked by gradual deterioration in cognitive abilities and a range of symptoms overlapping significantly with AD and PD. A-syn, a 140 amino acid protein encoded by the SNCA gene, is primarily manifested in the presynaptic terminals of neurons. It is composed of three areas: the amphipathic N-terminal region, the central hydrophobic region, and the C-terminal region. In PD, a-syn misfolding and aggregation form LB and Lewy neurites (LN), disrupting cellular processes and leading to neuron dysfunction and death. In DLB, a-syn aggregation shows a distinct cortical distribution pattern, contributing to cognitive impairment. LB in DLB consists of a-syn fibrils surrounded by various proteins, disrupting cellular function. While DLB and PD share similarities, including a-syn aggregation, the diseases exhibit distinct clinical manifestations. Symptoms include motor issues such as tremors and slow movement, as well as non-motor issues, namely autonomic dysfunction and cognitive impairments. Emerging therapeutic approaches targeting a-syn, such as GV-971 and TLR4 agonists such as MPLA, offer promising avenues for following disease progression and improving patient outcomes. This review explores the disease progression of PD as well as DLB, focusing on the role of a-syn in these disorders. In addition, it highlights the protein's potential therapeutic strategies. Continued research is essential for developing effective treatments.

Introduction

PD is the second most pervasive, progressive neurodegenerative disorder after AD (Olivares et al.). Additionally, PD is the most widespread synucleinopathy, meaning the disease is distinguished by the clustering of a-syn, otherwise known as LB in intracellular inclusion bodies (Dunning et al.). The disease was originally identified and described by the medical practitioner, Dr. James Parkinson in 1817 (Lewis). Today, PD affects approximately 1% of people over 60 and is primarily known for its rapid cognitive decline as well as its effect on motor function inflicted by the depletion of dopamine-synthesising cells in the substantia nigra (Alarcón et al.; Cory-Slechta et al.). Much like PD, DLB is a prevalent kind of neurodegenerative dementia. It manifests in older individuals but is often overlooked and commonly misdiagnosed. DLB is characterised by the gradual spread of LB and LN. On a clinical aspect, DLB is marked by a progressive deterioration in cognitive abilities and presents a range of clinical signs that overlap significantly with those of AD and PD (Chin et al.). DLB was first discovered by Dr. Friedrich Lewy in the early 20th century. He identified abnormal protein deposits in the brains of individuals with Parkinsonian and Dementia symptoms (Liu et al.).

Following AD, DLB is the second most prevalent neurodegenerative condition that results in dementia (Prasad et al.). A-syn is a protein of 140 amino acids, encoded by the SNCA gene and primarily expressed in the presynaptic terminals of neurons in the brain. Consequently, presynaptic terminals transmit signals through neurotransmitters from one cell to another (Takahashi). This review will explore the pathogenesis of PD and DLB through a comprehensive investigation of symptoms caused by the protein a-syn. Additionally, the function and role of a-syn in these disorders will be examined as a source of potential therapeutics.

Alpha-Synuclein Structure and Distribution

A-syn is composed out of three distinguished regions: the amphipathic N-terminal region, the central hydrophobic region and the C-terminal region (Kim et al.). The amphipathic N-terminal region is positively charged and contains both hydrophobic and hydrophilic elements. This facilitates its interaction with lipid bilayers in cells. The binding to these cellular membranes allows a-syn's role in the discharge of neurotransmitters and the recycling of synaptic vesicles (Emamzadeh; Kang and Sun; Arakaki et al.). The central region is composed by a hydrophobic amino acid sequence. Its role involves the formation of insoluble fibrils, which are fundamental elements of neurodegenerative diseases such as PD. The domain promotes the assembly of a-syn into pathological structures, consequently contributing to neuronal impairment and cell death (Vidović and Rikalovic; Arsito et al.). Finally, the C-terminal region modulates protein interactions and influences their conformation. The domain regulates the protein's role in vesicle trafficking and neurotransmitter release, ultimately affecting its aggregation properties (Gallardo et al.; McClain et al.).

Distribution and Impact in the Brain

High levels of the protein are found in parts of the brain such as the neocortex, the hippocampus, the substantia nigra, the thalamus and the cerebellum (Kim et al.). Nonetheless, discoveries have shown that a-syn is not exclusively expressed within the brain. It can be observed in nervous tissue, red blood cells and more (Nakai et al.). For instance, mRNA has been proven to contain low levels of a-syn, which generates additional core cellular activity beyond its standard impact on the brain (Baltic et al.; Burré; Askanas et al.).

Role in synaptic vesicle trafficking

A-syn plays a multifaceted role in synaptic vesicle trafficking, which is essential for neuronal communication (Román-Vendrell et al.). First and foremost, a-syn regulates the synaptic vesicle pool. It maintains the balance and recycling of synaptic vesicles for efficient neurotransmitter release (Zaltieri et al.). Additionally, a-syn links lipid membranes via its N-terminal region to stabilise vesicles and aid their fusion with the presynaptic membrane (Lai et al.). It also interacts with SNARE complex proteins by assisting in the merge of vesicles with the presynaptic membrane (Agliardi et al.). Moreover, a-syn ensures vesicles are refilled and prepared for neurotransmitter release through endocytosis and affects vesicle mobility within the

presynaptic terminal. This helps their distribution and availability for release (Huang et al.). Nonetheless, when faced with the disease, the clustering of a-syn disrupts these functions, resulting in synaptic dysfunction and neurodegenerative pathology (Peña-Bautista et al.).

The Role of Alpha-Synuclein in Parkinson's Disease

A-syn takes on a substantial role in the progression of PD. LB as well as LN are formed by misfolded a-syn in neurons and constitute a hallmark of PD pathology (Peña-Bautista et al.). First and foremost, triggers such as genetic mutations, environmental factors, or age-related changes influence the initiation of the misfolding process (Picca et al.; J. Ma et al.; Salahuddin et al.). Some of these environmental conditions include the presence of toxins and oxidative stress. For example, a-syn clumps have been shown to form in environments with elevated levels of reactive oxygen species (ROS), and in turn, a-syn increases ROS production. Additionally, cytochrome as well as hydrogen peroxide, both sources of oxidative stress, are areas where a-syn tends to clump more frequently (Dunning et al.). Once a-syn molecules misfold, they are likely to aggregate and form small clusters called oligomers. These oligomers then continue to combine, creating larger, insoluble structures known as fibrils (Casella et al.; Alam et al.). Subsequently, the fibrils propagate from one cell to another via mechanisms such as cell-to-cell transmission, thereby perpetuating the misfolding process (Luk et al.). Consequently, the buildup of insoluble a-syn aggregates disrupts regular cellular processes and may lead to brain cell death, particularly those crucial for motor control (Guo et al.). Moreover, lysosomal dysfunction may occur, resulting in the accumulation of abnormal a-syn within cells (Papagiannakis et al.). This underscores the pivotal role dysfunctions in both a-syn aggregation and lysosomal pathways have in the development and progression of PD. These a-syn clumps disrupt multiple cellular processes, ultimately contributing to neurodegeneration and affecting both the function and homeostasis of a-syn within the cell (Dunning et al.).

Spread of Pathology

Per the Braak hypothesis, PD pathology spreads in a predictable pattern throughout the brain. It is conjectured that the spread begins from the olfactory bulb or the dorsal motor nucleus of vagus in the caudal medulla. After that, it progressively moves to the medulla, continues towards the pontine tegmentum and other midbrain areas, including the substantia nigra, affecting dopamine neurons (Visanji et al.). Furthermore, the pathology advances to the cortex, extending its reach throughout the central nervous system. However, the spread to the spinal cord only occurs after the central nervous system has already been affected. Consequently, the spinal cord is not considered a probable pathway for the initial spread of the disease (Rietdijk et al.). In summary, PD pathology is conjectured to spread from the peripheral nervous system to the central nervous system.

Symptoms and Progression of Parkinson's Disease Related to Alpha-Synuclein Pathology

As previously stated, PD is a chronic and gradual neurodegenerative condition defined by worsening symptoms over time, often resulting in increased disability. A-syn pathology significantly influences both the symptoms and progression of PD. Patients typically experience two categories of symptoms: motor symptoms, affecting movement, as well as non-motor symptoms, involving other bodily functions (Yang et al.).

Motor Symptoms.

Motor symptoms include tremors, rigidity, bradykinesia, postural instability etc. In PD patients, motor symptoms appear due to a decrease of dopamine levels following the neuron dysfunction and death caused by the a-syn aggregates (Zaltieri et al.; Müller). Dopamine is essential for smooth muscle movement control meaning that decreased dopamine levels result in an imbalanced neuronal signalling in the basal ganglia (Paul; Zhai et al.). This consequently leads to a dysfunction in the basal ganglia which affects motor control, coordination, and contributes to the motor symptoms (Zhong et al.). This also applies to bradykinesia, a symptom characterised by slowness of movement and compromised motor function (Hasan et al.; Lemos et al.). The reduced dopamine levels also contribute to increased muscle stiffness and rigidity. Finally, these dysfunctions lead to postural reflex impairments, contributing to the development of postural instability (Hendy et al.).

Non-Motor Symptoms.

Non-motor symptoms are made up of autonomic dysfunctions, cognitive impairments, mood disorders and sleep disruptions. Autonomic dysfunctions appear when the decline of neurons inflicted by the accumulation of a-syn affects regions involved in autonomic function. These regions include the enteric nervous system and autonomic ganglia (Calabresi et al.). Dysfunction in the enteric nervous system affects gastrointestinal motility, leading to constipation while the dysfunction in autonomic ganglia affects blood pressure regulation, contributing to orthostatic hypotension (Dommershuijsen et al.; Ghalandari et al.). Cognitive impairments, namely memory and executive function deficits can arise from the presence of a-syn aggregates in parts of the brain associated with cognitive function. These include the cortex and hippocampus (Henderson et al.; Gargouri et al.; Oltra et al.). Cognitive processes and neurotransmitter systems, including dopamine and acetylcholine, which are vital for memory and executive function, are disrupted by a-syn pathology (Sanchez-Catasus et al.). This ultimately results in deficits in memory formation, retrieval, and executive function (Blatt et al.). Furthermore, a-syn aggregates affect regions involved in mood regulation, such as the limbic system and prefrontal cortex (Alwani et al.). These crucial mood regulation pathways face a deficit of neurons and dysfunction due to the accumulation of a-syn. Neurotransmitters involved in mood regulation, such as serotonin and norepinephrine face an imbalance due to the pathology which leads to neuronal dysfunction. This contributes to the progression of common mood disorders including depression and anxiety (Werner and Olanow). Lastly, a-syn aggregates in

brain regions that control sleep-wake cycles, such as the brainstem and hypothalamus. The build up of a-syn in these regions leads to their degeneration which disrupts sleep regulation pathways (Thangaleela et al.; Baschieri et al.). Furthermore, serotonin and dopamine, both influential in regulating sleep, are targeted neurotransmitters affected by a-syn pathology (Davla et al.; Rothman et al.). Once again, this leads to neuronal dysfunction and neurotransmitter imbalances which contribute to rapid eye movement, otherwise known as REM, sleep behaviour disorder (Nigam et al.).

Alpha-Synuclein in Dementia with Lewy Bodies

Lewy Body Formation and Composition

LB are formed following the misfolding of a-syn proteins into small clusters called oligomers which later aggregate further to form larger, insoluble fibrils. These fibrils make up the central core of LB (Guerrero-Ferreira et al.). Moreover, ubiquitin is a protein that tags other damaged proteins for degradation. It incorporates itself into the LB, marking the aggregated proteins for removal (Alexopoulou et al.). Additionally, neurofilament proteins originating from the neuronal cytoskeleton entangle themselves within the growing LB, which eventually contributes to the pathological structure characteristic of DLB (Hill et al.). Lastly, various enzymes such as proteases become trapped in the LB due to the aberrant a-syn aggregation, disrupting their normal cellular functions (Wennström et al.). In summary, the mature LB consists of a dense core of a-syn fibrils, surrounded by a halo of ubiquitin, neurofilament proteins, and associated enzymes. These components collectively disrupt cellular function, contributing to neuronal dysfunction and cell death in DLB.

Comparison with Parkinson's Disease Pathology

Although PD and DLB are very similar and sometimes have overlapping elements, there are significant differences that separate them. While PD and DLB share similarities and occasional overlap, notable distinctions can set them apart. The main overlapping factor consists of the build up of misfolded a-syn and therefore the synthesis of LB and LN. A-syn pathology begins in brainstem nuclei in both PD and DLB whereas its spread in the cortical regions differs (Guerreiro et al.). Both DLB and PD involve dysfunction of neurotransmitter systems, particularly dopamine, contributing to motor as well as non-motor symptoms (Stockbauer et al.; Nemade et al.). In DLB, a-syn pathology spreads extensively to cortical regions, contributing to cognitive impairment, whereas in PD, it predominantly affects subcortical structures such as the substantia nigra (Tysnes and Storstein; Emre). Furthermore, cognitive deterioration symptoms emerge early during DLB, whereas cortical pathology usually develops later in the progression of PD (Gallagher and Schrag; Zhang et al.). Moreover, a multitude of symptoms seem to be more prevalent in DLB rather than PD. These include, visual hallucinations, fluctuations in cognition, REM sleep behaviour disorder etc. Motor symptoms are less severe in LBD, while non-motor symptoms are more common compared to PD (Fei et al.; Wang et al.). Lastly, both diseases have

distinct diagnostic criteria, with DLB requiring the presence of dementia symptoms within one year of parkinsonism onset, while PD focuses on motor symptoms with later cognitive decline (Marsili et al.; Walker et al.).

Unique Aspects of Alpha-Synuclein Aggregation in Dementia with Lewy Bodies

It is worth highlighting certain unique aspects of a-syn aggregation in DLB. For instance, its aggregation exhibits a distinct cortical distribution pattern, one of which begins in the brainstem, like PD (Ye et al.; McKeith et al.). Furthermore, another unique aspect of a-syn aggregation in DLB is its distinctive pattern of spread throughout the brain. However, unlike PD, in DLB, a-syn pathology spreads extensively into the cortex through neuronal pathways, axonal transport, and prion-like propagation (Imamura et al.; Menon et al.; Weber et al.). The clumps spread transsynaptically and retrogradely along neuronal pathways and across synaptic connections (Mezias et al.). These cortical regions are responsible for cognitive functions, which deteriorate due to the pathology and lead to some of the most prevalent dementia symptoms such as visual hallucinations (Taylor et al.; Dudchenko and Vasenina).

Symptoms of Dementia with Lewy Bodies Related to Alpha-Synuclein Pathology

The aggregation of a-syn is central to the symptoms of DLB. The specific symptoms vary based on the brain regions in which these clumps form. For example, a-syn pathology in the cortex and hippocampus often disrupts cognitive processes responsible for cognitive recall, executive function, and visuospatial perception (Lim et al.). Moreover, when the aggregations migrate towards the occipital and temporal lobes, it leads to dysfunction in visual processing pathways. Consequently, this results in the perception of visual hallucinations (Perini et al.). Furthermore, the presence of a-syn in the brainstem and hypothalamus has consequences on the sleep-wake cycles. LB affects brainstem nuclei responsible for regulating REM sleep. This results in abnormalities in REM sleep regulation, including acting out dreams (Matar et al.). Finally, if the aggregations find their way into the autonomic ganglia and spinal cord, autonomic functions such as blood pressure and bladder control are targeted. This orthostatic hypotension can lead to dizziness, lightheadedness, or even fainting (Orimo; Isik et al.).

Alpha-Synuclein as a Biomarker

Given its central key involvement in the pathology of PD and DLB, a-syn levels may serve as markers for disease presence and progression (Ganguly et al.). The different types of a-syn biomarkers can be distinguished as follows: Cerebrospinal Fluid (CSF), blood plasma and tissue biopsies.

Cerebrospinal Fluid.

When the misfolded a-syn is released from neurons into the extracellular space, it enters the CSF through the brain's interstitial fluid (Menéndez-González et al.). The concentrations of a-syn in CSF can be monitored using approaches like enzyme-linked immunosorbent assay

(Mollenhauer et al.). These measurements show the changes in a-syn levels in the CSF and are used as biomarkers to aid with diagnosis and monitor any progression (Majbour et al.).

Blood Plasma.

Moreover, when released into the extracellular space, the misfolded a-syn enters the bloodstream. Then, it circulates in the blood plasma, either freely or bound to blood cells and other proteins (Foulds et al.). To monitor a-syn levels, blood samples are collected from patients measured using techniques such as ELISA or mass spectrometry (Barbour et al.; Zhao et al.). These measurements show the variations in a-syn levels in the blood plasma and are used as biomarkers to aid with diagnosis and monitor any progression (Kluge et al.).

Tissue Biopsies.

Additionally, biopsies can be taken from tissues such as the skin, salivary glands, or gastrointestinal tract, where a-syn aggregates can be present (Yan et al.; L. Y. Ma et al.; Gibbons et al.). The samples are then stained using specific antibodies that bind to a-syn and examined under a microscope to detect and quantify a-syn aggregates (Tsukita et al., 2019). These observations are once again used as biomarkers to aid with diagnosis and help distinguish PD and DLB from other neurodegenerative disorders (Coughlin & Irwin, 2023). Despite these biomarkers, there are still several challenges when it comes to studying a-syn. The different methods used to measure a-syn levels can produce inconsistent results. There is a need for standardised methods to ensure reliable and comparable data. Addressing these challenges is essential for accurate research and diagnosis. In conclusion, the study of a-syn's role in PD and DLB highlights its critical involvement in the pathology and progression of the neurodegenerative disorder. A-syn aggregates disrupt cellular processes and lead to neuronal dysfunction and death, particularly in regions essential for motor control and cognitive function. In summary, LB in DLB consists of a-syn fibrils surrounded by various proteins, disrupting cellular function. While DLB shares similarities with PD, there are unique aspects contributing to DLB's diverse symptomatology, affecting cognitive, visual, and autonomic functions.

Therapeutic Approaches Targeting Alpha-Synuclein

Throughout recent years, a-syn studies aiming to modify disease progression in conditions such as PD and DLB have grown substantially. In a study published in 2024, researchers tested GV-971's ability to reduce a-syn protein clumps using lab tests and brain samples from PD and DLB patients. They also assessed motor skills in mice treated with GV-971. GV-971 is derived from seaweed oligosaccharides. Results showed that GV-971 successfully stopped a-syn clumping and broke down existing clumps. Moreover, it protected neurons from damage and reduced harmful cell particles. An improvement in motor function was also observed in the mice (Yu et al., 2024). Another study suggests that Toll-like receptors, which play a role in the immune system, help clear a-syn. Researchers hypothesised that using MPLA, a TLR4 agonist, could potentially enhance a-syn clearance without causing harmful

inflammation. They divided six-month-old PLP-a-syn mice into four groups and gave them weekly doses of LPS, MPLA, or a control solution for twelve weeks. Results showed a boost in a-syn uptake by microglial cells, which induced motor function improvement without triggering widespread inflammation. All in all, the study aims to highlight the positive outcomes of long-term MPLA treatment in transgenic PLP-a-syn mice, suggesting its potential as a promising option for pathology-modifying trials in a-syn-related diseases such as DLB or PD (Venezia et al., 2017).

Conclusion

In conclusion, a-syn is crucial in the development of PD and DLB, disrupting cellular function as well as contributing to neuronal dysfunction and death. While both diseases share similarities in a-syn aggregation, they exhibit distinct clinical manifestations. Emerging therapeutic approaches and studies targeting a-syn offer promising avenues for mitigating disease progression and improving patient outcomes.

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Neurology of Music By Vivian Younger

Introduction

Music has been used to connect and inspire since prehistoric times, and continues to permeate all aspects of human life - from entertainment and culture, to personal emotional experience. Even tens of thousands of years ago, the Sumerians used music to enhance their worship rituals and communicate with one another (“Music of Ancient Times,” 2015). Yet, we do not have a comprehensive understanding of a) how it is processed in the brain, b) how we have individual differences in our music preference, and c) how music impacts our brains long term. Here we review recent literature and studies to examine how musical aspects such as beat, tone and pitch are processed within the brain, and how having a comprehension of these things can help navigate our understanding of musical preferences. Past work has solely investigated the neural mechanisms involved in processing single musical elements without (1) accounting for how they may rely on similar (or differing) neural substrates and without (2) connecting them to the psychology of why we may prefer them. Through this review we aim to further explore this relationship.

Music is composed using multiple different elements and features, including pitch, beat, melody, rhythm, tempo and tone. These terms are referenced in the table below, along with their definitions. Each feature requires different neurological functions for them to be cognitively processed. This paper will focus on pitch, beat and tone.

Term	Explanation
Pitch	The quality of a sound, governed by the rate of vibrations producing it; the degree of highness or lowness of a tone.
Beat	A main accent or rhythmic unit in music. Can be a unit of time or pulse of sound.
Melody	A melody, also tune, voice or line, is a linear succession of musical tones that the listener perceives as a single entity.
Rhythm	The placement of sounds in time within a piece of music. Rhythm is a strong, repeated pattern of movement or sound.
Tempo	The pace or speed at which a section of music is played.
Tone	Tone is the use of pitch in language to distinguish lexical or grammatical meaning. This is commonly used to convey emotion or settings in music.

Neural correlates supporting music

How sound gets from the ear to the brain

The auditory system is fundamental to identifying sounds. This system consists of two major parts- the brain and the ears. Sound is processed starting from the outer ears as it makes its way into our brains, where it involves multiple neural regions with differing functions and can sometimes even trigger emotional responses. Sound waves enter the outer ear, also known as the pinna, and are then channeled into the ear canal. The ear canal leads to the eardrum, which vibrates from the sound waves. This area is known as the middle ear. The vibrations from the eardrum are sent to three bones called the ossicles within the middle ear, known as the malleus, incus, and stapes, which amplify these vibrations (see Figure 1). These vibrations are then sent to the cochlea, a snail-shaped structure filled with fluid within the inner ear. The cochlea contains 20-30,000 hair cells which actually process tones and pitches from sounds. When the fluid within the cochlea is triggered from vibrations, it creates a ripple effect, which bends the hair cells, opens them up like pores, and allows a certain chemical to rush into the cells. This translates the vibrations into electrical signals. These electrical signals are transmitted to the brain with neurons, via the cochlear nerve system. The base of the hair cells is the basilar membrane, arguably the most important part of the cochlea, which has graded stiffness properties and thus does these vibrations. These make their way into the cerebral cortex, where the understanding of sound takes place. This further activates multiple regions of the brain related to rhythm, pitch, tone, and even emotion, which will be discussed below.

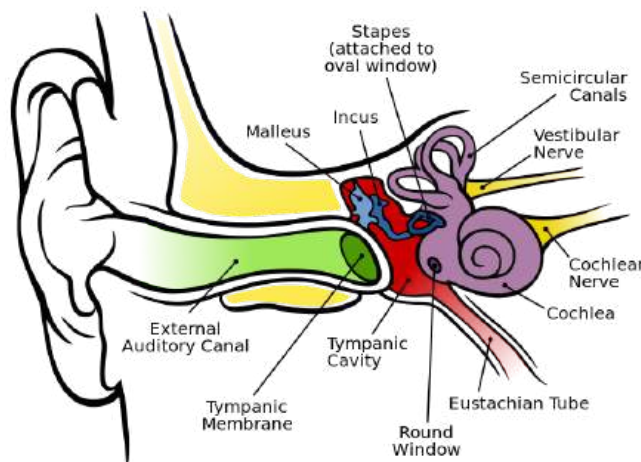


Figure 1. This figure shows the auditory transduction pathway discussed above.

Figured from: Chittka L, Brockmann A (2005) Perception Space—The Final Frontier. PLoS Biol 3(4): e137.

<https://doi.org/10.1371/journal.pbio.0030137>

Tone processing in the brain

Tone is the use of pitch in language to distinguish lexical or grammatical meaning. This is commonly used to convey emotional settings in music. The processing of tone in the brain is primarily carried out within the auditory cortex. The inferior frontal lobe evaluates the tone of one's voice, and the premotor cortex determines the vibrations of vocal chords (Sammler et al 2015). This helps the brain determine the pitch in one's voice. When understanding tone, the

right hemisphere of the brain generally processes pitch whereas the left hemisphere mediates the processing of linguistic information (Zatorre & Gandour, 2008). Thus, both hemispheres have been found to work together when processing tone. Lexical tone perception has also been found to activate the left pre-cingulate gyrus subregion for tonal language speakers, as well as other auditory elements such as prosody. Tonal language speakers also had activation in the left superior temporal gyrus when analyzing lexical tone (Liang et al 2018). Tonal languages make up most of the world's languages. These are languages that use the pitch of different syllables to determine differences between words (Linguist Tunes In To Pitch Processing In Brain, n.d.).

The majority of research on tone has focused on how there may be differences in tone processing between musicians and non-musicians. We would expect musicians to differ when it comes to processing music because they have more experience recognizing different musical components and differentiating even the smallest changes in pitch, tone, etc. Musso et al., (2020) investigated how tones may be processed differentially between musicians and non-musicians. They identified that musicians use regions while processing music that are typically used for other purposes, such as speech, like the left posterior superior temporal sulcus (STS), superior temporal gyrus-3 (STG3), and fusiform gyrus (FG). The authors suggest that this could be due to functional adaptation within these regions as musicians continued practicing and building up experience.

Beat and Rhythm Processing in the Brain

Binaural (using both ears) beats are the sounds we can conceptualize after our brain processes two tones at slightly different frequencies in both ears. Binaural beats are what we actually hear when listening to music. These beats are processed in the brain by two different sound frequencies that are introduced to each ear separately. The sound waves created by each beat are processed in the superior olivary nucleus (see Figure 2) (LIONSTALKSCIENCE, 2022). The resulting frequency is the difference between the two sound waves. This is where a beat forms. We perceive beats at a range of 250 ms - 2s (Patel & Iversen, 2014). Beat perception has been found to require interaction between two regions of the brain - auditory regions and motor planning regions. Beat perception primarily relies on a simulation of periodic action in motor planning regions of the brain, and on bidirectional signaling between these regions and auditory regions (Kasdan et al., 2022).

Rhythm is a strong, repeated pattern of movement or sound and is processed similarly to beat. The belt and parabelt are located on the right side of the brain within the superior temporal gyrus and are responsible for processing rhythm. The cerebellum is involved in beat and rhythm processing, as well. The cerebellum plays a major role in accurate and predictive timing. As all these regions work together, the brain is able to make highly accurate temporal predictions when it comes to processing these musical elements.

Pitch processing in the brain

Pitch refers to the quality of a sound, governed by the rate of vibrations producing it; the degree of highness or lowness of a tone. We detect pitch when a sound is harmonic and by how

temporally periodic it is. The brain determines the frequency of harmonies by detecting the spatial pattern of excitation along the cochlea's basilar membrane (see section: “How does sound get from our ears to our brains?”). There are multiple theories as to how the brain turns sound into pitch. The most reliable and evidential theory is the temporal theory, which is that we process all the overtones of a sound and condense it into one periodic signal. This signal is the inverse of the frequency we perceive (Yost, 2009). Pitch perception begins at the cochlea, inside the ear, and travels into the brainstem. The signal travels from the brainstem into the auditory cortex where it is processed in the brain's pitch center, which has not yet been discovered. Research has yet to find where exactly the processing occurs, yet there is an understanding of which parts of the brain have different functions relating to pitch. The secondary auditory cortex is responsible for early processing intervals of melody and harmony (Peretz & Zatorre, 2005). Pitch chroma is processed in the anterior part of the temporal lobe, and pitch height in the posterior part (Griffiths 2014). Pitch chroma tracks information according to one sound source whereas pitch height is used to separate sources. The superior temporal gyri is involved with consonants and dissonance.

Brain Region Map

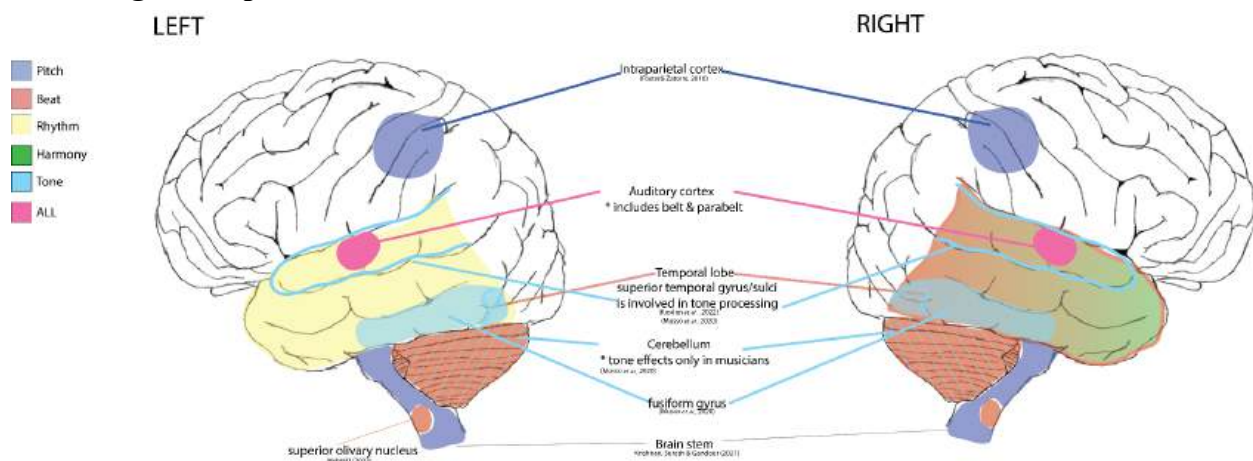


Figure 2. Regions labeled and colored in on this map indicate regions that have been linked to the corresponding musical feature. Notably, the majority of musical properties rely on the temporal lobes, brainstem and cerebellum. Other work (not reviewed here) has shown a role of frontal regions in the emotional responses to music, which may be tied to certain musical properties (ex: faster rhythm, more of an emotional response), but functional responses in these regions are not specific to music (i.e., they will respond to other emotionally-inducing stimuli).

Articles cited: (Binaural Beats: What Are They and What Are the Benefits?, n.d.; Foster & Zatorre, 2010; Kasdan et al., 2022; Krishnan et al., 2017; Musso et al., 2020)

Psychology of Music

Given that human brains largely process musical features in the same way, how do we wind up with different music preferences? And, what are the psychological effects of listening to music? The next three sections will explain how personality affects music preference, how

music affects cognitive function and why we experience a physical sensation when listening to music.

Personality and Musical Preference

The Big 5 is a broad psychological trait measurement used to categorize the different dimensions of one's personality (John et al., 2012). These include Openness, Conscientiousness, Extraversion, Agreeableness, and Neuroticism, commonly abbreviated as OCEAN. These traits have been used by neuroscientists and psychologists to research the effect personality has on one's music taste. Vuoskoski & Eerola (2011) conducted a study that showed that highly neurotic individuals (as judged by their scores on OCEAN trait measures) as compared to all other tested personality groups perceived certain music pieces to be more emotionally salient (sad). Within the same study, music with high emotional intensity was found enjoyable for those who scored highly in Agreeableness, Extraversion, and Openness, whereas those who scored highly for Neuroticism and Conscientiousness did not enjoy the emotionally salient music. Individuals scoring high on Openness had unique results throughout the same study, as it was shown to be the only trait where participants experienced “chills,” and other transient emotional responses while listening to music. Those who are open were found to be more likely to have profound and intense listening experiences (see Figure 3).

A study done by David Greenberg revealed numerous correlations between personality and musical preference by asking over 4,000 participants a number of psychological questions and tests (Greenberg et al., 2015). Participants with high scoring empathy tended to prefer mellow music, such as R&B and soft rock. The empathists were distasteful to more intense music, including heavy metal and punk. People who scored highly on systemizing, rather than empathizing preferred just the opposite- music with more intensity and energy, like punk and heavy metal. Overall, this work indicates that personality can alter one's emotional response to music, and that there is a correlation between being open and experiencing a physical sensation, such as chills, while listening to music. This work also revealed that an individual's highest scoring OCEAN traits correlated with the genre of music they enjoy.

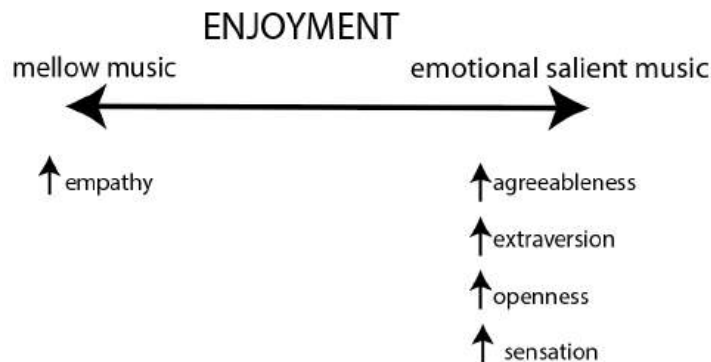


Figure 3. Summary of the interaction between psychological traits and responses and music.

Physical sensations & music

Experiencing a physical sensation while listening to music is a complicated experience that occurs due to a list of different factors, which I detail here. Those who are more creative with higher intellectual curiosity and openness were found to experience these sensations, such as chills, at a higher rate than those who are not. A study conducted by the University of Southern California found using MRI scans that those who felt a chill had a higher volume of fibers connecting their auditory cortex to the areas that process emotion than those that did not feel a chill when listening to music (Sachs et al., 2016). Experiencing chills or goosebumps as a response to music is a phenomenon that is known as frisson. A French study using EEG scanning found that frisson caused increased activity in the orbitofrontal cortex, the supplementary motor area, and the right temporal lobe (Chabin et al., 2020). The orbitofrontal cortex is involved in processing emotion, and the supplementary motor area is involved in movement control. This study also found that when highly pleasurable music was played, dopamine was released in the dorsal striatum before participants actually experienced chills, and throughout the peak pleasure phase they recorded a dopaminergic release in the ventral striatum (Salimpoor et al., 2011). Dopamine is then a neurotransmitter associated with feeling pleasure and satisfaction.

The effects of music on Cognitive Function

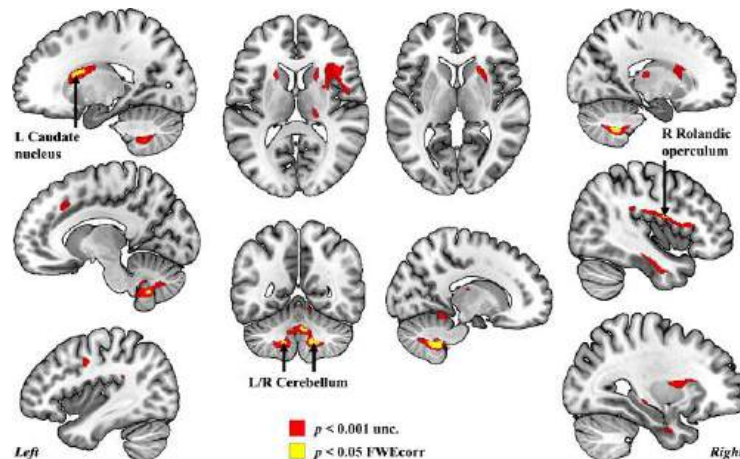


Figure 4. Univ. of Geneva Experiment - regions that display positive gray matter changes after musical training (Marie et al., 2023)

Expertise and experience with music such as playing an instrument, producing music, and composing music has been linked with an increase in cognitive brain functioning, in the fields such as language, attention span, memory and even mathematics. A study conducted at the University of Geneva collected over 100 retired people and studied the differences in their brains using fMRI scanning before vs. after taking lessons in piano and musical awareness (Marie et al., 2023). After six months of consistent lessons, they noticed an increase in gray matter in multiple brain regions, including the cerebellum and other parts of the neocortex linked to working memory and directed attention (see Figure 4 for a detailed map of these regions).

Musical training did not rejuvenate any areas of the brain, but increased the brain's plasticity and slowed the cognitive declining process. They also noticed that inevitable atrophy within the brain was not associated with behavioral decline. A study by George & Coch (2011) found that musicians, as opposed to non-musicians, had shorter latencies of electrical brain responses to visual and auditory stimuli, as well as a better awareness of tonal changes. A study conducted by (Halwani et al., 2011) focused on the arcuate fasciculus, a white matter tract connecting frontal and temporal brain regions, and the differences within its development in musicians vs. non musicians. They found that musicians with long term motor-vocal training had an increase in volume and microstructural complexity of the arcuate fasciculus as opposed to non-musicians. The arcuate fasciculus and surrounding regions of the brain are strongly linked with speech processing, concluding there is a positive correlation between experienced musicians and processing of language and speech.

Conclusion

In this article, we first reviewed how the brain processes sound through the ears, then reviewed the neurology of music in relation to beat, pitch and tone processing. We summarized the brain regions that are involved in processing these musical elements. We later reviewed how personality and the big 5 personality traits correlate with certain preferences of music, and highlighted that Openness was linked to more physical sensations when listening to music, and discussed the neurobiology and plasticity behind frisson. We also explored research studies that showed that music was linked with a positive change in cognitive function, and an increase of gray matter and plasticity. Throughout this paper we compared how musicians and those with musical experience have different neurological functions than those who do not play music. This paper aimed to further explore the relationship of neuroscience and music, and how music affects us psychologically.

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Are There People Starving In A Rich Community? A Case Study of the Silent Challenges in the North Shore By Samuel Chen

Abstract:

The North Shore in Illinois is also known for being a wealthy community so many people assume that there are no poor people in the area. However, food insecurity remains a critical issue in the North Shore, where the pandemic caused rising living costs and economic disparities and led to increased reliance on assistance programs. This research examines the effectiveness of food insecurity assistance programs, such as food pantries, in this region, using ethnographic observations from a local food pantry and qualitative interviews with volunteers and clients in multiple pantries. The findings reveal that while these programs provide crucial support, gaps in accessibility and lack of cultural foods and information persist, particularly among vulnerable populations. This study underscores the need for targeted improvements in food assistance strategies to ensure more equitable outcomes in the North Shore community. This research also addresses the difficulties that many other clients in other areas face since similar issues also affect people beyond the North Shore as well.

Introduction

One particularly hot summer, a single mother and her children arrived at the food pantry just as the doors were opening. Her children peeked inside with hopeful eyes. The pantry was stocked with donated goods, thanks to the generosity of local residents and businesses. As the mother selected items—canned beans, rice, and a few fresh vegetables—she noticed the relief in her children’s faces. For a moment, the weight of financial stress was lifted, replaced by the comfort of knowing that there would be enough food for the week.

In the North Shore, the area was known for its wealthy neighborhoods. But behind the façade lay a pressing issue: food insecurity was affecting more families than anyone had anticipated. Every day, many families would walk into food pantries looking for food that would sustain them for the next few weeks. With their modest income, they often had to choose between paying bills or buying groceries.

This scene was not unique to this particular family. Many families in the North Shore faced similar struggles, hidden behind the wealthy neighborhoods. Food insecurity was a silent challenge, affecting the daily lives of many, despite the region’s affluence. The food pantry’s efforts were a lifeline, but they also highlighted a larger issue: the need for sustainable solutions to address food insecurity in an area where the disparities between prosperity and poverty were stark and often overlooked.

This research focuses on four key food pantries: New Trier Township Food Pantry, Wilmette Food Pantry, Willow Creek Care Center, and Northfield Township Food Pantry. Each pantry serves distinct client demographics and operates under different structures and funding models, yet all share a common goal of alleviating hunger and providing essential services to their communities. For example, the New Trier Township Food Pantry and the Wilmette Food

Pantry are pretty small while the Willow Creek Care Center and the Northfield Township Food Pantry are massive. The differences in pantry size makes it harder for the smaller pantries to serve the same amount of people as the bigger ones. In the following sections, this study will dive into the state of food insecurity in the region, what pantries have been doing, and what they can do better to serve their clients.

Literature Review

Food insecurity is not a unique problem in the North Shore but a pressing issue nationwide. Many scholars have seen the issue and examined the effectiveness of government assistance programs and food pantries along with how food insecurity is a growing concern within the U.S. According to Alaimo, food insecurity is “the limited or uncertain availability of nutritionally adequate and safe foods, or limited, or uncertain ability to acquire acceptable foods in socially acceptable ways” (Alaimo 282). Food insecurity has risen over the past few years, especially in adults. Between the years 2021 and 2022, the percentage of people in the U.S with food insecurity rose from 10.2%, 13.5 million people, to 12.8%, 17 million people (USDA). There are many factors that impact how much food insecurity increases, including race, place of residence, household composition and geographic location. For race, Blacks and Hispanics’ food insecurity rates increased the most, at 2.6% and 4.6% respectively (USDA). Not only that, the place of residence is also important, with the highest increase outside metropolitan cities at 3.9% and inside the principal cities with an increase of 3.1% (USDA). The geographical area is also important when it comes to food insecurity increase, with the South having the highest increase at 3.1% and the Northeast at 2.8% (USDA). Children are also being affected by the increase of food insecurity and they are also struggling to put food onto the table. 12 million children don’t have the comfort that there will be food on the table (“Hunger in the U.S”). More than 1 in 5 children are at risk of food insecurity, with 1 in 3 children for Latinos and African Americans (“Hunger in the U.S”).

Food insecurity is also very important because it is linked to many health issues, such as diabetes, Chronic Kidney Disease, obesity and cardiac issues. 6.8 million U.S households suffer from very low food insecurity, which is defined as when the “normal eating patterns of one or more household members were disrupted and food intake was reduced at times during the year because they had insufficient money or other resources for food” (USDA). This is significantly higher than the 5.1 million people in 2021 (USDA). A survey that was conducted in two states interviewed benefactors from SNAP and charitable food pantries reported that 60% of households identified health conditions as the main challenge that comes with food insecurity (Jacknowitz et al.). One of the main reasons is because of the lack of nutrition that people get from cheap food (Diabetes and Food Insecurity). Cheap food is not that healthy for the body because it has high amounts of sugar, saturated fat and sodium (Diabetes and Food Insecurity). Not only that, they are also high in calories and carbs which can cause spikes in blood sugar levels (Diabetes and Food Insecurity). High blood sugar levels increase the risk of diabetes complications like heart and kidney diseases, vision loss or lower-limb amputations (Diabetes and

Food Insecurity). Food insecurity can also cause people to skip meals, which can lead to lower blood sugar levels and that can cause diabetes complications (Diabetes and Food Insecurity). Health care costs can also be difficult for people who are already struggling with their budget (Diabetes and Food Insecurity). As a result, these people have to choose between health care or food. This can cause people to put off filing their prescriptions and checking their blood sugar level in order to save supplies (Diabetes and Food Insecurity). This can be very dangerous and lead to further health issues (Diabetes and Food Insecurity).

According to Feeding America, around 49 million people (1 in 6 Americans) received some sort of charitable food assistance in 2022 (Charitable Food Assistance Participation). Various assistance programs, such as food pantries, community kitchens, and government subsidies like SNAP are used by people in need. However, SNAP has been underperforming in many counties because the average modest meal in the U.S costs \$3.14, but the SNAP benefits only cover \$2.74 (Gupta and Waxman). Considering that 4 out of 10 SNAP users make zero net income, rising food costs and inadequate benefits make it hard for households to feed themselves (Gupta & Waxman). According to data from NielsenIQ on geographic variations in food prices, benefits were inadequate in one out of five 5 counties (Gupta & Waxman). SNAP benefits are particularly inadequate in urban areas with high food prices and in some rural areas that face above average food prices (Gupta & Waxman). Among adults with very low food insecurity, 51.3% drew on savings to pay for food and 1 in 10 used cash from loans to pay for groceries (Gonzalez & Martinchek). Continually using payday loans will undermine a family's financial stability and increase debt burdens to unsustainable levels (Gonzalez & Martinchek). SNAP is not sufficient enough for people with very low food insecurity, which is why they have to rely on savings and loans.

There have been many studies on food insecurity in the U.S and mostly focus on how widespread food insecurity is and how well the government assistance programs are at dealing with it. Some findings that all the sources commonly have is that government assistance programs are not doing enough (Chilton and Rose; Gonzalez and Martinchek; Gupta and Waxman; Jacknowitz et al.). The studies urged the government to increase the funding for government assistance programs as they are very important to those who cannot afford food comfortably (Chilton & Rose; Gonzalez & Martinchek; Gupta & Waxman; Jacknowitz et al.). While other studies are focusing on the increase in food insecurity ((Alaimo 282; CDC Surveillance System: Trends in Prevalence of Food Insecurity in U.S. Adults; USDA). There also have been sources that have mentioned that government assistance programs like SNAP aren't enough for families who have little to no income(Gonzalez & Martinchek). However, there are some limits to the studies, as some of them only have data from surveys that are merely part of a few states (Policy and Benefits). Some of the studies are also outdated as they were before the pandemic and a lot of policies changed because of it (Alaimo 282; Chilton & Rose; David; Myers & Painter; Zepeda). This could have influenced the study findings and made the study different. There are also other studies that try to cover nationwide data, which means their analysis may miss the grounded interactions and real world mechanisms (CDC Surveillance

System: Trends in Prevalence of Food Insecurity in U.S. Adults; Charitable Food Assistance Program; "Hunger in the U.S"; Urban). Many papers do not focus on wealthy communities because they assume that everyone in such communities does not confront issues related to food insecurity (CDC Surveillance System: Trends in Prevalence of Food Insecurity in U.S. Adults; Charitable Food Assistance Program; "Hunger in the U.S"; Urban).

This research will contribute to this topic because it focuses on the micro-level analysis of how communities deal with food insecurity and it demonstrates how food insecurity is still prevalent in wealthy communities. To reach these ends, data from a small community in a wealthy neighborhood is used. My paper also has a lot of data from the client side and pantry coordinators, which means it will have both sides of the narratives and behaviors. This makes my paper more reliable as I have information from the giver and the receiver in the food pantry operation. I got the information by utilizing ethnographic observations and interviews. These methods are in-depth and provide context for the study.

Methods

This study employs a mixed-method approach to evaluate the effectiveness of food insecurity assistance programs in the North Shore. The research combines two qualitative methods, interviews and ethnographic observations, to gain a comprehensive understanding of how these programs function and their impact on food insecurity in the region.

The research was conducted in the North Shore, a region characterized by its wealthiness. My fieldwork focused on food pantries across various communities within the North Shore region. Participants in this study included program administrators, volunteers, and clients who utilize these food assistance programs. I ensured that there was representation from different types of programs and demographic groups. The final sample included 4 program administrators, 2 volunteers, and 13 clients. Most of the samples were female, with the volunteers being male. During my ethnography, I noticed that most of the volunteers were middle aged or elderly and they were mostly women. However, the race of the sample was mostly white while the clients were a diverse group. Even though the North Shore is predominantly known for being mostly white, there were still a decent amount of African Americans, Latinos and Asians.

The administrators that I interviewed, mostly women, were very kind and patient. The interview invitations were sent out through email to the pantry administrators. In the email, I also attached an interview outline so that the interviewees were able to see what we were going to talk about in advance. These interviews were conducted with program administrators and sometimes clients to understand the operational structure, challenges, and perceived effectiveness of the programs. While I was volunteering at the Willow Creek Care Center, I connected with clients because I talked with them while I was putting their information into the system. After I put their information in, I asked them if they were willing to be interviewed for my paper. Each interview lasted between 30 to 60 minutes and was recorded with participants' consent. My participant observation took place at the Northfield Township Food Pantry. I participated in volunteer activities, such as food distribution and meal preparation, and observed

what was going around in that space at the same time. The total observations amounted to 20 hours. This method provided insights into the everyday operations of the pantry and interactions between volunteers and clients that cannot be fully conveyed in the interviews.

I made sure that for each of these interviews, informed oral consent was obtained from all participants before their involvement in the study. The consent process included a clear explanation of the study's purpose, procedures, potential risks, and benefits. I also had the interviewees sign a consent form that outlined all the permissions and what I was going to do with the interview.

By integrating interviews and ethnographic data, the study provides a better understanding of these programs' strengths and areas for improvement.

Operational Structure and Pantry Services

The food pantries on the North Shore serve their clients by adapting their services to meet the needs of diverse populations and relying heavily on donations, though their operational structures vary. The New Trier Township Food Pantry is affiliated with New Trier Township and accountable to taxpayers through the township supervisor and board. It also serves clients who meet income guidelines and issues cards for their bi-weekly visits. During the pandemic, the pantry adapted its services to ensure safety by offering outside pickups before recently allowing clients to shop inside. Similarly, the Wilmette Food Pantry is managed by 8 to 10 dedicated volunteers, who handle all aspects of food distribution and organization. While the pantry caters to a broad and diverse group with minimal entry requirements, it also responds to increased demand with specific services like holiday food items. In contrast, the Willow Creek Care Center manages a high volume of clients with structured visit limitations and provides a comprehensive range of services beyond food, including clothing and clinics. They relied on a mix of government funding (60%), donations (20%), and church support (20%). The Northfield Township Food Pantry, a 501(c)(3) charity, does not check income levels but verifies residency. It sources food from the Greater Chicago Food Depository (GCFD), local supermarkets, and community donations. With about 50 regular volunteers and 150 additional ones who help occasionally, the pantry handles various tasks, including client interaction, stocking, cleaning, and picking up donations. Collaborating with other local pantries and schools for food drives and donations, the Northfield Township Food Pantry has strong partnerships with local companies and organizations that support the pantry through food drives and volunteering.

However, most of these pantries suffer from either the lack of government aid or the decreased government aid. The New Trier township food pantry operates on donations (85 to 90%) and a modest \$25,000 budget in 2023. The pantry receives contributions from schools, churches, local businesses, and volunteer-organized food drives. Although the New Trier Township Food Pantry operates with the township, the pantry's heavy reliance on donations asks the question of whether or not New Trier Township is given enough funding. This relates to how the New Trier Township Pantry limits the amount of food that each client could choose. According to Brian Leverenz, a New Trier Township pantry staff:

People can't take more than 2 cereals, no more than 2 toiletries, 2 cans of food, 2 cookies, or 2 crackers. People can take 2 dairy items out of the fridge. We have milk, eggs, yogurt, and butter. People can take 2 meats out of the freezer, like chicken breasts, fish, pork chops, sausages.

This limit means that there will be some clients who will not be able to get all the food that they want and will have to make do with the food that they were able to get. Perhaps if the New Trier Township government had invested more funding into the pantry, it would be able to increase the amount of food that each client would be able to take out.

Similarly, Wilmette food pantry operates independently without government funding, relies solely on donations and is housed rent-free at Trinity Methodist Church. The pantry also only operates on Tuesdays, which gives clients a very limited time period to get food. The pantry's heavy reliance on donations raises questions about whether the government provides sufficient support. The Wilmette Food Pantry is a very small pantry, which is why the pantry only has 8 to 10 volunteers since they do not need any more. According to Barbara Young, a Wilmette Food Pantry staff:

The pantry is allowed to only give the people what they have. If the pantry doesn't have a certain item, the people are just going to have to wait.

Since the Wilmette Food Pantry solely operates on donations, perhaps the pantry would be able to give out more food and operate on more hours if they had government support.

Although the Willow Creek Care Center gets more than half of their food from the government, they have recently faced cuts in government aid. This forces the Willow Creek Care Center to depend more on the community donations. According to Esther Desantiago, a Willow Creek Care Center staff:

The COVID pandemic opened up many more pantries but everyone still gets the same amount of food from the government. This caused the government to lower what everyone was getting, which dropped Willow Creek's funds by 20% two years before and another 20% last year.

Since the Willow Creek Care Center is the biggest food pantry in the North Shore, a lot of people get impacted by this decrease. The decrease in government aid could be the reason why the Willow Creek Care Center limits clients to use the pantry once a month. This raises even more concern about the funding that the government is putting into local food pantries since the pandemic has caused the amount of clients to increase.

Clients at the Willow Creek Care Center have also reported that they use SNAP and go to the Willow Creek Care Center. According to 2 male clients, they both reported that they also

participate in SNAP and receive benefits from it. This shows how SNAP is not enough for their families since they also visit pantries to receive food.

Similarly, the Northfield Township Food Pantry is a pretty big food pantry as it serves 880 monthly families that live in the Northfield Township Area. However, according to Julie Schaefer, the coordinator for the Northfield Township Food Pantry:

“I don't have a hard number but I would say about 20% of everything we distribute is from the GCFD [the Greater Chicago Food Depository]. The rest of it is either things that we purchase or food items that are donated by community members.”

This shows how little the government is providing food for the pantry, even though they serve more than 800 families per month. The pantry provides many limits to each category of food because they don't want them to run out for clients. During my ethnography, I have seen many clients having to put back food because they went over the limit by 1 or 2 items. However, if the government provides more help to the pantry, they will be able to provide more food for clients and remove some limits on food so clients will be able to get their desired amount.

Client Diversity and Community Outreach

The food pantries in the North Shore also serve a variety of clients and they also try to get their existence known in their community while also making the pantry seem welcoming toward clients. The New Trier Township food pantry collaborates with over 40 local nonprofits, schools, and police departments to enhance visibility and support, maintaining consistent volunteer recruitment through community engagement efforts. Wilmette responds to heightened demand driven by rising food prices and economic instability, adapting its services to meet specific needs such as offering holiday-related food items and accommodating various dietary restrictions. The Northfield Township Food Pantry encounters similar challenges, serving over 1,200 families with 800 visiting monthly and four new families joining weekly, yet 20% of the township's population remains unaware of its existence. Volunteers have become proficient and now operate independently after initially struggling with the pantry's workflow. Volunteers will also speak slower for clients who have trouble hearing in order to accommodate their hearing condition. They will also do this for people who don't know English that well. Sometimes clients like to bring in a second person to translate between the volunteer and the client to bring down the language barrier. The volunteers at the Northfield Township Food Pantry still often talk to each other about their life outside the pantry and coordinate with each other. Willow Creek addresses language barriers with Spanish-speaking staff and operates with a dedicated team of nine staff members and up to 800 volunteers weekly from diverse backgrounds. Not only that, but they also have signs all around the pantry that are in Russian, Spanish and English so that clients can read the signs in their native language and access the pantry. The center gains visibility through its website, church gatherings, and word-of-mouth referrals among customers. Another way that pantries like to get more clients to visit their pantry is to make the pantry more welcoming

toward clients. Many clients are ashamed and embarrassed to be using the pantry so the Willow Creek Care Center makes itself more welcoming for clients by making the pantry seem more like a grocery store.

However, New Trier Township faces challenges in meeting increasing demand, particularly during economic downturns or periods of inflation. This is a reflection of the increasing food insecurity in our nation these days. A study found that 62% of adults whose grocery costs increased reported either reducing or not buying the food that they wanted (Martinchek et al). They also found that adults whose grocery costs increased were almost twice as likely to have food insecurity (29% compared to 16.5%), which shows the direct correlation between inflation and food insecurity (Martinchek et al). At the same time, many people are unaware of what support they can get from the community. For example, despite community outreach through flyers, business cards, and periodic media coverage, the Wilmette food pantry remains relatively unknown due to its small size. According to Brian Leverenz:

8 main reasons for people to visit the pantry: Disability and disease is the biggest. The most frequent disability is mental illness. Displacement/job loss is usually temporary. Divorce, death, desertion, drugs and alcohol, domestic violence. Some people fall in multiple categories... Many clients are kind of embarrassed or ashamed that they're using a food pantry.

There are many people who are being affected by these reasons, however, they do not visit the pantry because they either do not know about it or they are too ashamed to visit it. One way that pantries can improve their effectiveness in their community is to make the pantry more welcoming to people who are in these times of trouble.

The New Trier Township Food Pantry also has trouble getting a variety of foods onto the shelves since they mostly receive American food from donations and government aid. Similarly, the Willow Creek Care Center also has similar problems. According to Esther Desantiago:

The Care Center does have a goal of getting cultural foods for the families so that they get more comfortable food that they see at home, not just American food but also other types. They will get these mostly from donors because the government only gives out American foods. They do want the government to increase funding of the cultural foods because of migrants

The increase of migrants due to the Russia-Ukraine war and the cartel in Central and Southern America make this issue more pressing because they are not used to eating American food at home. According to the Migration Policy Institute, nearly 46.2 million immigrants lived in the US in 2022, which is the highest ever in US history (Batalova). Foreign population in the US also grew by 912,000 people between 2021 and 2022, which is 65% of the total population increase during that year (Batalova). According to a survey done on 33,000 immigrants by Kaiser Family Foundation, about 53% of them reported challenges accessing government assistance programs such as food insecurity programs due to the language barrier (KFF). Around

25% of the participants also reported that they feel overqualified for their job (KFF). This results in less income for their family since they are working a job that pays less. When observing the clients in the Willow Creek Care Center, I also found that there was a diverse population in the 22 clients that I observed: 36% English, 18% Russian or Ukrainian, 18% Venezuelan, 14% Spanish, 9% Mexican, and 4.5% Columbian. If they are able to get cultural food, it will make the pantry truly seem like a grocery store that they will find in their neighborhood, and it will be more effective than American food. This is why the government needs to add more variety to the foods that they give to food pantries because not everyone likes to eat American food.

Another issue that comes with the increase of immigrants is the language barrier between volunteers and clients. During my ethnography at the Northfield Township Food Pantry, I observed that some clients brought another person with them into the pantry. The person they brought was a translator who would interpret what the volunteer was saying and translate it for the client. This would help them understand the rules of the pantry and access it easier. However, not everyone has a relative that can speak both English and the client's native language very well. According to Esther Desantiago:

Language barriers are one of the challenges that the care center faces because they mostly serve migrants. The Care center doesn't have a single mandarin speaker and it's also hard for Ukrainian and Russian clients because the care center only has a few volunteers who can speak those languages. However, the care center has enough Spanish speakers because Desantiago was able to build a connection between the English and Spanish speakers.

Language barrier is a big issue in the North Shore as well because clients will not be able to get the full service of the pantry since they do not understand everything in the pantry. Pantries can increase the effectiveness of pantries for non-English speaking immigrants by getting volunteers who can speak multiple languages or start making guides that are in multiple languages. Non-english speaking clients can also bring in another person who can translate what the volunteer says to the client.

The age imbalance of volunteers is also remarkable. During my ethnography at the Northfield Township Food Pantry, there was a lack of volunteers from the younger generation. Most of the volunteers were middle aged or elderly men and women. According to Ron Kochman, a volunteer at the New Trier Township Food pantry:

I have been retired for 10 years and almost every pantry volunteer leaves after one year because of various reasons. I have also been living in the township since 1975: most friends and neighbors don't know the pantry exists.

The fact that Ron has been retired for 10 years brings up the question about volunteer availability. It makes a point that a volunteer needs to have enough spare time in order to

volunteer at the pantry. This also proves the volunteer demographics in the ethnography since almost all of them were middle aged or elderly men and women.

Ron also makes the point that many friends do not even know that the pantry exists. This shows how the community is fully using the pantry since many of them are unaware of its existence. Even though the Northfield Township food Pantry serves over 1,200 families with 800 visiting monthly and four new families joining weekly, yet 20% of the township's population remains unaware of its existence. Similarly, Wilmette food pantry also has a problem with the lack of awareness of people in the community. According to Barbara Young:

The [Wilmette] pantry does have a website, and they try to have articles written about them and have a banner outside of the church, but there are still a lot of people who miss out because they are a small food pantry.

This raises the question of how many people are missing out on the food pantry because they do not know there is one in their community. This shows that pantries have a lot more potential in helping the community since there are many people in need of food who are unaware of the pantry in their community. These findings all show that there is a lot more that pantries can do to help their community by making themselves more known and by adapting for the people who cannot speak English that well.

Table 1. Comparison of Pantries

Pantries	Features	Challenges
New Trier Township Food Pantry	<ul style="list-style-type: none"> ● Is affiliated with the New Trier Township Board ● Operates on 85-90% donations and a modest budget of \$25,000 ● Clients must meet income guidelines and are issued a card for bi-weekly visits. ● Clients come from affluent areas like Winnetka, with varying needs due to disabilities, job loss, or other crises. 	<ul style="list-style-type: none"> ● More government aid ● Increasing amounts of clients among economic downturns ● Diversity in the foods given by the government ● Lack of awareness in the community of the local pantry
Wilmette Food Pantry	<ul style="list-style-type: none"> ● Housed at Trinity Methodist Church, which provides space rent-free. ● No formal identification is required; the pantry operates on an honor system and serves anyone in need. 	<ul style="list-style-type: none"> ● Not enough government aid ● Lack of awareness in the community

Willow Creek Care Center	<ul style="list-style-type: none"> • Offers a comprehensive range of services including food, clothing, and clinics (dental and vision). • Clients are limited to one visit per month for grocery shopping, and the quantity they can take is based on available inventory. • Operates with a team of 9 staff members and utilizes up to 800 volunteers weekly across various shifts. 	<ul style="list-style-type: none"> • Not enough government aid • Increasing amounts of clients • Diversity in the foods given by the gov • Language barriers in the pantry
Northfield Township Food Pantry	<ul style="list-style-type: none"> • The pantry serves approximately 1,200 families, with around 880 visiting monthly. • Works closely with local grocery stores like Whole Foods, Trader Joe's, and Costco, receiving daily deliveries of food donations. • Pantry is in high demand when the children are out of school. 	<ul style="list-style-type: none"> • Unable to expand the pantry • Clients are limited because of the increase • Clients cannot access full pantry due to barrier • Pantry needs volunteers to operate • Still 20% of the township don't know the pantry exists.

Overall, the findings in this section prove that both the government and society have a lot of work to do in order to improve the effectiveness of the pantries. These findings are related to the existing literature because they both support the fact that the government is not doing enough to help the food assistance programs. For example, studies say that SNAP is not doing enough to support people with food insecurities since they have to take out loans and the money provided in SNAP is not enough for a modest meal due to inflation (Gonzalez & Martinchek; Gupta & Waxman; Jacknowitz et al.). Many studies support the findings except these findings are more specific towards the North Shore in Illinois. The North Shore in Illinois is unique compared to the other places studied because the North Shore is known for being a wealthy community and it mostly is. So these findings are targeted toward the heavy minority that have food insecurity in the North Shore. Other studies focus on areas that have more food insecurity than the North Shore or national statistics (“CDC Surveillance System: Trends in Prevalence of Food Insecurity in U.S. Adults”; Feeding America; "Hunger in the U.S"; Urban). The government and society can take action and make life easier for people in need by reducing the stigma against them and by providing more job opportunities for them. Many people in need have experienced shame and embarrassment because of their financial state which makes their life a lot harder. The

government can start connecting pantries with job opportunities so that people can maintain a stable income and financial state. Everyone has a role to play when it comes to helping the more unfortunate people in the community.

Conclusion

Overall, pantries need more government support and they also need to adapt to the clients more, either through breaking the language barrier or giving them more diverse foods. Food pantries play an essential role in addressing food insecurity through tailored approaches. Understanding the dynamics of volunteer-client interactions, operational structures, and community outreach efforts is crucial for enhancing the effectiveness and sustainability of these vital community resources. The government should increase the funding for the food pantries so that they can expand their operations and increase the amount of food they give to each client. The government should also increase its funding for the government assistance programs that people nationwide use. Studies have found that government aid is not enough for SNAP recipients as they need to use their own money to buy modest meals (Gonzalez & Martinchek).

Future research should explore strategies for improving client satisfaction, volunteer engagement, and community awareness to further support these pantries' missions. They can also consider the question: "Are there any other ways that will better help people than giving food only?"

People can also help clients by connecting them to job opportunities so that they can find jobs and get a better income. With a better income, clients would be able to move on from the food pantry and be able to sustain themselves. Furthermore, clients would benefit greatly if they were educated on tips and tricks on how the economy and job opportunities work. People can also help clients by working on their houses and making sure that they have a safe place to live or by helping them find an affordable house so they can live comfortably.

On the other hand, people can start considering what social structures are generating food insecurity and poverty consistently. For example, the state of job opportunities forces people to take low-paying jobs and the income is not enough to sustain them and their family, which then gives them food insecurity. Another social structure is the healthcare and social safety nets. These government programs are sometimes discriminatory toward certain ethnic minorities and that causes the clients to spend more of their budget on those programs. This leads to less of the budget being allocated for food, which leads to the family being food insecure. Political instability and unsafe environments also force clients to move areas into a new environment which can be hard for them to adapt to. During these hardships, clients often need support from the food pantry since they are unable to manage the budget that well. Food insecurity is not an issue that is isolated from other social issues and structures. Instead of a program that only takes food insecurity into consideration alone, a comprehensive program that provides solutions to poverty is much preferred.

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Epigenetic Mechanisms: A Promising Frontier in Cancer Treatment By Sanjana Thodupunoori

Abstract

Epigenetics is the study of heritable chemical modifications that regulate gene expression and DNA transcription without directly modifying the DNA sequence. Two key epigenetic mechanisms are histone modifications and DNA methylation. Epigenetic changes influence many biological processes and diseases, including cancer. Cancer arises when genetic mutations in the DNA cause a cell to multiply rapidly. Malignant tumors can spread to other tissues, allowing cancer to affect many areas in the body. Cancer cells can have epigenetic alterations that allow them to evade current treatments. This review provides an overview of epigenetic mechanisms and discusses how targeting these mechanisms can lead to additional cancer therapies. Specifically, we explore the potential of epigenetic drugs to target the small percentage of cancer cells that resist conventional treatments due to their epigenetic states. By understanding how epigenetics contributes to cancer development and treatment resistance, researchers can develop more effective and targeted therapies.

Introduction

DNA Replication and Regulation

DNA, or deoxyribonucleic acid, is the genetic material that encodes for all life processes. The genetic blueprint of DNA exists in almost every cell of the body. It consists of a sequence of nucleotide bases—adenine, thymine, guanine, and cytosine—held together by hydrogen bonds to form a double-stranded helix. Different parts of the DNA nucleotide sequence encode different genes in the body. In order to contain all this genetic information, cells package double-stranded DNA as chromatin in the cell nucleus. Chromatin is further condensed into chromosomes, which are complex structures located inside the nucleus. Through a tightly-regulated mechanism, chromosomal DNA is wrapped around histone-containing proteins known as nucleosomes (Alberts et al.).

When a cell is ready to divide, the DNA must first be copied to ensure that genetic information is passed on to the newly divided cell. This process, known as DNA replication, is essential for cellular growth, development, and the replacement of damaged cells. As such, it is tightly regulated to maintain the integrity of the genetic code. When a cell is ready to replicate and divide, tightly packaged chromosomal DNA is “unwound” and its double-helix is separated to allow for DNA replication and transcription (T.M.).

Initially, the DNA unwinds so that the double stranded helix can be split into two separate strands by an enzyme called helicase. Then a short piece of RNA primer is attached to each one of the strands. DNA polymerase then matches each of the DNA nucleotide bases on the strand with a complementary base in the 5' to 3' direction on one strand, while the other strand is done in short segments called Okazaki fragments (Shen). Once both DNA strands go through this

process, the primers are replaced with DNA nucleotide bases and the DNA ligase enzyme joins the Okazaki fragments together. This process results in two identical DNA molecules with one copy of the original strand and one copy of the new strand in each (Kornberg).

Overview of cell cycle/replication

DNA replication is especially important in the context of cell replication. There are two primary types of cell division: mitosis and meiosis. While mitosis is how most cells in the body divide and replicate, meiosis occurs in germ cells such as sperm and egg cells (Potapova). Mitosis is the cell division process most implicated in cancer. In the cell division process, there are 3 main stages called the growth (interphase) stage, the mitosis stage, and the cytokinesis stage. The interphase stage is broken up into 3 parts called G1, S, and G2 phases. In the G1 phase, normal cell functions and cell growth occur. In the S phase, the DNA is replicated and produces two copies of each chromosome. During the G2 phase, the cell continues to grow and prepare for the next phase. The next phase is mitosis which includes prophase, metaphase, anaphase, and telophase where chromosomes condense, the microtubules are attached to each sister chromatid, the chromosomes move away from each other, and finally new nuclear envelopes are formed around the separated chromosomes. The last phase, called cytokinesis, is the division of the cell which forms two identical daughter cells (Cooper).

Epigenetics Role in Regulating DNA Transcription

While DNA provides the blueprint for many cell functions, epigenetics can affect how and when genes are expressed without affecting the DNA sequence. Specifically, epigenetic modifications are chemical changes to DNA or associated proteins that do not alter the underlying DNA sequence, but can influence gene activity (CDC). These modifications act as switches, turning genes on or off in response to various factors, including environmental cues and developmental signals. For example, in one study in preclinical rodent models, scientists were able to detect how maternal care affected gene expression and stress response through epigenetic changes (O'Donnell and Meaney).

In this process, there are two main mechanisms: histone modification and DNA methylation. In histone modification, acetyl and methyl groups are added onto the histone tails directly, acting as chemical tags. DNA methylation is a process by which a methyl group is added onto the cytosine-guanine residues on strands of DNA.

Mechanisms Contributing to Cancer

Cancer is caused by certain mutations that can lead to abnormal cell growth. These mutations can occur due to environmental factors like tobacco smoke, alcohol, obesity, pollution, certain infections, and sun exposure (Trichopoulos et al.). The mutations can also occur spontaneously due to random errors that happen during the DNA replication process.

Although there are DNA repair mechanisms, at times such mechanisms fail, causing cells to continue through the cell cycle unchecked, ultimately leading to more cell division. For

example, a gene called the p53 tumor suppressor gene plays a crucial role at the G1 checkpoint. Its function is to sense DNA damage. When it does, p53 halts the cell cycle allowing time for DNA repair to happen. However, when there is a certain mutation in the p53 tumor suppressor gene, it can cause the cell cycle to proceed through the G1 checkpoint without fixing the DNA damage, leading to the replication of the mutation (Torii et al.). Usually, the cell division process is well regulated and prevents cell division from occurring uncontrollably. However, cancer cells are able to extensively replicate and divide to an extent that is hard to be controlled (Williams and Stoeber).

Cancer hallmarks and their importance

There are six key hallmarks of cancer. These include: sustained proliferative signaling, evasion of growth suppressors, activation of invasion and metastasis, replicative immortality, angiogenesis, and resistance to cell fatality. These hallmarks are a set of characteristics that scientists use to define cancer cells to distinguish them from normal cells (Lazebnik). By identifying which characteristics a cell or tumor has relevant to these hallmarks, scientists can develop diagnostic tools and therapeutics that are more targeted to the condition. Also, having knowledge of the hallmarks can help scientists predict how the cancer might develop and progress, leading to better treatment (Lazebnik).

Cancer Diagnosis and Existing Treatments

The clinical course and underlying mechanisms of cancer can vary extensively from patient to patient, making it difficult to treat. The risk of cancer occurring can also increase with age due to changes in metabolic profile, weakening of the immune systems, compounding genetic mutations, and chronic inflammation (Pesheva and University). It is crucial to diagnose cancer at an early stage as once it undergoes metastasis, it makes it far more challenging to treat. Metastasis can occur microscopically and silently across various locations, leading to multiple treatment targets, the development of drug resistance, and evasion of the immune system (Gerstberger et al.). Some examples of methods used to diagnose cancer include biopsy, laboratory tests, imaging tests, and physical exams (Hamilton).

Many different types of treatment options for cancer also exist, including surgery, chemotherapy, radiotherapy, immunotherapy, hormone therapy, and gene therapy (Miller et al.). A key goal of cancer treatment is to remove the cancerous tumors while minimizing the effect of the treatment on healthy cells. However, targeting the cancer cells specifically while leaving the healthy cells unaffected is very challenging. For example, chemotherapy's goal is to target rapidly dividing cells. Although this does kill cancer cells, it also kills other healthy cells that rapidly divide such as the cells in the hair, bone marrow, and digestive tract (Nygren). The effect of cancer treatments on healthy cells can lead to many side effects. Some examples of side effects patients may experience from chemotherapy can include fatigue, mouth sores, skin irritation, hair loss, digestive issues, and nausea (Silveira et al.).

Literature Review

Tumor Suppressor and Proto-Oncogene Mutations in Cancer

Despite cells having identical DNA, cells can vary vastly based on how genes are expressed. When a cell gains certain mutations that lead to inappropriate over- or under-expression of key genes involved in regulating cell cycle, this can lead to the development of cancer. This could happen in a variety of ways such as through mutations in tumor suppressor genes or oncogenes (Figure 1).

Tumor suppressor genes function as the "brakes" of the cell cycle, ensuring that cells only divide when necessary. The proteins encoded by these genes act as checkpoints, halting the cell cycle to prevent abnormal cell division and promoting apoptosis in cells that do not meet specific criteria. Overall, tumor suppressor genes are involved in inhibiting cell growth and repairing damaged DNA, while making the cell undergo apoptosis if the damage is not repairable (Williams and Stoeber). However, with DNA methylation and histone modifications, these tumor suppressor genes can be silenced, causing the cell to evade growth control mechanisms. Therefore, the cell is able to go through the cell cycle process unchecked when there are inactivating mutations in tumor suppressors, potentially leading to the formation of a tumor.

Similarly, there are proto-oncogenes. Proto-oncogenes are genes that promote cell growth and division. When these genes are overexpressed, they can become oncogenic, meaning they can cause uncontrolled cell growth leading to cancer development. While the two main mechanisms of epigenetics most commonly affect tumor suppressor genes, they can also affect proto-oncogenes. Ultimately, the interplay between epigenetic mechanisms, tumor suppressors, and proto-oncogenes can disrupt cell division and growth, opening the way for cancer (Lee and Muller).

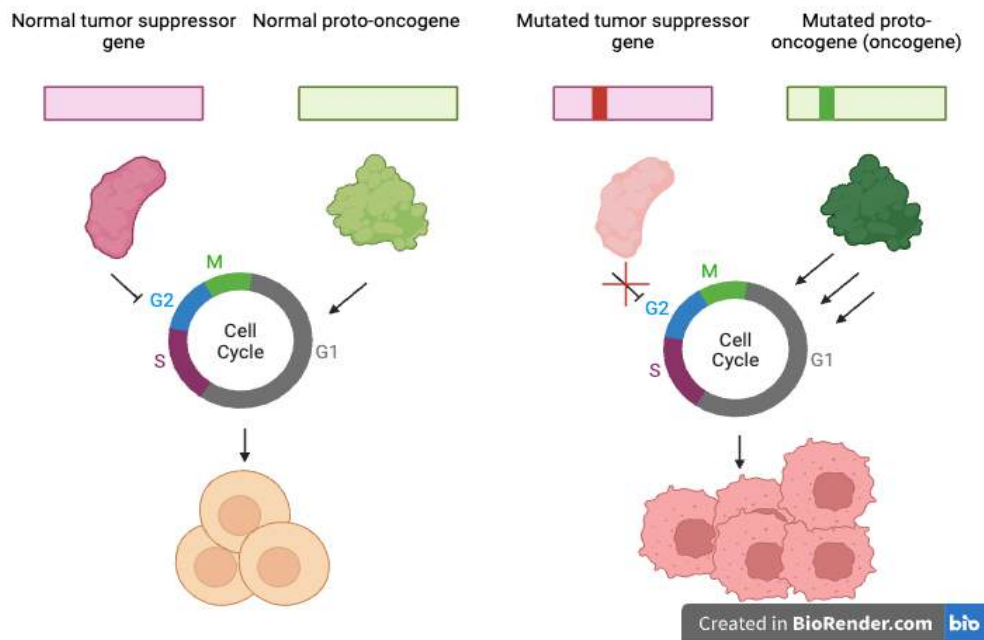


Figure 1. This figure is a diagram representing the difference in the function of a normal tumor suppressor gene and proto-oncogene compared to mutated versions of these genes. The mutated tumor suppressor gene as represented, stops the inhibiting mechanism of the cell cycle while the oncogene (mutated proto-oncogene) is overexpressed causing the cell cycle to continue.

Histone Modifications

Histones are the proteins that DNA is wrapped around in the chromatin structure. Histone modifications, as the name suggests, are the modifications that alter the structure of histones, which can affect DNA transcription and gene expression. Tiny chemical tags are added to or removed from the histone protein in this process. There are 4 main mechanisms of histone modification: acetylation, deacetylation, methylation, and demethylation. These mechanisms can affect gene expression based on how tightly the DNA is wrapped (Cheriyedath).

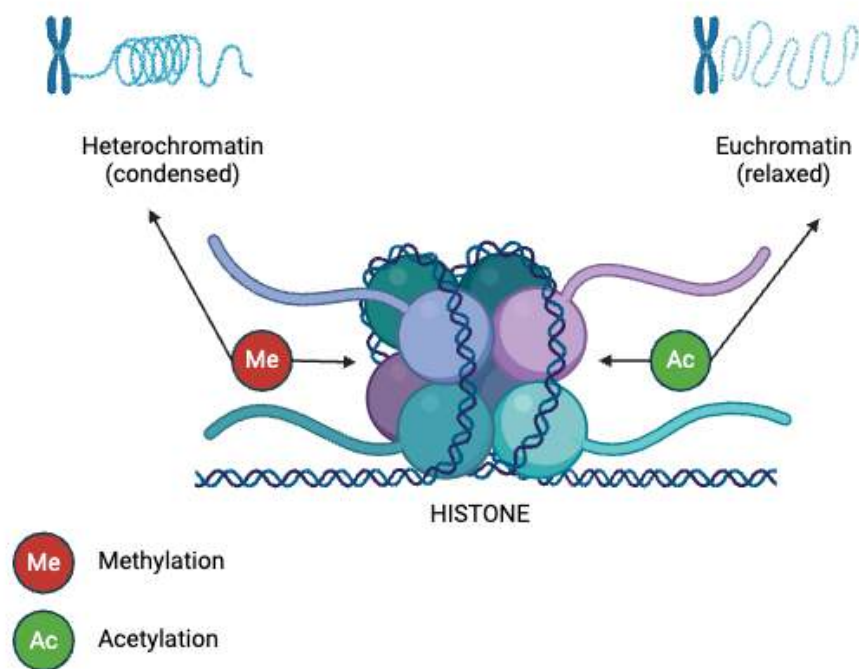
In histone acetylation, an acetyl group is added onto the Lysine or Arginine residues on the histone tail. The process of forming acetyl-lysine and acetyl-arginine is done by an enzyme called HAT, which stands for histone acetyltransferase. Since it is an enzyme, its purpose is to catalyze the transfer of the acetyl group onto the Lysine or Arginine residues on the histone proteins. The acetylation process is associated with the euchromatin state, also known as the active chromatin state. Essentially, this means that the chromatin structure is loosely packed, making it accessible to RNA polymerase during transcription (Figure 2). The genes in this loosely packed structure are more accessible and have the potential to be transcribed and expressed (Liebner et al.).

The reverse reaction, which is deacetylation, is instead triggered by the HDAC enzyme, also known as histone deacetylase. Deacetylation is the removal of the acetyl group from the

residues on the histone protein. Deacetylated histones are associated with the heterochromatin state, which is a tightly packed up state, limiting transcription in the unexposed areas. Since RNA polymerase cannot access the tightly wrapped up structure, it will not be transcribed (Barnes et al.).

Methylation and demethylation work in similar ways. While methylation is the addition of a methyl group onto the Lysine or Arginine residues, demethylation is the removal. However the heterochromatin state is associated with histone methylation where the HMT (histone methyltransferase) enzyme is responsible, while the euchromatin state is associated with demethylation where the LSD (lysine-specific demethylase) is responsible (Figure 2). Methylation can occur in several ways. It includes monomethylation, dimethylation, and trimethylation, which are names that indicate the amount of times the methyl group is engaged with the particular residue (Deng et al.).

Histone acetylation, deacetylation, methylation, and demethylation are all epigenetic mechanisms which can contribute to cancer development. When the structure is packed tightly, it can lead to the silencing of certain tumor suppressor genes. When the structure is loosened, it can cause oncogenes to activate. These mechanisms can both lead to genomic instability making them strongly linked to cancer.



Created in BioRender.com

Figure 2. This is an image representing how methylation and acetylation work as chemical tags on the histone protein to either condense or relax the chromatin for transcription needs.

DNA Methylation

DNA methylation is another epigenetic process in which a methyl group is added to the DNA molecule by a group of enzymes called DNMTs (DNA methyltransferases). While this methyl group being added doesn't alter the DNA sequence itself, it can alter the activity of segments. There are two types of DNA methylation, *de novo* methylation and maintenance methylation, and there are 3 enzymes, called DNMT3A, DNMT3B, and DNMT1 (Feinberg and Vogelstein). *De novo* methylation involves adding methyl groups to unmethylated sequences. This is usually done by DNMT3A and DNMT3B enzymes in early embryonic cells. DNMT1 maintains DNA methylation in the maintenance methylation process. The methyl groups that are attached to the sequence are usually added at cytosine-guanine sites, also called CpG sites. The methylation occurring at these promoter CpG islands affects DNA transcription as the methylated CpG islands can prevent transcription regulators from binding to the gene, hindering transcription.

There are also TET enzymes that, instead of adding methyl groups, remove them from the DNA. They help restore the original state of the DNA, allowing for accurate gene expression by converting the 5-methylcytosine (methylated cytosine) to cytosine (Moore et al.). This process is essentially called DNA demethylation. After demethylation, transcription factors have the ability to bind to genes and the genes can be expressed.

In cancer however, DNA methylation patterns may be disrupted either causing hypermethylation or hypomethylation. Hypermethylation refers to excess methylation at CpG sites which can cause gene silencing, and hypomethylation is decreased methylation at the CpG sites which can cause increased gene expression (Singal and Ginder). While DNA methylation is crucial for normal gene regulation, its disruption in cancer can lead to uncontrolled cell growth by altering gene expression patterns.

Leveraging Epigenetic Drugs Against Cancer

Primary mechanisms of epigenetic cancer therapies include inhibiting the enzymes or mechanisms that add or remove chemical tags to stop the overexpression or silencing of certain genes. Some examples of epigenetic cancer therapies that already exist are HAT inhibitors, HDAC inhibitors, Histone methylation inhibitors, and DNA methylation inhibitors. For example, HAT inhibitors called Garcinol and Anacardic Acid downregulate global gene expression and induce apoptosis (Kopytko et al.). HDAC inhibitors such as Vorinostat (SAHA) induce hyperacetylation of histones and non-histone proteins, promoting apoptosis and sensitizing tumors to other treatments (Li and Seto). Histone methylation inhibitors like DZNep inhibit the trimethylation of certain histone residues, causing the reactivation of silenced genes (Miranda et al.). Finally, DNA methylation inhibitors such as 5-Azacytidine (Vidaza) can cause degradation of DNMT1 and result in reduced methylation (Palii et al.) (Table 1). By targeting these epigenetic mechanisms and enzymes, the drugs can restore normal gene expression and stop the growth of cancer cells.

There also exist several other epigenetic therapies, such as HAT inhibitors. Curcumin

inhibits p300/CBP HAT activity, leading to cell cycle arrest and apoptosis (Marcu et al.). C646, a p300 inhibitor, replicates the proapoptotic effects of RNA-mediated p300 knockdown (van den Bosch et al.). Some HDAC inhibitors like Entinostat are used with other drugs for enhanced demethylation (Connolly et al.). ACY-1215 targets HDAC6, which has been shown to work in combination with other treatments for certain conditions (Wen et al.). Another example of a histone methylation inhibitor is Tazemetostat which targets the EZH2 enzyme, preventing histone protein methylation (Li and Seto). Finally, other examples of DNA methylation inhibitors include Zebularine which inhibits DNMT1, which have been shown to be effective in mouse models and can be taken orally (Yoo et al.). RG108 is a small molecule that inhibits the active site of DNMT1, reducing toxicity and demethylating certain genes (Zheng et al.). Overall, there exist many epigenetic drugs against cancer that target the mechanisms and enzymes responsible for promoting cancer.

Table 1. Overview of the epigenetic drugs for cancer based on mechanism

HAT inhibitors	<p>Garcinol + Anacardic Acid → downregulate global gene expression and induce apoptosis</p> <p>Curcimin → inhibits p300/CBP HAT activity, leading to cell cycle arrest and apoptosis</p> <p>C646 → a p300 inhibitor that replicates the proapoptotic effects of RNA-mediated p300 knockdown</p>
HDAC Inhibitors	<p>Vorinostat (SAHA) → induces hyperacetylation of histones and non-histone proteins, promoting apoptosis and sensitizing tumors to other treatments</p> <p>ACY-1215 → targets HDAC6, and has been shown to work in combination with other treatments for certain conditions</p>
Histone Methylation Inhibitors	<p>DZNep → inhibits the trimethylation of certain histone residues, causing the reactivation of silenced genes</p> <p>Tazemetostat → targets the EZH2 enzyme, preventing histone protein methylation</p>
DNA Methylation Inhibitors	<p>5-Azacytidine (Vidaza) → causes degradation of DNMT1, resulting in reduced methylation</p> <p>Zebularine → inhibits DNMT1, showing efficacy in mouse models and can be taken orally</p> <p>RG108 → a small molecule that inhibits the active site of DNMT1, reducing toxicity and demethylating certain genes</p>

Conclusion

This review overviewed the role of epigenetic mechanisms in cancer and the potential gap in cancer treatment that can be addressed with epigenetic drugs. By targeting the small percentage of cancer cells that have epigenetic states allowing them to resist cancer therapeutics, we are one step closer to developing more effective cancer treatments for resistant tumors. While there already exist some epigenetic therapeutics for cancer, in the future, the development of additional drugs and combining these drugs with other treatments such as immunotherapy or chemotherapy may enhance how efficient they are. Therefore, a potential area for future research is to conduct clinical trials where epigenetic drugs are used with existing cancer treatments, to make them more targeted to cancer cells. Additionally, understanding the epigenome in each individual can help scientists build personalized medicine to expand the possibilities of treatment. Epigenetic drugs combined with existing cancer treatments, may improve the effectiveness of therapeutics in treatment-resistant tumors.

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Next Generation Food Packaging By Ariel Sakhai

Abstract

In the modern world, plastic plays a crucial role in food preservation due to its versatility, durability, and affordability. However, its environmental drawbacks, including non-renewability and prolonged degradation, have intensified the search for sustainable alternatives. This review examines the latest advancements in alternative food packaging materials, with a focus on essential oils, animal-based coatings, algae-based coatings, and plant-based coatings. Essential oils like thyme, clove, and cinnamon offer antimicrobial and insect-repellent properties, enhancing food safety and extending shelf life. Animal-based coatings, including beeswax and chitosan, provide superior hydrophobicity and antibacterial effectiveness. Algae-based coatings exhibit excellent tensile properties and thermal stability, making them viable substitutes for traditional plastics. Plant-based coatings, treated with TiO₂ nanoparticles, enhance mechanical and barrier properties, preserving food quality for longer periods. We provide a novel cross study analysis to compare these components across four different parameters to rank the best materials for engineering next generation food packaging. Through our studies, we determined Chitosan is the superior material, ranking high across all four parameters: material flexibility, heat stability, anti-microbial properties and environmental safety. Despite the promising developments, challenges such as scalability, cost, and long-term stability need further research. The continued exploration of these innovative materials is essential for addressing environmental concerns and meeting consumer demand for eco-friendly packaging solutions.

Introduction

In modern society, plastic—a polymer-based synthetic material—dominates food preservation methods. Plastic's versatility, durability, and low cost make it the preferred choice for packaging and storing food items, helping to extend shelf life and reduce food waste. Complex food packaging did not become commonplace until the 19th century. Before this period, people stored food in natural materials such as shells, leaves, and gourds, often weaving these materials into containers (Berger). The need for dedicated food packaging arose with the transition from nomadic to more permanent living arrangements. Evidence dating back to 7000-1500 B.C. indicates that glass and pottery were used for food storage, demonstrating early human ingenuity in food preservation (Berger). Plastic packaging entered the food industry in the early 1900s during World Wars I and II, revolutionizing food preservation and distribution (Brody et al.). The mid to late 20th century saw the emergence of active packaging, with companies like Tetra Pak Co. and Dean Foods leading the way. These innovations included packaging that contained food and interacted with it to prolong shelf life. A notable example is silica gel, developed in the 20th century, which extends the shelf life of products by absorbing humidity. Such advancements marked significant milestones in the evolution of food packaging.

However, the environmental impact of plastic has sparked a significant debate and led to increased interest in alternative, more sustainable materials. In 2015, it was estimated that 164

million tons of plastic were consumed in the food packaging industry (Rhodes). However, plastic is neither renewable nor easily recyclable and takes an extraordinarily long time to decompose. Additionally, the production of plastic releases significant amounts of methane and CO₂, exacerbating global warming. These environmental concerns have intensified the need for more sustainable alternatives (Risch).

The market demand for alternative and active packaging is growing rapidly. Environmental awareness and consumer demand for sustainability have driven companies to explore and adopt eco-friendly packaging solutions. By 2028, the market for plastic packaging alternatives is projected to reach \$28.94 billion (Ma). This growth is fueled by both regulatory pressures and corporate commitments to reduce environmental impact. Companies are increasingly turning to biodegradable materials, edible films, and other innovative solutions that offer both functionality and sustainability. As more companies commit to eco-friendly and sustainable practices, they increasingly turn to these innovative packaging solutions, heralding a shift towards more sustainable food preservation. This transition not only addresses environmental concerns but also meets the evolving expectations of consumers who prioritize sustainability. The continued development and adoption of sustainable food packaging represent a critical step in mitigating the environmental impact of plastic waste and fostering a more sustainable future.

In recent years, many groups have heavily researched more sustainable food packaging methods, highlighting their potential benefits. The studies have evaluated these novel materials for numerous food safety parameters. However, no study to date has yet conducted a thorough analysis to effectively compare these different materials and determine which material is superior for alternative food packaging while preserving essential food safety properties. In this review, we will perform a comparative analysis between cutting edge plastic alternatives used in novel food packaging. We will discuss the latest innovations in novel biopolymer production technologies that address extending shelf life to prevent pathogenic bacterial growth, improving flavor during this preservation and maintaining environmental sustainability. We will finally assess each of these metrics across many different animal and plant-derived compounds.

Essential Oils

Essential oils are made of a mix of organic compounds which are derived from different parts of the plant (Wani et al.). In recent years, essential oils have become a promising alternative form of food packaging due to their wide range of applications, from insect repellent to antibacterial qualities (Wani et al.). Recent studies have demonstrated the strong anti-pest properties found in essential oils such as Thyme oil, which is extracted from the thyme plant. Thyme oil microcapsules have smooth surfaces, good encapsulation properties, and excellent thermal stability, making them a great potential alternative (Chung). For four weeks, these microcapsules exhibited over 90% insect repellency, demonstrating their potential for usefulness in pest control (Chung). Additionally, Thyme essential oil successfully loaded and stabilized with Halloysite nanotubes (HNTs) are encapsulated using vacuum pulling and surface coating

techniques (Jang et al.). These HNT nanocapsules provide prolonged antibacterial activity, preventing *Escherichia coli* growth for up to 10 days when incorporated with flexographic ink for food packaging (Jang et al.). Its antibacterial and Insect repellent properties highlight the versatility that essential oils, such as Thyme oil, can and have played in the active packaging world. Essential Oils have also been shown to help improve the structure and brittleness of packaging materials. To increase the brittleness and rheological properties of PLA, essential oils including clove, garlic, and cinnamon are combined with polyethylene glycol (PEG) (Ahmed). These films show promise as sustainable substitutes for traditional packaging materials in improving food safety and prolonging shelf life by inhibiting microorganisms such as *Staphylococcus aureus* and *Campylobacter jejuni* (Ahmed). In addition, zein-based films that have been modified with essential oils, eugenol, and monolaurin show improved surface contacts, thermal stability, and structural integrity (Jana Sedlarikova). These films also inhibit a variety of microorganisms, such as molds, yeasts, and bacteria (Jana Sedlarikova). Essential Oils have also been proven to increase the shelf life of perishable goods, such as Sea Bass. Sea bass filets' shelf life is greatly increased by chitosan nanofibers infused with a mixture of liquid smoke and thymol (Ceylan et al.). When comparing the control test to the test that contained the essential oils, the shelf life of the filets lasted longer (Ceylan et al.). Given that these nanofibers can tolerate temperatures of up to 150 °C, pasteurization and reusability are possible uses for them (Ceylan et al.). This robust body of research demonstrates how alternative food packaging can improve food safety, lengthen shelf life, and offer sustainable food packaging options. Microencapsulation of essential oils would also help preserve the ability of the essential oils, consequently increasing the shelf life of the packaged goods. The process of microencapsulation, which involves coating the essential oils in an emulsion made up of organic materials such as lipids, proteins, and polysaccharides, provides them with a coating that helps protect and preserve them for longer. With microencapsulation, the shelf life, durability, effectiveness, and overall performance are advanced (Reis et al.).

Animal-based Coatings

Animal-based coatings can vary from beeswax which is made from bees to chitosan which is made up of shellfish. As the animals and types of products vary, so do their physical properties such as their durability, antibacterial effectiveness, and brittleness. For example, the hydrophobic properties of beeswax and shellac wax were measured as sustainable alternatives to plastic petroleum-based coatings for paper packaging. The study examined the thermal, chemical, surface, and barrier properties of these alternatives. Shellac wax, due to its longer fatty acid chain, demonstrates higher water contact angles than beeswax, thus providing superior hydrophobicity (Ahuja and Kumar Rastogi). Among various formulations tested, the BS-2-60 coating is identified as the most promising, showing significant resistance to penetration by food and liquids (Ahuja and Kumar Rastogi). Similarly, research into the nanoencapsulation of liquid smoke and thymol in chitosan nanofibers highlights their efficacy in extending the shelf life of sea bass filets by inhibiting the growth of total mesophilic aerobic bacteria (TMAB),

psychrophilic bacteria (TPB), and yeast and mold (Ceylan et al.). Furthermore, looking at the effectiveness of antimicrobial chitosan coatings enriched with essential oils and bioactive compounds on fresh-cut broccoli reveals effective inhibition of *E. coli* and *Listeria monocytogenes*, with notable antimicrobial activity from tea tree, rosemary, pollen, and propolis, while pomegranate and resveratrol exhibit less effectiveness (Alvarez et al.). These coatings prevent the growth of bacteria without compromising the sensory attributes of the broccoli like its taste, texture, or smell. Additionally, research on chitosan-coated paper for oil barrier applications explores chitosan's interactions with fatty acids, particularly in acidic conditions, where the polycationic nature of chitosan stabilizes fatty acid emulsions by binding with anionic lipid molecules (Ham-Pichavant et al.). The study notes that chitosan-coated paper exhibits effective fat barrier properties, although high treatment costs are mitigated by incorporating natural molecules such as cellulose ethers and alginates into the chitosan coating formulations (Ham-Pichavant et al.). These studies underscore the potential of natural and sustainable materials to enhance food safety, extend shelf life, and provide innovative solutions in food packaging, positioning these materials as viable replacements for traditional, environmentally detrimental packaging options.

Algae-based Coatings

Algae has also proven to be a promising alternative in the active and alternative food packaging industry. Coming from the ocean, these plant-based materials are often in abundance. Some researchers are finding innovative ways to use marine algae for sustainable solutions in packaging and marine coatings. Testing semi-refined kappa-carrageenan, derived from algae, for edible packaging, they found that when plasticized with sorbitol, the films had excellent moisture content, water vapor permeability, and oxygen barrier properties, making them ideal for eco-friendly packaging (Farhan and Hani). These coatings, created by heating an azide and an alkyne together, proved to be more stable and resistant to fouling than physically cross-linked versions, offering durable, eco-friendly options for marine use, which can hopefully be applied for liquid food packaging. Another type of Algae has also been researched: *Ulva* sp. from the Arabian Gulf. By using whole biomasses combined with plant-based plasticizers like *Plantago ovata* seeds and polyethylene glycol, biodegradable bioplastics with excellent tensile properties and thermal stability were able to be produced (Nermin El Semaary). This approach not only reduces waste and costs but also presents a sustainable alternative to traditional plastics, contributing to environmental conservation efforts. These studies collectively highlight the promising potential of algae-derived materials in tackling environmental issues and promoting sustainable practices.

Plant Coatings

Plant-based packaging poses an intriguing and improvement-convincing argument in the active packaging discussion. One example is made up of paper sheets treated with TiO₂ nanoparticles: the paper's tensile strength, air permeability, and water vapor properties were all

significantly enhanced by the treatment (Nermin El Semary). The mechanical, barrier, and thermal characteristics of the paper are improved through the use of sugarcane fibers and TiO₂ nanoparticles. By oxidizing membrane lipids, which damages bacterial cell walls and cytoplasmic membranes and prevents bacterial growth, TiO₂ nanoparticles are very useful in establishing bacterial inhibitory zones (Maślana et al.). The study, which used various ratios of SA/TiO₂NPs@OCN, showed that treated papers outperform untreated controls in terms of burst strength, tear resistance, tensile strength, air permeability, and water vapor permeability. Applying these papers to current paper-based packaging could hopefully increase the durability and overall performance of the materials. The study's photographs demonstrate that strawberries maintained with these treated paper sheets kept their freshness for an additional eighteen days when compared to those preserved with control sheets, highlighting the treatment's efficacy in prolonging shelf life and preserving food quality. Moreover, the stability of these films is validated by FTIR and TGA tests, demonstrating its practicality. Another Study made materials for sustainable food packaging by using beechwood xylans, cellulose nanofibers, and lignosulfonates (Silva et al.). The resulting nanocomposite films demonstrated excellent homogeneity, thermal stability, mechanical properties, antioxidant capabilities, and UV barrier effectiveness (Silva et al.). The films also effectively delayed fruit browning during storage, highlighting their potential in active food packaging applications and the viability of using low-cost, eco-friendly resources. There have also been innovations towards creating antioxidants for food packaging using study natural colorants from beetroot peel extract (BPE) in bioactive antioxidant films for food packaging. Incorporating BPE into gelatin-sodium alginate films significantly improves mechanical, physical, antioxidant, and antibacterial properties, particularly at higher concentrations (Moufida Chaari). During 14-day storage at 4°C, meat samples coated with these films showed reduced microbial deterioration, delayed oxidation, and improved sensory attributes, highlighting their potential as an environmentally friendly alternative to synthetic additives in food packaging (Moufida Chaari). Overall, these studies show how plant-based treatments and alternatives can be used to create cutting-edge, environmentally friendly food packaging solutions.

Discussion

We have covered many different natural food packaging additives and alternatives that offer an immense opportunity to improve food quality and shelf life, as well as help the environment. However, many concerns remain around the stability and integrity of these materials in comparison with current single-use plastics. In direct comparison to petroleum-based plastics, studies we reviewed emphasize some attributes where alternative materials are superior to traditional plastics like polyethylene such as heat stability and melting temperature. Potential solutions to the limitations revealed by these studies include mixing various biopolymers or using sophisticated formulation techniques to increase the stability and cost-effectiveness of natural packaging materials. Combinations like these could increase the strength of the packaging and make it more well-suited to act as a replacement for plastic.

Studies on alternative packaging materials often start debates and disagreements within the scientific community, particularly concerning their practicality and effectiveness compared to traditional options. For example, whereas natural essential oils and chitosan-based coatings in packaging have demonstrated encouraging antibacterial qualities, concerns remain over their long-term durability and cost-effectiveness. Despite its subpar durability, algae-based packaging offers an array of strong properties such as flexibility, antimicrobial properties and low environmental impact. We recommend more resources be put towards turning algae into large-scale packaging that is suitable for carrying food all across supermarkets. Algae could be a sustainable option as the limited supply of petroleum struggles to meet the demands of an ever-growing human population. Algae-based materials offer a renewable alternative that can help alleviate the pressure on finite petroleum resources, contributing to long-term environmental sustainability. As petroleum reserves continue to diminish, the importance of sustainable and renewable alternatives like algae becomes increasingly critical. On the other hand, due to factors such as changing pH levels and temperatures, there is now a harmful excess of algae in the oceans. This surplus of algae could solve the problem of over-reliance on petroleum-based goods as the amount of petroleum begins to diminish. Furthermore, Algae can be grown and harvested, making it a potentially endless source of material.

Others highlight the possibilities for using essential oils with antibacterial qualities—like thyme and cinnamon—integrated into packaging as essential oils show high antimicrobial effects when incorporated into packaging. They help slow down and reduce the growth of mold, help preserve the food giving it a longer shelf life, and can even act as an insect repellent. With that in mind, there are still problems with incorporating essential oils. Some studies concluded that when compared to the control, the food held in essential oil packaging tasted different. Essential oils are expensive, posing obstacles to scalability and feasibility. One possible solution would be microencapsulation of the essential oils. With microencapsulation, many strong odors and tastes can be masked, which would help solve the problem with taste. Microencapsulation allows for a slower and more direct release rate, which could in turn require less essential oil to execute the same anti-microbial properties.

Subsequent investigations may concentrate on refining the incorporation of essential oils and additional bioactive substances to optimize their effectiveness while reducing expenses. Stronger testing protocols to assess long-term efficacy and environmental impact could offer more precise recommendations for using these substitutes. There are still many unanswered questions about natural and biobased packaging materials. Further research is required to determine these materials' long-term stability, environmental effects, and potential interactions with food. There is not yet enough research on these materials' cost-effectiveness in comparison to conventional packaging options and their scalability for industrial applications. The growing replacement of traditional plastics with natural and biodegradable materials, such as chitosan, alginates, and essential oils, is one of the emerging themes in sustainable packaging research. Technological developments in nanotechnology, such as the encapsulation of essential oils in halloysite nanotubes, are a major trend in improving the functionality of packaging materials.

The field has demonstrated plant-based materials and waste products, such as algae and beetroot peel, to produce bioactive packaging films that can improve food preservation and benefit the environment.

To continue to explore the potential of sustainable food packaging, future research should concentrate on a few important aspects. This entails creating more affordable manufacturing processes for natural and biobased materials, enhancing their performance and stability in a range of settings, and carrying out thorough life cycle analyzes to comprehend the environmental impact of these materials. To guarantee safety and efficacy, more research should look into the materials' possible interactions with food products as well as their scalability for large-scale industrial use.

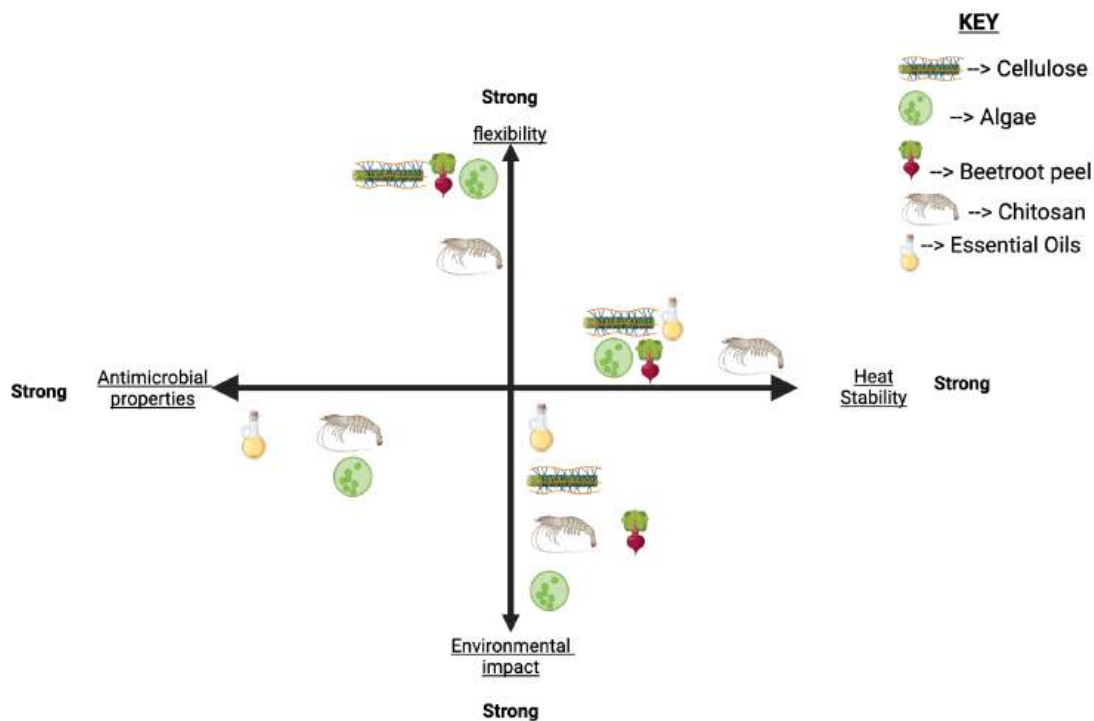


Fig 1: Summation of the Heat Stability, Flexibility, Antimicrobial Properties, and Environmental Impact for each food packaging component. These are plotted on axes radiating from a central point. The distance from the center represents the strength or effectiveness of each property—stronger attributes are plotted further from the center, while weaker attributes are closer to the center.

Material	Heat Stability	Flexibility	Antimicrobial properties	Environmental impact
Chitosan	Strong (up to 150°C)	Medium	Medium	Medium
Algae	Medium	High	Medium	strong
Essential Oils	Strong (up to 150°C when microencapsulated)	ND	Strong	Poor
Beetroot Peel Extract	Poor	High	ND	Medium
cellulose	Strong (up to 153°C)	High	ND	medium/poor

Material	Name of Study	Durability	Preserving Flavor	Microorganisms
Chitosan	Liquid smoke Chitosan for seabass	The nanofibers retained 85% of their mass up to 150°C, ensuring the packaging's durability under heat treatment conditions.	ND	<p>TMABc (Total Mesophilic Aerobic Bacteria Count):</p> <ul style="list-style-type: none"> - Initial: 2.92 log CFU/g <p>Day 7:</p> <ul style="list-style-type: none"> - Control: 7.44 log CFU/g - CN: 5.03 log CFU/g - LSCN: 7.14 log CFU/g - LSTLCN: 4.5 log CFU/g <p>TYMc (Total Yeast and Mold Count):</p> <ul style="list-style-type: none"> - Initial: 3.43 log CFU/g <p>Day 7:</p> <ul style="list-style-type: none"> - Control: 8.32 log CFU/g - CN: 4.76 log CFU/g - LSCN: 7.12 log CFU/g - LSTLCN: 7.05 log CFU/g
chitosan	Fat resistance properties of chitosan-based paper packaging for food applications	Improved durability with chitosan-paper bi-layers compared to untreated paper.	ND	<p>Bacteria Count (TMAB): Dropped from 7.44 to 4.5 log CFU/g with the best coating</p> <p>Cold Germs (TPB): Dropped from 8.46 to 7.07 log CFU/g with the best coating</p> <p>Yeast and Mold (TYMc): Dropped from 8.32 to 7.05 log CFU/g with the best coating</p>
Chitosan	Chitosan and broccoli	7 days - Microbial Decay Reduction: 2.5–3.5 log CFU/g reduction in mesophilic and psychrotrophic bacteria counts.	Higher scores for treated samples (dark green, bright, score of 5) - Texture:	<p>In Vitro Assay (Agar Diffusion Method)</p> <ul style="list-style-type: none"> - Tea Tree EO: <ul style="list-style-type: none"> - E. coli: 18.0 ± 1.0 mm - Native microflora: 17.8 ± 1.2 mm - Rosemary EO: <ul style="list-style-type: none"> - E. coli: 19.0 ± 2.3 mm - Native microflora: 16.9 ± 2.0 mm

			<p>Treated samples remained crispy (score of 5) Smell: No significant differences Browning: CH coatings inhibited browning.</p>	<ul style="list-style-type: none"> - Pomegranate: <ul style="list-style-type: none"> - E. coli: 18.8 ± 0.9 mm - Native microflora: 19.5 ± 1.8 mm - Resveratrol: <ul style="list-style-type: none"> - E. coli: 17.5 ± 1.1 mm - Native microflora: 16.6 ± 1.5 mm - Pollen: <ul style="list-style-type: none"> - E. coli: 16.5 ± 0.9 mm - Native microflora: 17.0 ± 1.2 mm - Propolis: <ul style="list-style-type: none"> - E. coli: 16.0 ± 1.0 mm - Native microflora: 16.2 ± 1.5 mm <p>In Vivo Assay (Pathogen Survival)</p> <ul style="list-style-type: none"> - Tea Tree EO : <ul style="list-style-type: none"> - L. monocytogenes: 5 log reduction - E. coli: 3 log reduction at 0.5–1 µL/mL - Propolis : 5–7 log CFU/g reduction at 20 µL/mL - CH Plus Tea Tree EO : Significant reductions between days 5 and 7
Algae	Novel Active Food Packaging Films Based on Gelatin-Sodium Alginate Containing Beetroot Peel Extract	Adding beetroot peel extract (BPE) to the film improved its strength and flexibility. The strongest film could stretch up to 91.1% before breaking, with a tensile strength (TS) of 36.43 MPa.	BPE films helped preserve the sensory qualities of meat, maintaining appearance, color, aroma, and overall acceptability over 14 days of storage.	The BPE films significantly reduced microbial counts in meat: Total Bacteria (APC): Decreased from 7.810 to 5.102 log CFU/g Cold Bacteria (PTC): Decreased from 6.98 to 6.18 log CFU/g
Essential Oil	Release of cinnamon essential oil from polysaccharide bilayer films and its use for microbial growth inhibition in chilled shrimps	ND	The cinnamon essential oil (EO) impacted the flavor of the shrimp, but it was not negatively received by the panelists. The films helped in maintaining the freshness and sensory qualities of the shrimp during chilled storage.	Agar bilayer films consistently showed approximately 1.4 times higher antimicrobial activity than alginate bilayer films across all tested bacteria
Cellulose	Zero-Valent Metal-Incorporated Cellulose Films	High, confirmed by FTIR** and TGA*	ND	>99% reduction in 24h

Algae	Applications of algae for environmental sustainability: Novel bioplastic formulation method from marine green algae	Although algae are rich in precursor materials for bioplastics, they need the addition of strengthening agents, plasticizers, or fillers in order to give a high degree of flexibility and durability to the produced bioplastic.	ND	Both natural polysaccharides are excellent precursors for the formation of bioplastics and are completely biodegradable by soil microorganisms.
Cellulose	Wood-inspired biobased nanocomposite films composed of xylans, lignosulfonates and cellulose nanofibers for active food packaging	All films displayed thermal stability up to 153 °C, indicating their ability to withstand typical sterilization processes required for food packaging.	ND	ND
Essential Oil	Microencapsulation of Essential Oil for Insect Repellent in Food Packaging System	- All microcapsules exhibited thermal stability up to 150 °C, making them suitable for various packaging applications. -The particle sizes ranged from 1 to 10 µm, with microcapsules using SLS having a smooth and spherical shape, indicating better structural integrity compared to those using T80 and P127	ND	All microcapsules demonstrated high insect repellent efficacy (>90%) over a period of 4 weeks, indicating long-lasting repellent properties suitable for protecting food products from pests.
Essential Oils	Antimicrobial, Rheological, and Thermal Properties of Plasticized Polylactide Films Incorporated with Essential Oils to Inhibit <i>Staphylococcus aureus</i> and <i>Campylobacter</i>	Glass Transition Temperature (T_g): The T _g of films was significantly lowered by the incorporation of essential oils, indicating increased flexibility. The clove oil-based film had the lowest T _g . Melting Temperature (T_m): The T _m values also decreased with the addition of essential oils, with the lowest T _m observed in the cinnamon oil-based film.	ND	It can be seen from Table 1, the inhibition zones increased significantly with increasing cinnamon oil concentration (0.4 to 1.6 mL) in the films, and a significantly higher zone of inhibitions (ZoI) was observed for the Gram-negative <i>C. jejuni</i> than the Gram positive <i>S. aureus</i> . It clearly indicates the selectivity of microorganisms towards inactivation induced by the EO. <i>Overall Average for S. aureus: 1.02 reduction</i> <i>Overall Average for C. jejuni: .87 reduction</i>
Essential Oil	Halloysite Nanocapsules Containing Thyme Essential Oil: Preparation, Characterization, and Application in Packaging	ND	ND	When used in packaging applications these antimicrobial agents could extend the shelf-life of goods and inhibit contamination and spoilage by microbes. They act to reduce or obstruct the growth of microorganisms and fungi in foods

	Materials			
Essential Oil	Halloysite Nanocapsules Containing Thyme Essential Oil: Preparation, Characterization, and Application in Packaging Materials	ND	ND	Thyme Oil (TO): Demonstrated strong antibacterial activity against E. coli, effective in eliminating E. coli during the first 5 days and maintaining strong activity for about 10 days. Cinnamon Oil: High effectiveness with complete inhibition of S. aureus and C. jejuni at higher concentrations. Garlic Oil: Least effective, showing the lowest antimicrobial activity against both S. aureus and C. jejuni. Clove Oil: High effectiveness with complete inhibition of S. aureus and C. jejuni.
Carrageenan	Characterization of edible packaging films based on semi-refined kappa-carrageenan plasticized with glycerol and sorbitol	The films show good mechanical properties (high tensile strength and elongation at break) indicating high durability.	ND	ND
	Extraction of agar from Gelidium sesquipedale (Rodhopyta) and surface characterization of agar based films	Good, with high mechanical strength and moderate water resistance	ND	ND
	Starch-vegetable fiber composites to protect food products	Increased storage modulus and decreased moisture content with higher wheat bran content. Improved mechanical properties with higher fiber content.	ND	NS
Essential Oil	Zein-Based Films Containing Monolaurin/Eugenol or Essential Oils with Potential for Bioactive Packaging Application	Moderate tensile strength, increased elongation with eugenol and ML combinations	ND	Effective against Gram-positive and Gram-negative bacteria, yeasts, and molds
Insect waxes	Spray coating of edible insect waxes for liquid food packaging	High durability with <5% tape test failure post-annealing; low water absorption (0.64%); maintains hydrophobicity (>130° contact angle) even after crinkling.	Excellent flavor preservation with high contact angles (>130°) against water, acids, and alcohols; low liquid absorption.	Indirect antimicrobial benefits due to reduced surface moisture and high hydrophobicity

***FTIR Analysis:** This technique was used to evaluate the chemical stability of the films. The FTIR spectra showed no significant changes in the characteristic peaks of the cellulose acetate polymer matrix after the incorporation of the metal nanoparticles. This indicates that the structural integrity of the polymer was maintained, and the metal nanoparticles were well integrated without causing degradation to the polymer matrix.

****TGA Analysis:** TGA was employed to determine the thermal stability of the films. The analysis revealed that the films exhibited high thermal stability, with decomposition temperatures remaining consistent before and after the incorporation of the metal nanoparticles. The presence of metal nanoparticles did not adversely affect the thermal degradation profile of the cellulose acetate films, suggesting that the films can withstand processing and storage conditions without significant degradation.

Conclusion

As researchers seek sustainable food packaging solutions to reduce plastic waste and enhance food preservation, many recently researched innovations have demonstrated significant potential. One such innovation is essential oils, such as thyme, clove, and cinnamon, which have been shown to provide antimicrobial and insect-repellent properties, thereby extending shelf life and improving food safety. Another such innovation is animal-based coatings, including beeswax and shellac wax. Animal-based coatings offer superior hydrophobicity and stability, while chitosan nanofibers infused with bioactive compounds have effectively inhibited bacterial growth and maintained sensory attributes. Algae-based coatings have also proven effective in providing fat barrier properties and reducing treatment costs. Furthermore, plant-based coatings treated with TiO₂ nanoparticles and oxidized nanocellulose have significantly enhanced mechanical and barrier properties, and have been proven to prolong the freshness of produce.

These advancements are highly promising for the future of sustainable food packaging, highlighting the potential of natural, sustainable materials to replace traditional plastic packaging. These new innovations have the potential to meet consumer demand for eco-friendly options reducing the carbon footprint and minimizing waste through the use of biodegradable and recyclable materials and address environmental concerns by decreasing reliance on non-renewable resources and reducing landfill waste. Continued research and development in this field are essential for ensuring long-term stability, economic feasibility, and regulatory compliance, paving the way for a more sustainable future in food packaging. However, despite these promising developments, there are potential obstacles in the implementation of these innovations, among them the feasibility of scaling up production because sustainable materials can be more expensive and less durable than traditional plastics, making large-scale adoption challenging. Further research will be necessary to understand how these innovations can be used in a wide-scale consumer capacity. Furthermore, these packaging innovations have only been tested on a limited amount of produce and other foods, so we don't yet know the full range of possible applications.

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Prion Diseases: a Comprehensive Review of Pathogenicity and Transmission

By Diya Somnali

Abstract

Prion diseases, which can be categorized into sporadic, genetic, and acquired types, involve the misfolding of the normal prion protein (PrP^C) into its pathogenic form (PrP^{Sc}). Prion disease can be either sporadic, genetic, or acquired. Prion disease is incurable and fatal. The pathogenic conversion of PrP^C to PrP^{Sc} involves a structural change which promotes the formation of neurotoxic aggregates that are resistant to degradation. Despite numerous clinical trials, therapeutic strategies have been largely unsuccessful due to the challenges posed by the resilience and propagation of PrP^{Sc}. Attempts to reduce PrP^C levels, inhibit PrP^{Sc} formation, or use immunotherapy have shown limited success due to issues such as poor brain penetration, toxicity, and autoimmune reactions. Future research aims to translate the growing molecular understanding of prion replication into effective treatments. Here, I review the current literature describing the pathogenesis of prion disease and the efforts to develop effective treatments. I also make recommendations for further exploration which could identify viable therapeutics in the future.

Introduction

Prion diseases, or transmissible spongiform encephalopathies (TSEs), constitute a distinct and fatal class of neurodegenerative disorders that affect both humans and other mammals. Prion disease affects about 1.2 people per million, with a median age at the time of death of 67 (Tam, Johnny et. al). Prion diseases are marked by their unusual etiology—the transformation of a cellular protein into a misfolded, pathogenic form that induces similar misfolding in like proteins. The agent at the center of these diseases is the prion protein (PrP), which exists in a benign cellular form (PrP^C) under normal conditions (Figure 1). Under certain pathological circumstances, PrP^C undergoes a profound conformational change into a “Scrapie” isoform (PrP^{Sc}). This review aims to delve deep into the multifaceted aspects of prion diseases, including their intricate classifications, the complex molecular mechanisms underlying their pathogenesis, and the ongoing challenges in developing effective treatments.

Prion diseases are notable not only for their poor prognosis but also for their ability to transmit between individuals and across species. The propagation of these diseases involves the accumulation of PrP^{Sc}, which can seed the conversion of native PrP^C into more pathogenic protein. This templating leads to a cascading of pathogenic events that results in widespread neurodegeneration. The unique mechanism of prion replication challenges conventional understandings of infectious diseases and has profound implications for diagnosis, treatment, and prevention.

The clinical manifestations of prion diseases vary but typically include rapid neurodegeneration, leading to symptoms such as memory failure, personality changes, and difficulties with motor coordination, culminating in death (Ford, Liz et al.). The pathogenic

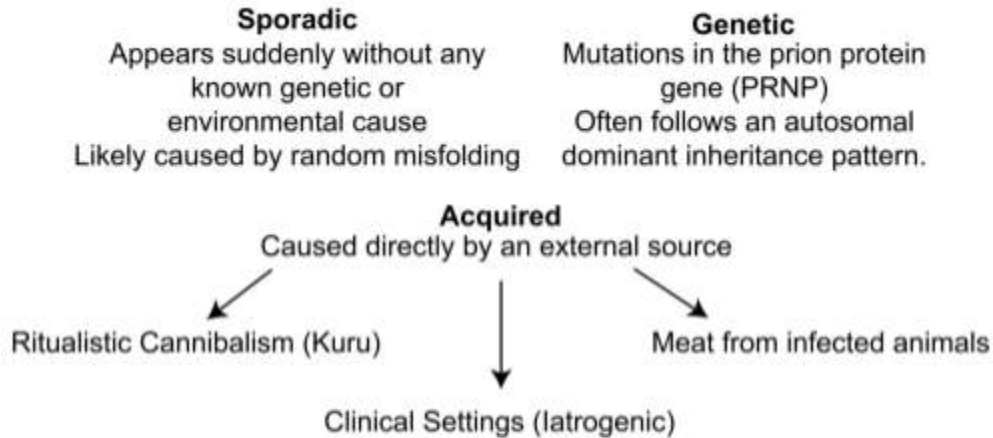


Figure 1. Acquisition routes of prion diseases. A brief description of the three main causes of prion disease, including genetics, sporadicism, and acquirement.

process is notably similar across different species, with diseases such as bovine spongiform encephalopathy (BSE) in cattle, scrapie in sheep, and chronic wasting disease (CWD) in deer and elk mirroring human conditions like Creutzfeldt-Jakob Disease (CJD), Gerstmann-Sträussler-Scheinker Syndrome (GSS), and kuru (Baiardi et. al). These diseases can arise sporadically, from genetic mutations, or through transmission, which includes iatrogenic and dietary routes. Each form of disease leads to distinct deposition patterns of PrP^{Sc} in the brain, contributing to the diverse clinical and pathological profiles observed within this disease class.

This comprehensive review explores the detailed categorization of prion diseases, delves into the molecular pathogenesis that defines them, and discusses the significant challenges that have hindered the development of effective therapeutic measures. By integrating findings from advanced research and clinical trials, this review aims to present an enriched understanding of prion diseases and highlight the critical areas where further investigation and innovative approaches are needed to combat these devastating conditions.

Categorization

Human prion diseases are classified into three primary categories based on their etiology: sporadic, genetic, and acquired. This classification not only helps in understanding the origin of the disease but also guides the clinical approach to diagnosis, management, and research strategies (Figure 1).

Sporadic Prion Diseases

The most common form of prion diseases is sporadic, with Creutzfeldt-Jakob disease (CJD) being the predominant type (Tam, Johnny et. al). Sporadic prion diseases occur in the absence of any known risk factors or genetic mutations, suggesting a random misfolding of PrP^C

into PrP^{Sc}. This form appears without warning and is not associated with any familial pattern or external source of infection. The mechanism behind the sporadic occurrence is not well-understood but is hypothesized to involve stochastic events within the normal protein folding processes in the body. Such events may be influenced by unknown environmental factors or intrinsic cellular processes that have yet to be fully elucidated (Denouel, Angéline et al).

Genetic Prion Diseases

Genetic prion diseases arise from pathogenic mutations in the prion protein gene (PRNP). These mutations increase the propensity of the prion protein to misfold, facilitating the spontaneous formation of PrP^{Sc}. Familial diseases like Gerstmann-Sträussler-Scheinker syndrome (GSS), fatal familial insomnia (FFI), and familial CJD are examples where prion diseases transmit through families with a genetic basis (Gough et al.). These conditions often exhibit a pattern of inheritance that follows an autosomal dominant trait, meaning a single mutated copy of the gene can increase an individual's risk of the disease. Genetic screening can identify mutations in the PRNP gene, providing crucial information for family counseling and managing the disease progression in symptomatic and asymptomatic carriers (Mastrianni).

Acquired Prion Diseases

Acquired prion diseases are those contracted through an external source of infection. This category includes variant Creutzfeldt-Jakob disease (vCJD) often associated with the consumption of beef products contaminated with the prion protein from cattle affected by bovine spongiform encephalopathy (BSE), also known as "mad cow disease" (Will, Robert G). Another example is kuru, historically found among the Fore tribe in Papua New Guinea and transmitted through endocannibalism. Specifically, funeral rites of the tribe involve consumption of the deceased (Liberski et. al). Iatrogenic CJD is another form, transmitted through medical procedures that inadvertently transfer prion-contaminated tissues or instruments from one person to another (Brown et. al). This category highlights the transmissible nature of prion diseases, emphasizing the critical need for stringent controls in medical procedures and the handling of biological materials from affected individuals. Due to small structural differences in PrP^C among different species, interspecies prion transmission is usually highly impeded (Figure 2) (Afanasieva et. al).

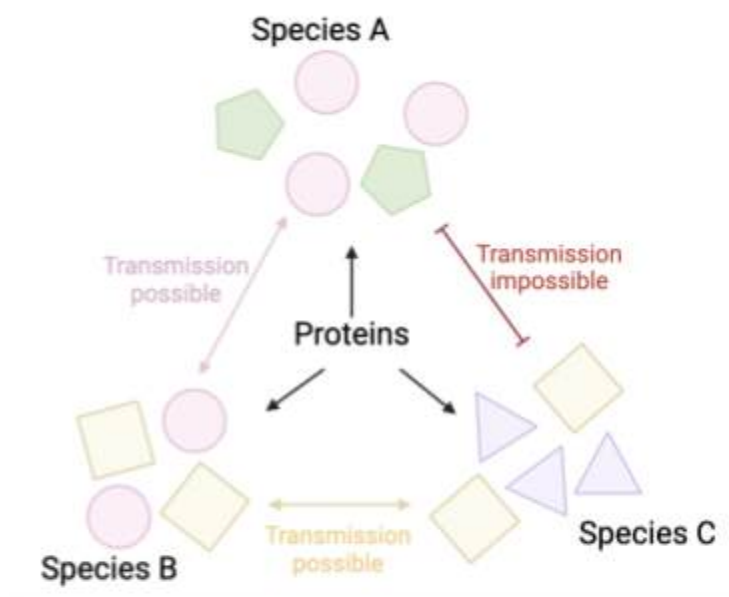


Figure 2 Transmission of prion diseases across species. Interspecies transmission of prion diseases is usually impossible due to structural differences.

These diseases further manifest distinct neuropathological features depending on the deposition patterns of PrP^{Sc} in the central nervous system. The sporadic and genetic forms typically show widespread spongiform changes throughout the brain. In contrast, the acquired forms, particularly those linked to dietary exposure, might present more localized amyloid plaques (Jankovska et. al). These pathological distinctions are crucial for neuropathologists in diagnosing the type of prion disease, which in turn influences treatment approaches and prognostic evaluations.

Understanding the categorization of prion diseases is essential for developing targeted therapies, preventive measures, and for the broader implications of public health related to food safety and medical practices. Each category presents unique challenges and opportunities for research, particularly in uncovering the precise mechanisms of disease transmission and propagation within and between species.

Pathogenesis

The pathogenesis of prion diseases revolves around the transformation of the normal cellular prion protein (PrP^C) into its pathogenic isoform, PrP^{Sc}. This transformation involves a profound conformational shift from a structure that is predominantly alpha-helical to one that is rich in beta-sheets (Figure 3) (Eghiaian et. al). This change not only makes PrP^{Sc} resistant to proteolytic digestion but also promotes its aggregation into neurotoxic forms. The pathogenic process of prion diseases is multifaceted, involving genetic predispositions, cellular interactions, and complex neurotoxic mechanisms.

Genetic Predispositions

The susceptibility to prion diseases often involves genetic factors, most notably mutations in the PRNP gene that encodes PrP^C. Such mutations can alter the conformational stability of the protein, making it more prone to misfold into PrP^{Sc}. Familial forms of prion diseases such as GSS, FFI, and familial CJD are directly linked to specific mutations in the PRNP gene (Bernardi et. al). These genetic alterations can reduce the energy barrier for the conformational transition of PrP^C to PrP^{Sc}, facilitating the spontaneous formation of the disease-causing isoform. Genetic screening and molecular analysis of PRNP in families affected by prion diseases have provided significant insights into the hereditary nature of these conditions and have highlighted the role of genetic counseling in managing these diseases (Goldman et. al).

Cellular Interactions

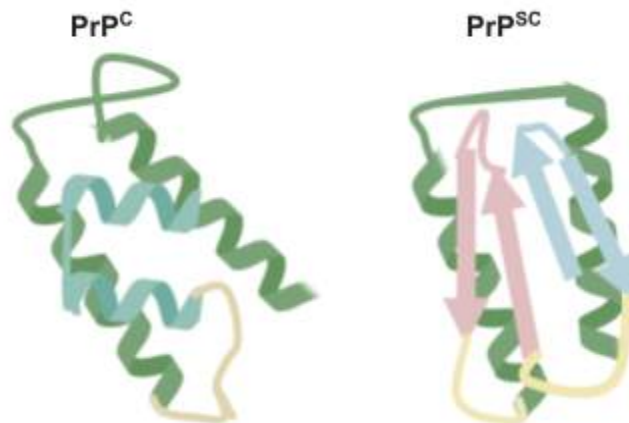


Figure 3 Cellular vs. Scrapie forms of PrP.
The Scrapie form displays beta pleats in addition to the alpha helices.

At the cellular level, the interaction between PrP^C and PrP^{Sc} is critical in the pathogenesis of prion diseases. PrP^C is typically located on the cell surface, anchored by a glycosylphosphatidylinositol (GPI) anchor, where it performs its normal physiological functions, involving neuronal differentiation and T-cell activation (Cazaubon et. al). The pathological PrP^{Sc} can recruit the normal PrP^C and stimulate its conversion into more PrP^{Sc} through a template-assisted mechanism (Figure 4) (Wickner et. al). This process not only amplifies the quantity of the pathogenic protein but also contributes to the spread of the disease within the central nervous system. The exact cellular mechanisms and factors influencing this conversion are subjects of intense research, focusing on how PrP^{Sc} interacts with PrP^C and the role of cellular cofactors in this process.

Neurotoxic Mechanisms

The accumulation of PrP^{Sc} leads to the formation of neurotoxic aggregates that disrupt normal cellular functions. These aggregates interfere with cellular homeostasis and can lead to

the impairment of the ubiquitin-proteasome system, a critical pathway for protein degradation (Lin et. al). This impairment can cause a buildup of misfolded proteins, exacerbating cellular stress and leading to neuronal death. The aggregates may also induce neuroinflammation by activating microglial cells, which further contributes to neurodegeneration (de Melo et. al). Moreover, the presence of PrP^{Sc} can affect synaptic function and neuronal communication, leading to the clinical symptoms observed in prion diseases such as cognitive impairment, motor dysfunction, and ultimately, death.

Understanding the complex pathogenesis of prion diseases is essential for developing effective therapies and preventive measures. Future research efforts are directed towards unraveling the intricate interactions at the molecular and cellular levels that lead to PrP^{Sc} propagation and identifying potential therapeutic targets to inhibit or reverse the misfolding process. Additionally, insights into the neurotoxic mechanisms may offer avenues for symptomatic treatments that could alleviate the burden of these diseases.

Clinical Trials

The inherent resistance of the pathogenic prion protein (PrP^{Sc}) to proteolytic enzymes and

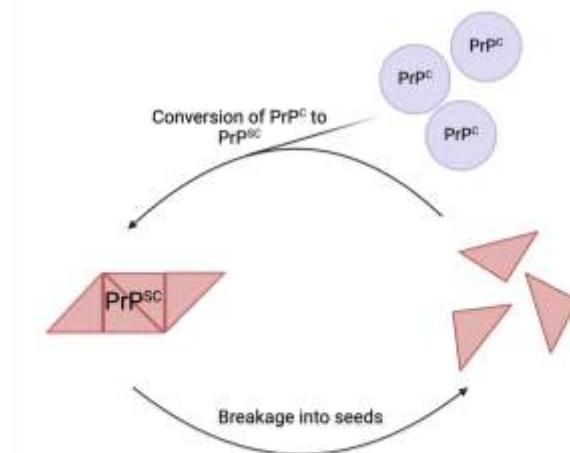


Figure 4. Conversion of cellular protein into prions. PrP^C is converted into PrP^{Sc} by prion 'seeds', then converts back into 'seeds' to convert more PrP^C.

its ability to propagate by seeding the conversion of normal prion protein (PrP^C) into more of itself have presented substantial obstacles in developing effective treatments for prion diseases (Burchell et. al). Historical and ongoing research efforts have explored a variety of therapeutic approaches, aiming primarily at reducing the levels of PrP^C or blocking its pathological conversion to PrP^{Sc}. Despite these efforts, clinical trials and therapeutic experiments have generally resulted in minimal efficacy, failing to significantly alter the course of disease progression.

Reduction of PrP^C Levels

One of the initial therapeutic strategies was to reduce the substrate available for conversion, PrP^C. This approach hypothesized that lower levels of PrP^C would result in reduced formation of PrP^{Sc} and thus slow the progression of the disease. Genetic knockout studies in animal models provided some support for this theory, showing that animals lacking PrP^C were resistant to prion disease (Salvesen et. al). However, translating this approach to human therapies has proven challenging due to the functions of PrP^C and the potential side effects of drastically reducing its levels in humans.

Inhibition of PrP^{Sc} Formation

Another major area of research has focused on developing compounds that can inhibit the conversion of PrP^C to PrP^{Sc}. Various molecules, including polythiophenes, amphotericin B, and Congo red, have shown some ability to inhibit this conversion in vitro (Margalet et. al). Despite these promising in vitro results, these compounds have not been successful in clinical settings. Issues such as poor blood-brain barrier penetration, toxicity at effective doses, and limited efficacy in advanced stages of the disease have hindered their clinical application.

Immunotherapy Approaches

Immunotherapy has been considered a promising approach to treat prion diseases by targeting PrP^{Sc} for clearance from the brain or by stabilizing the conformation of PrP^C to prevent its misfolding. Active immunization with prion protein fragments and passive administration of anti-PrP antibodies have been tested in animal models (Brazier et. al). While some studies reported delayed disease onset, the overall results have been inconsistent, and similar strategies in humans raise concerns about potential autoimmune reactions and the difficulty in achieving sufficient antibody penetration into the central nervous system.

Challenges in Clinical Trials

Clinical trials for prion disease treatments have been fraught with challenges, including the rapid progression of the disease and the late stage at which diagnosis typically occurs. Trials with drugs like amantadine, flupirtine, and quinacrine have demonstrated little to no extension in survival or improvement in disease symptoms (Shim et. al) For instance, quinacrine was initially found to inhibit PrP^{Sc} formation in cell cultures but failed to show efficacy in clinical trials, with patients showing no significant improvement in survival or disease progression (Gayraud et. al).

Future Directions

The insights gained from past research and clinical trials, however challenging, are invaluable. They highlight the critical need for early diagnostic tools that can detect prion diseases before clinical symptoms manifest and for therapeutic strategies that can intervene at a molecular level early in the disease process. Innovations in genetic screening, biomarkers, and

advanced imaging techniques need to be integrated into clinical practice to facilitate earlier diagnosis and tailored therapeutic interventions.

Therapy and Research

The path forward will require a concerted effort to translate the molecular understanding of prion replication and the body's response to prion accumulation into effective treatments. This may involve novel pharmacological agents that can specifically target pathological PrP^{Sc}, immunotherapeutic approaches that harness the body's immune response to prions, or cutting-edge technologies like CRISPR for gene editing that might correct or mitigate the effects of deleterious mutations in the PRNP gene (Castle et. al).

Multidisciplinary and Collaborative Approaches

Furthermore, tackling prion diseases will necessitate a multidisciplinary approach that combines neurology, molecular biology, pharmacology, and other fields to develop comprehensive strategies for managing these diseases. Collaboration across various scientific and clinical disciplines, along with partnerships with the biotechnology and pharmaceutical industries, will be essential. Such collaborative efforts can accelerate the pace of discovery and the application of research findings to patient care.

The Role of Global Health and Regulatory Bodies

Prion diseases, with their ability to cross species barriers and their implications for food safety and blood supply, also pose significant public health challenges and a pronounced need for stringent regulatory oversight and enhanced global surveillance. Regulatory bodies and global health organizations must work together to ensure stringent controls and safety protocols in food production and medical procedures to prevent the transmission of prion diseases.

Integration of Advanced Diagnostics and Therapeutics

Critical insights derived from these challenges emphasize the imperative need for early diagnostic modalities that can detect prion diseases prior to symptomatic manifestation and therapeutic strategies that can interrupt the prion pathogenesis at a molecular level before extensive neurological damage occurs. Emerging technologies in genetic screening, molecular biomarkers, and sophisticated neuroimaging must be refined and integrated into clinical protocols to facilitate preemptive diagnosis and intervention.

Novel Therapeutic Avenues and Molecular Targeting

Future therapeutic initiatives must pivot towards leveraging molecular insights into prion replication mechanisms and host-prion interactions. This may include the development of small molecule inhibitors that can specifically target misfolded PrP^{Sc}, advanced immunotherapeutics to modulate the immune response effectively, and potential applications of CRISPR-Cas9 mediated gene editing to correct or suppress deleterious mutations in the PRNP gene (Ma, Jiyan).

Conclusion

Taken together, while the path to understanding and effectively treating prion diseases is fraught with complexity, the relentless pursuit of advanced scientific knowledge and innovative treatment modalities holds promise. The integration of cutting-edge scientific research, combined with global collaborative efforts and the application of novel biotechnological advances, is essential for achieving significant breakthroughs in the prevention, management, and eventual eradication of prion diseases. As we enhance our molecular arsenal and refine our strategic approaches, the scientific community is poised to unravel the intricacies of prion pathologies and improve clinical outcomes for those afflicted.

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“Pale, Male, and Yale” Diplomacy: Analyzing the Impact of Gender in Diplomacy on Promoting Global Gender Equality By Alexia Lara

Abstract

The lack of cisgender female representation in diplomatic circles and foreign policy organizations has led to largely cisgender male-dominated foreign policymaking. However, the public sector has seen efforts towards increasing female representation. In Congress, women are more likely than their male counterparts to introduce foreign policy legislation that targets action toward women and girls because of surrogate representation. Surrogate representation, initially coined by Harvard political scientist Jane Mansbridge in 1998, occurs when an elected official takes on forms of identity-based representation that exceed normative representation standards. This article examines whether there is a difference in the effectiveness of male versus female diplomatic visits to foreign countries in promoting gender equality issues. By comparing data on gender policies with data from diplomatic visits, I analyze the impact of female diplomatic visits. Through quantitative analysis controlling for regime type, gender inequality index, and gender of the diplomats themselves, there is no evidence that female diplomats are more successful in terms of gender equality outcomes. While not dismissing the value of female diplomats, this work discusses the limitations of quantitative studies on their impact. Although a structural change of adding more female diplomats may not be the answer, the mere presence of females in diplomatic and ambassadorial positions highlights the steps to break heteropatriarchy in the foreign service sector.

Introduction

The U.S. State Department is overwhelmingly “pale, male, and Yale,” a famous adage in diplomatic and journalistic circles highlighting the lack of racial, gender, and socio-economic diversity that has persisted for centuries (Bass 2020). Cisgender men have long dominated the diplomatic sector, and opportunities for the advancement and employment of cisgender, female diplomats are often a low priority. According to the 2022 Women in Diplomacy Index results, women remain largely underrepresented in ambassadorship positions worldwide, with women comprising 21.6% of total ambassadors (Chebab 2022). When a woman does hold a position of authority or power, she may still be overpowered by men. Diplomatic powers wielded by leaders at the highest levels of the State remain primarily in the hands of men. At the ministerial level, the sphere of foreign affairs remains disproportionately dominated by men, with women ministers holding just 25% of this type of portfolio (Mohammed 2023). The underrepresentation of women in diplomatic circles leads to a myriad of issues. Primarily, when females are not represented or included in the decision-making process, policies are passed and enacted under a hegemonic masculine lens that does not factor in the nuances of women like reproductive health, socialization, and gender roles. Moreover, legislation passed under male-dominated circles most likely excludes issues surrounding women because of the absence of a female ambassador to

represent the voices of women worldwide. Thus, the representation of female ambassadors in policymaking is vital to the continued inclusion of gender-based issues in legislation.

To further investigate this, I ask: Is there a difference in the effectiveness of male versus female diplomatic visits to foreign countries when promoting gender equality? I argue that women are more likely to be seen as representing women's issues like gendered violence, economic empowerment, and peace and security (U.S. Department of State) by diplomatic officials because it is in the national interest of women to combat these issues. Because women's issues will be discussed more, female diplomatic visits are more likely to promote gender equality. Thus, with the inclusion of women in diplomatic policy making, issues concerning women's rights are more likely to be discussed because of their ability to act as global surrogates, individuals who introduce legislation on behalf of women worldwide (Angevine 2016).

The intersection of foreign service and gender is based on preconceived stereotypes—such as the notion about women's family responsibilities and their inability to adequately negotiate—that push women out of the diplomatic sector. I analyze the social, political, and gendered gap determining the likelihood of a diplomatic visit promoting gender equality. Building on prior work examining the gendered nature of foreign service (Erlandsen et al.; Towns; Angevine; Baskaran and Hessami; Warsi), I analyze the intersection between gender and foreign service. This work also significantly borrows from The University of Gothenburg's GenDip dataset by Ann Towns, investigating the correlation between gender and diplomatic representation. Comparing data from diplomatic visits with data on gender policies in different countries, I analyze the effect of female diplomatic visits. I also study datasets that concern increased gender equality. Additionally, to further analyze the intersection between gender and foreign service, I utilize the findings of political scientist Sarah Angevine, economists Thushyanthan Baskaran and Zohal Hessami, and journalist Farkhanda Warsi. I find that the gender of a diplomat does not directly affect the gender equality scores of a country where the visit is held. This is contrary to initial expectations and expectations in the literature that diplomatic visits by female diplomats are more successful in promoting gender equality because it would allow the increased representation of the female body to discuss pressing gender issues and create gender-targeted legislation in the receiving country.

Surrogate Representation

Prior studies have shown that women are underrepresented in foreign diplomacy (Chebab 2022, Mohammed 2023, Warsi 2020, Erlandsen et al. 2021). Compared to the mere presence of women in legislatures, women's movements and policy agencies can be more impactful in expressing women's perspectives on legislation addressing violence against women. American political scientist Jane Mansbridge initially coined the notion of surrogate representation in 1998, describing when an elected official tackles identity-based representation that exceeds normative representation standards by “acting for” the specific interests of their constituents. Mansbridge furthers surrogate representation as compensation for inadequate voter representation whose candidate lost in their districts or voters whose preferred policies attract a minority group in their

districts (Lavi 2023). Surrogate representation has primarily been observed in the legislative branch, where Congresswomen serve as a global surrogate for women's rights issues as they introduce gender-based foreign policy legislation. Nonetheless, surrogate representation is not restricted to Congress and can further be applied to diplomatic circles. The concept of surrogate representation arises from the significance of representing women in policy making and advocacy. In the context of foreign service, the lack of representation in diplomatic circles and foreign policy organizations has led to largely male-dominated foreign policymaking. Empirically, surrogate representation theorizes the presence of a female diplomat, as a representative of women can increase the likelihood that foreign policy reforms targeted toward women and girls in foreign countries are introduced. As a result, they have the ability to act as global surrogates for issues concerning women, which also affects entire populations. The idea that a female diplomat can represent most women's beliefs and objectives can allow for more successful outcomes of gender equality goals.

Surrogate representation is reflected in Sara Angevine's work. Angevine explains that female Members of Congress are more likely to introduce foreign policy legislation targeting women and girls, indicating their role as global surrogates for women's issues (2016). Surrogate representation can spill over into the diplomatic sector, especially as female diplomats work to promote gender equality during diplomatic visits. The idea that a female diplomat can represent most women can allow for more successful outcomes of gender equality goals. Despite being a non-pivotal minority, women in councils effectively influence policy outcomes, as they can shift the focus and direction of discussions during council meetings.

Women in Politics

As Thushyanthan Baskaran and Zohal Hessami argue in *Women in Political Bodies as Policymakers*, the representation of women in political and local councils can accelerate the desired outcomes of the policies being passed, especially concerning human rights issues like childcare. The representation of women in a diplomatic body can mobilize the discussion and promotion of gender equality in this sector. When observing the possible benefits of female diplomats regarding promoting women's rights during diplomatic visits, it is also essential to understand the dynamic between men and women in the diplomatic sector. In "Gender Difference Among Employees Working In Diplomatic Missions," Farkhanda Warsi reveals that gender differences were observed, with fewer women occupying top positions in most Diplomatic Missions than men. Additionally, women were treated unequally in terms of employment opportunities. Warsi and Baskaran, as well as Hessamani, agree on the notion of gender equality in politics, especially because of the significant lack of female representation.

Diplomatic Visits

In this work, data from Erlandsen et al.² are borrowed and assessed. Erlandsen et al. have conducted extensive and comparative work examining the gendered nature of ambassadorial appointments in Latin America. Their findings are compiled in databases on several variables concerning the data.

One innovation this article proposes is analyzing whether the election of women to the presidency in Argentina, Brazil, Chile, and Costa Rica affected the gender disparity at the top of those nations' foreign service sectors. While other scholars have suggested various factors that might explain the gendered nature behind ambassadorial appointments, few have accounted for the political authority factor. Erlandsen et al. suggest that the inclusion of women in the presidency would allow for (1) women to act on behalf of other women and (2) the appointment of more women due to their gendered networks and homophily (as described by Crowder-Meyer in 2015 and Reyes-Housholder in 2016).

This work also significantly builds upon The University of Gothenburg's GenDip dataset on Gender and Diplomatic Representation led by Towns et al. The GenDip dataset analyzes the changing gendered nature in diplomatic circles and uses several variables concerning the data. This research analyzes a sample from 1968 to 2021 and examines the extent to which states appoint men, women, and other diplomats to various ambassador positions. Within the time frame of 1968-2021, the dataset relies on 10-year increments from 1968-1998 because of the low share of women during that period. For each year, the authors record several variables about the diplomatic visit. Notably, these include the name of the sending country (taken from ISO in 2020), main posting of the diplomat (taken from the Europa World Yearbook), title of the diplomat, gender of the diplomat (taken from the Europa World Yearbook), and the name of the sending country (taken from ISO).

Methods

To better understand how diplomatic visits impact gender equality cross-nationally, I constructed a dataset encompassing variables that measure gender equality scores and female representation in ambassadorial positions. I analyzed five countries' diplomatic visits—Benin, Chile, Mexico, South Africa, and Poland³—across a 10-year period from 1998-2008. The time

² The authors' dependent variable is a binary indicator of whether a diplomatic mission was headed by a woman *ambassador* in a given year or not. This research analyzes a sample of ten Latin American countries from 2000-2018; its analysis unit is a country year. For each year, the authors record the appointment of women ambassadors as heads of diplomatic missions and numerous other variables hypothesized to affect the appointment of women ambassadors. These include discretionary appointment, foreign minister headed by gender, the percentage of women in the cabinet (taken from Nyrop and Bramwell in 2020), the percentage of women with a university degree (taken from UNESCO Institute for Statistics in 2020), the percent of female labor force participation (taken from the International Labor Organization in 2020), the GDP per capita (taken from the World Bank), and the host status (taken from the Correlates of War's National Material Capabilities 5.0 by Singer et al. in 1972). This analysis includes two dummy variables. The first dummy variable is a head of government by gender measured with the value of 1 representing a woman governing a country and 0 if not in any given year. The second dummy variable was adapted from Murillo et al.'s dataset in 2011 and accounts for government ideology, such as the years in power without ideological change and the involvement of a leftist government.

³ Brigeovich's report on the post-Cold War wave of democratization highlights how the most successful transitions to democracy in the 90s are the countries of Benin and Poland. To expand my data to countries outside the regions of

frame began in 1998 partly because of data limitations and because the wave of post-Cold War democratization began in the 1990s.⁴ Moreover, according to Brigeovich's report on regime transitions in Sub-Saharan Africa and Post Communist States amid the wave of democratization, most regimes fully transitioned to democracy by 2008. The data best encapsulates the 10-year period and considers the global political changes and transitions. I captured every diplomatic visit from each of these five countries to every recipient country, with each diplomatic visit representing an observation. This results in a sample size of N=2053. For each observation, I captured meta-data, including the countries' names and the visit date.

For each diplomatic visit, I captured the countries engaging in the visit, the date of the visit, the gender of the ambassador, and the regime type of each country, representing the independent variables as categorical, binary variables. The gender of the ambassador on the visit (*Female*) was coded as 1 if their gender was female and 0 if male, collected from Erlandsen et al. (2021). The regime type variable represents the relationship between the sending and receiving countries' political regime and their intersection with gender equality scores. Incorporating the regime types within my data is an explanatory variable that can impact the outcome of gender equality scores from diplomatic visits. I gathered this variable's data from the Oxford Diplomacy Lab (2023). The Oxford Diplomacy Lab relies on data from the Regimes of the World (RoW) that distinguishes four types of political regimes from 1789 to 2022: closed autocracies, electoral autocracies, electoral democracies, and liberal democracies. The coding scheme for the regime type was 1 if they were the same type and 0 if not.

The dependent variable, or outcome, is the gender equality scores of a country. I argue and hypothesize that since women are perceived as representing women's issues and discussing them more, female diplomatic visits are more effective in promoting gender equality. Therefore, the higher the proportion of visits by female diplomats, the more I expect that country's level of gender equality to increase. To measure increased gender equality, I record the gender equality index of the country in the same year of the diplomatic visit and five years post the visit. I look five years down the road because it takes time for social and political change to occur (Hannigan-Page 2023). I utilize the United Nations Development Programme gender inequality index (GII) to measure increased gender equality in a country because it is a holistic and composite metric of gender inequality that analyzes three dimensions: reproductive health, empowerment, and the labor market. Within the health dimension, the GII measures gender equality by analyzing the maternal mortality ratio and the adolescent birth rate to create the female reproductive health index. The GII also analyzes female and male populations with at least secondary education and female and male shares of parliamentary seats to determine the female empowerment index. The GII analyzes the female and male labor force participation rates to create the index for the female and male labor market. According to the index, a low GII value

Sub-Saharan Africa and Central Europe, I utilize Erlandsen and Hernández-Garza et al.'s dataset that examines the diplomatic visits of ten Latin American countries, to where I selected two of them: Mexico and Chile.

⁴ The data collection borrows from the work of Erlandsen et al., whose dataset provides information starting in 1998.

translates to a low inequality between men and women in the receiving country. Utilizing the GII's indicators, the lowest number would mean reproductive health would be granted a right for all women, female empowerment would be robust—translating into increased female representation in every sector—and there would be equal employment rates and wages. With these results, I analyzed whether the diplomatic visit had an effect in contributing to an increase, decrease, or consistency with the gender index score. The lower a visit scores in the GII index, the more likely a country will improve its women's equality scores.

Results

I first present some general descriptive statistics about diplomatic visits of post-Cold War democratizing sending nations and their relationship to a country's gender inequality index. These appear in Table 1 and Table 2 as well as in Figure 1 and Figure 2. Here, I use the 0-0.9 GII score as the dependent variable on gender equality; this dataset is the most up-to-date and has the fewest missing values. I also note the change in a country's gender inequality score during and five years after a diplomatic visit, with a decrease in the GII reflecting an improving gender equality situation. In Figure 3, I present statistics on the change in GII for diplomatic visits with a female ambassador. Lastly, I display a regression model highlighting statistical significance in some of the variables in Table 3 and Table 4.

These data are, of course, impressionistic, but several items are worth mentioning. First, of the sample size of $N=2052$, 197 were female diplomats, and 1823 were male diplomats. 32 diplomatic visits did not report the gender of the diplomats. This is a limitation of the data collection because the null results could be due to so few causes with female visits. Second, the most significant changes in the GII occurred in countries with different regimes and male diplomats. Third, female diplomats did result—though not strongly—in a decreased gender inequality score in the receiving country. Overall, little evidence suggests a strong relationship between the presence of female diplomats and increased gender equality. The gender inequality score decreased in 181 cases (out of 197) five years after a female diplomat participated in a diplomatic visit. Of course, other factors might facilitate or hinder the work and impact of a diplomatic visit (as well as account for the change in gender equality performance), and this table does not allow the comparison of receiving countries that engaged in diplomatic visits with those that did not.

This analysis finds no substantial effect of female diplomats on gender inequality scores when looking at all receiving countries. This pattern holds when changing the dependent variable to the previous five years. The analysis does reveal, as expected, that regime type matters—democratic countries are more likely to report problems—and that countries in conflict have worse gender equality problems.

Table 1 and Table 2 provide data about the changes in gender inequality scores. Table 1 displays the countries with the top five largest decreases in gender inequality scores, quantifying an improvement in gender equality. Based on Table 1, the most significant decrease in the GII occurred in countries with different regimes and male diplomats. Notably, the United Arab

Emirates and Saudi Arabia, both Middle Eastern countries, displayed the most significant decrease in gender inequality scores. The United Arab Emirates experienced the most significant decrease in gender inequality scores by -0.404 from 1998 to 2003. As both Middle Eastern Nations, the GII change could likely result from the strengthening political climate. Middle Eastern countries saw mass waves of feminist activism in the early 2000s that might have decreased gender inequality scores (Tajali 2024, El-Husseini 2023).

Table 1. Highest changes in gender inequality score

Sending Country	Receiving Country	Year	Same Regime	Female Diplomat	Total Score	Gender Inequality (GII) Year of Visit	Gender Inequality (GII) Five Years After Visit	Change in Gender Inequality Score
Poland	United Arab Emirates	2003	0	0	0	0.597	0.193	-0.404
South Africa	United Arab Emirates	2003	0	0	0	0.597	0.193	-0.404
Mexico	Saudi Arabia	2008	0	0	0	0.66	0.307	-0.353
Poland	Saudi Arabia	2008	0	0	0	0.66	0.307	-0.353
South Africa	Saudi Arabia	2008	0	0	0	0.66	0.307	-0.353

Table 2. Lowest changes in gender inequality score

Sending Country	Receiving Country	Year	Same Regime	Female Diplomat	Total Score	Gender Inequality (GII) Year of Visit	Gender Inequality (GII) Five Years After Visit	Change in Gender Inequality Score
Chile	Lebanon	2008	0	1	1	0.42	0.458	0.038
Chile	Lebanon	2008	0	1	1	0.42	0.458	0.038
Mexico	Lebanon	2008	0	0	0	0.42	0.458	0.038
Poland	Lebanon	2008	0	0	0	0.42	0.458	0.038
Poland	Bangladesh	1998	0	0	0	0.695	0.737	0.038

The sending countries, Poland and South Africa, both were in the process of regime transitions to democracy during this time period and fully transitioned to democracy by 2008. Saudi Arabia experienced the second-largest decrease in gender inequality scores by -0.353 from 2008 to 2013. The sending countries, Mexico, Poland, and South Africa, who visited Saudi Arabia, completed their transition to democracy when the diplomatic visit was held. In two cases (out of the top five), Poland was the sending country when diplomatic visits were held, with Saudi Arabia as the receiving country.

Based on Table 2, the lowest changes in gender inequality score—indeed increases in gender inequality—were observed in five countries with different regimes and two countries with female diplomats (out of the bottom five). Lebanon and Bangladesh both displayed an increase in gender inequality scores five years after the diplomatic visit. Lebanon, a Middle Eastern country, experienced an increased gender inequality score of 0.038. Female diplomats were present in two visits (of the four) to Lebanon in 2008. Similarly, Bangladesh experienced an increased gender inequality score of 0.038 between 1998 to 2003. Chile and Poland were the receiving countries in two cases (out of the bottom five). Both Figure 1 and Figure 2 graph the results from Table 1 and Table 2. Figure 1 and Figure 2 display the changes in gender inequality scores. In Figure 1, South Africa and Poland are the most significant outliers for sending countries where their diplomatic visits correlated to a significant decline in the gender inequality index. As a receiving country, Saudi Arabia is the largest outlier, as it experienced three diplomatic visits from the sending countries of Mexico, Poland, and South Africa in 2008. Chile and Poland are the most significant outliers for sending countries in Figure 2, as they were the most occurring countries to participate in a diplomatic visit to Lebanon and Bangladesh. As a receiving country, Lebanon is the most significant outlier because it experienced four diplomatic visits from the sending countries of Chile two times (out of four), Mexico, and Poland.

Figure 3 displays the changes in gender inequality scores for countries with the same regime and a female diplomat. In 61 cases (95% of cases), the combination of a female diplomat in a diplomatic visit where both the sending and receiving countries had the same regime meant the gender inequality score decreased. The outcome of decreased scores on the GII is improved gender equality for the sixty-one receiving countries. The three cases (out of 64) reported an increase in the gender inequality index, indicating a worsening of gender equality. An increased gender inequality index was observed in 2008 when Panama and Hungary were the receiving countries. Hungary reported two visits in 2008, totaling three cases when Panama was added.

Figure 1. Highest changes in gender inequality score

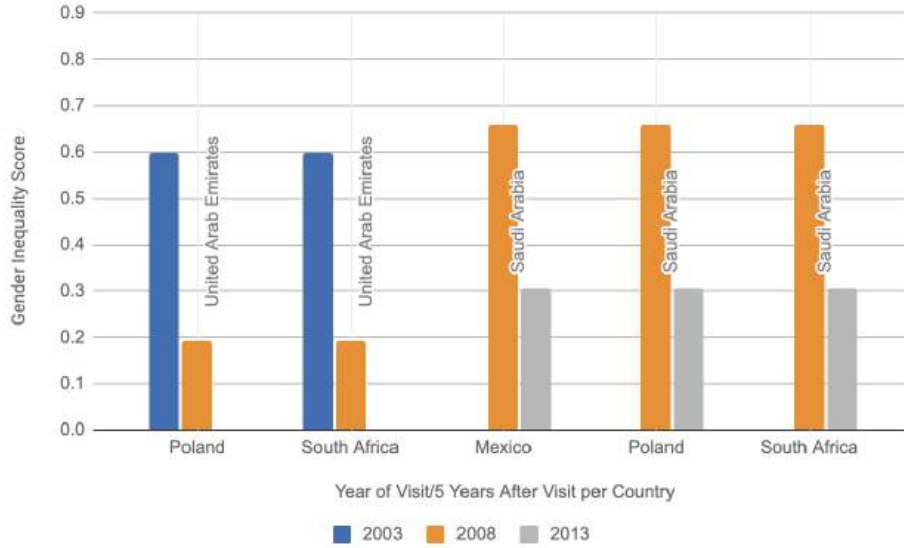


Figure 2. Lowest changes in gender inequality score

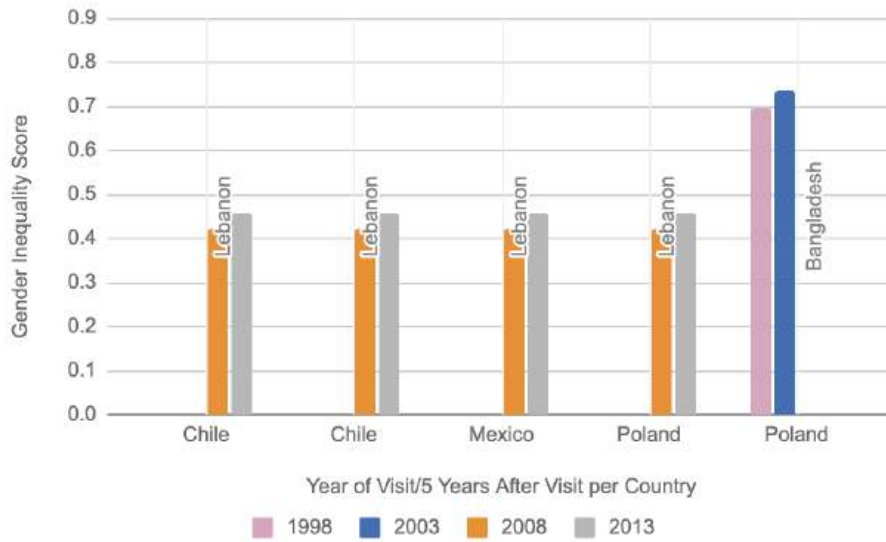
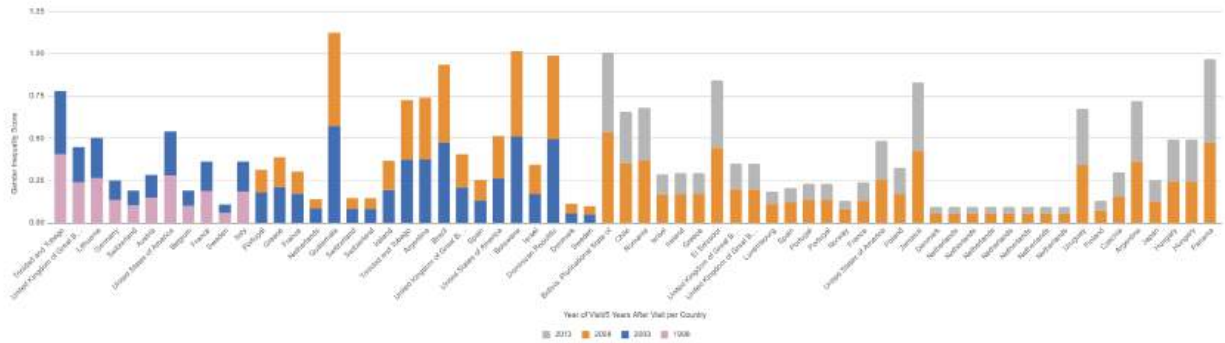


Figure 3. Changes in gender inequality scores with female diplomats



When considering all diplomatic visits held by the sending countries of Chile, Mexico, South Africa, Benin, and Poland, there is a mixed and statistically insignificant relationship between female diplomats and improvements in gender inequality scores. Table 3 displays a positive correlation ($p < 0.001$) between female diplomats and changes in gender inequality. However, the regression model in Table 3 depicts an increase in gender inequality scores by 0.008 when there is a female diplomat, signaling a worsening of gender equality in the receiving country. When both the sending and receiving countries have the same regime type, the regression model displays a statistically insignificant decrease in gender inequality scores by -0.002. When a female diplomat is present during the visit, and the sending and receiving countries have the same regime type, the expected change in gender equality score is -0.003 and statistically insignificant. There is also a positive correlation ($p < 0.001$) in Table 3 and Table 4 for the constant variable representing a male diplomat in a visit where the sending and receiving countries have different regimes. The change in gender inequality for the constant variable is -0.029, signifying an improvement in gender equality.

Table 3. Regression Model for Categorical Binary Variables Coded as 1

<i>Dependent variable:</i>	
Change in Gender Inequality	
Same Regime	-0.002 (0.004)
Female Diplomat	0.008*** (0.002)
Female Diplomat in Same Regime	-0.003 (0.005)
Constant	-0.029*** (0.001)
Observations	1,946
R ²	0.014
Adjusted R ²	0.013
<i>Note:</i>	* $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$

Table 4. Regression Model for Independent Variables Represented by Categorical Binary Variables Coded as 1

<i>Dependent variable:</i>	
Change in Gender Inequality	
Same Regime	0.008*** (0.001)
Female Diplomat	-0.004 (0.002)
Constant	-0.029*** (0.001)
Observations	1,946
R ²	0.014
Adjusted R ²	0.013
<i>Note:</i>	*p<0.05; **p<0.01; ***p<0.001

Table 4 displays a positive correlation ($p < 0.001$) between the sending and receiving countries having the same regime and changes in gender inequality. However, the regression model in Table 4 displays an increase in gender inequality scores by 0.008, signaling a worsening of gender equality in the receiving country. When a female diplomat is present, the regression model displays the change in gender inequality as statistically insignificant and decreases by -0.004.

Conclusion

Various scholars have examined the gender gap in the diplomatic sector, including the lack of representation of female diplomats. To date, there is no consensus on what, if any, effect female diplomats have on gender equality. This paper has assessed the impact of female diplomats in post-Cold War democratizing countries. The findings of this research suggest, however, that female diplomats do not substantially improve gender equality in the years following diplomatic visits that include female diplomats. Moreover, even when assessing the immediate "success" of a female diplomat, this study does not find a strong relationship with increased gender equality in the receiving country. This is not to say that female diplomats do not act for surrogate representation (variables like the discussion of women's rights within visits that this article does not examine). Nevertheless, this study suggests that female diplomats' representation in the foreign service sector remains lacking.

There are a series of reasons why female diplomats do not directly correlate to a substantial or lasting impact on gender equality. Often, they have a limited capacity for cooperation and resolution amidst their visit, perhaps when the ending and receiving countries are of different regimes. Similarly, in some cases, political leaders have been unwilling to embrace their calls for reforms. In other cases, authors would expect slight improvement, given that there is data about the discourse within these diplomatic visits and the direct topics discussed are unreleased primarily to the public. Lastly, it may simply be that the effect of female

diplomats on gender inequality cannot be "seen" because improvements in the receiving countries occurred before the introduction of a female diplomat.

This large-n analysis does not find that female diplomats, by themselves, substantially impact gender equality. This is not to say, of course, that individual female diplomats have no value. There is also a need to identify effective female diplomats and the conditions and mechanisms that allow their work to have a lasting, positive impact. Existing power structures within diplomacy jeopardize the achievement of gender parity. From a diplomatic standpoint, achieving gender equality requires analyzing the gendered system that foreign service was built on, data that this research does not examine.

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Molecular Mechanisms Underlying Tau Pathology and Disease Progression in Alzheimer's Disease and Chronic Traumatic Encephalopathy: Deciphering Molecular Characteristics for Therapeutic Target in Tauopathies

By Elle Scord

Abstract

Tauopathies, a division of neurodegenerative diseases defined by the deposition and aggregation of abnormal tau protein, include a diverse array of conditions that pose a relevant and withstanding concern in neurobiology and medicine. While associated tauopathies may share similar pathological features such as abnormal aggregation of tau protein, they differ in their clinical presentations, neuropathological patterns, and molecular mechanisms. Thus, highlighting the similarities and differences in the isoform variations and molecular pathology of tauopathies is crucial for advancing knowledge in the field and implying therapeutics. This review will comparatively outline the important molecular characteristics of Alzheimer's Disease (AD) and Chronic Traumatic Encephalopathy (CTE) by identifying their neuropathology, dominant isoform significance, post-translational modifications, and cellular response in order to determine unifying characteristics for therapeutic target. Thus, this review aims to provide a detailed analysis of the current knowledge surrounding tauopathies and their molecular characteristics and highlight areas for future research. With enhanced comprehension of the mechanisms that underlie these diseases, diagnostic techniques, potential biomarkers, therapeutics, and prevention mechanisms may be improved. However, conversely, understanding the molecular differences of tauopathies may not only enhance our understanding of these diseases, but also differential treatments for specific tauopathies.

Introduction

I. Function of Tau in the Neuron

Tau is an essential microtubule-associated protein (MAP) predominantly expressed in nerve cells, where it plays a crucial role in promoting the assembly and stabilization of microtubules (Weingarten et al. 1858–62). Microtubules, abundant in neurons, form an integral part of the cell's cytoskeleton, influencing its shape and function (Lasser et al.). Studies isolating tau have revealed its significance in regulating axonal and intracellular dynamics. Within the neuron, tau protein is concentrated in axons, where it attaches to microtubules to stabilize their function. Microtubule stabilization allows for kinesis-mediated transportation of cargo in neurons, therefore influencing neurotransmission (see Figure 1). Tau protein's N-terminal and C-terminal regions interact with proteins involved in cytoskeleton control, as well as motor proteins kinesins and dyneins. Thus, tau regulates intraneuronal dynamics, allowing for variable cytoskeleton rearrangement and synaptic transmission (Bodea et al. 71-94). Broadly speaking, in cases of pathology, abnormal alterations of tau lead to the development of neurodegenerative diseases known as tauopathies (Hernandez and Avila 2219-33).

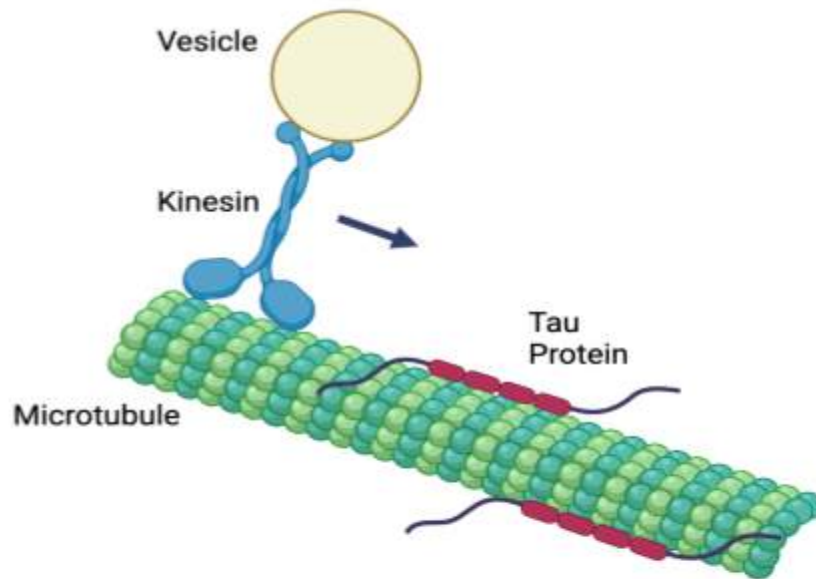


Figure 1: Schematic of Tau protein and microtubule. Tau protein is required for microtubule stabilization, which sets a precedent for the kinesis facilitated transport of cargo in neurons.

Tau is fundamental to the proper functioning of all neuronal subtypes. Notably, within the hippocampus, it is crucial for memory formation and the regulation of axonal transport and synaptic plasticity within hippocampal neurons (Wu et al. 45). There, tau interacts with motor proteins such as kinesins to facilitate the movement of vesicles, mitochondria, and other cellular materials along microtubules, thus regulating synaptic transmission and long term potentiation (LTP) (Sinsky et al. 9207). Interestingly, studies have shown that tau dysfunction can impair LTP in hippocampal neurons, suggesting that tau is necessary for synaptic plasticity, therefore abnormalities of this protein may contribute to learning deficits (Kimura et al. 179). Also prominent in the cerebral cortex, tau has crucial function in cortical neurons. It is specifically vital in preserving the shape and physical stability of nerve cells, maintaining the organizational and connective integrity of neurons (Bodea et al. 71). In subcortical subtypes of neurons such as the thalamus and basal ganglia, tau also modulates sensory relay processing and motor function (Bosch-Bouju et al.). While tau is typically found in these particular neurons of the central nervous system (CNS), specific isoforms such as “big tau” are primarily expressed in the peripheral nervous system (PNS), therefore diversifying its functions (Boyne et al. 279–93; Goedert et al. 393–99). Little is known of the function of big tau in neuronal pathology; however, dysfunction and abnormal accumulation of tau protein is primarily known to cause CNS neurodegenerative disorders (Medeiros et al. 514–24).

Interestingly, recent research has shown that tau may play a physiological role in dendrites and other synaptic interactions (Mietelska-Porowska et al. 4671–713). Aside from its axonal function, tau protein additionally occurs in pre and post synapses in a smaller concentration (Sinsky et al. 9207). In abnormal tau form accumulation, tau protein aggregates

damage tripartite synapses, disrupting the normal synaptic function (Stevenson et al.). Furthermore, microglia have regular interactions with synapses, therefore released extracellular tau could be capable of interactions with proteins present in the synaptic clefts, influencing synaptic functions (Sinsky et al. 9207). Thus, recognizing the complex interaction between tau protein and synaptic function is essential for understanding the pathophysiology of tau associated neurodegeneration and developing interventions aimed at preserving synaptic integrity.

II. Tau Gene Mutations and Isoforms

Previous studies have demonstrated that in humans, the normal functioning tau protein is encoded by the microtubule associated protein tau (MAPT) gene on chromosome 17q21 (Neve et al. 271–80). This gene in particular serves as an interesting subject of inquiry due to its many alterations that may occur in both healthy and abnormal tau production associated with tauopathies, such as isoforms and mutations (Rawat et al. 12841).

One mechanism of the MAPT gene and other genes associated with tau pathology would be expressed through missense mutations (Strang 912–28). Particularly in genes encoding tau proteins or their precursors, heritable and dominant forms of respective diseases may occur through these mutations (Wolfe 1–20). Furthermore, several other proteins have also been isolated and shown association with tau its pathology.

However, the most prominent and relevant alterations to the MAPT gene are isoforms produced as a result of alternative mRNA splicing (Goedert et al. 393–99). As shown in Figure 2, the human tau gene forms a primary RNA transcript, which upon further processing, produces isoforms. Alternative mRNA splicing of the MAPT gene produces six major isoforms known as 0N3R, 0N4R, 1N3R, 1N4R, 2N3R, and 2N4R (Goedert et al. 393–99). In order for tau to serve its optimal function, it is necessary that these 3R and 4R isoforms are distributed in a balance essential to drive proper function within specific brain regions (Vourkou et al. 12985). Studies have shown that 4R isoforms are more prone to forming neurotoxic aggregates than 3R isoforms (Goedert et al. 393–99).

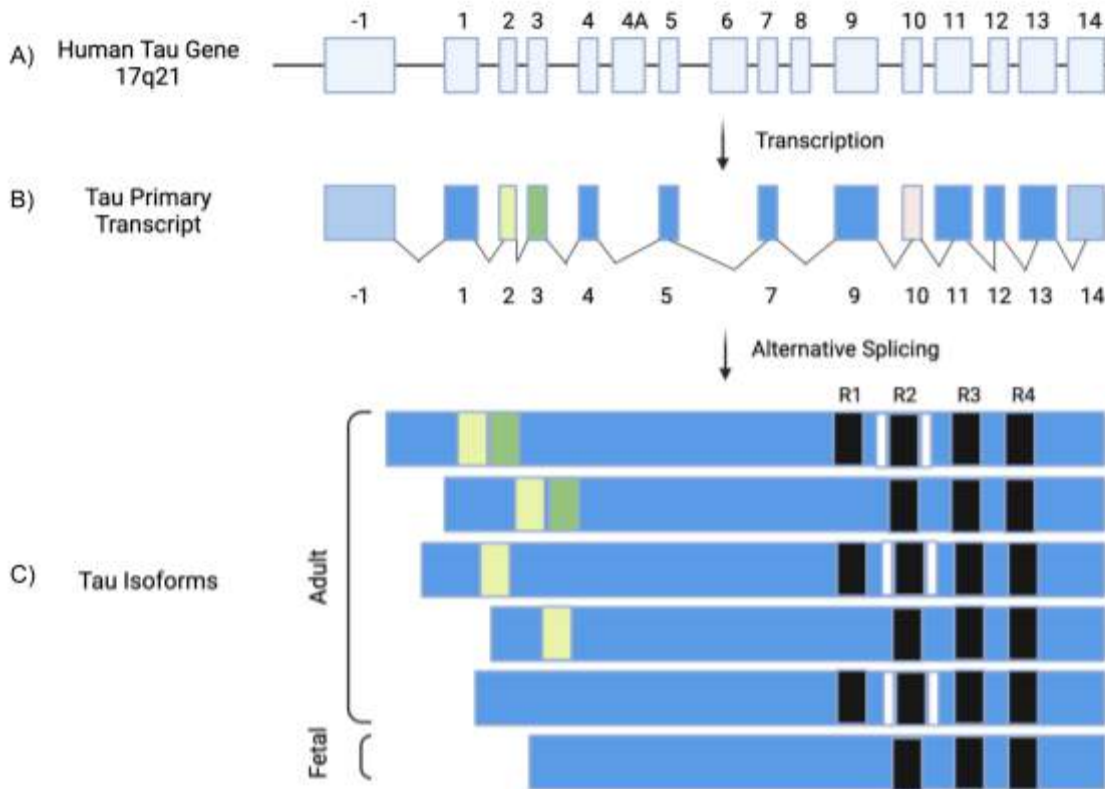


Figure 2. The human tau gene, tau primary transcript, and six tau isoforms that may result from alternative RNA splicing. A) the human tau gene spans approximately 100 kilobases and consists of 16 exons. B) In its primary transcript, all exons and introns are included. C) Following alternative splicing events, six isoforms are produced typically through the inclusion or exclusion of exons 2, 3, and 10.

Consequently, the distribution of tau isoforms within specific brain regions may contribute to increased vulnerability to different tauopathies. In other words, certain isoforms may be more prevalent in regions affected by different tauopathies (Vourkou et al. 12985). Thus, these imbalances have been implicated in the clinical manifestations of neurodegeneration, making them a promising source to identify biomarkers and develop isoform specific interventions for associated tauopathies (Buchholz and Zempel).

III. Post-Translational Modifications

Further, another alteration to tau that is commonly associated with the specialization of the protein through post-translational modifications (PTMs) (Haj-Yahya and Lashuel 6611–21). PTMs are essential to normal tau protein function, as their main purpose is to regulate and increase tau's functional diversity. This allows it to perform specialized tasks in neurons, regulating its diverse functions (VandeVrede et al. 134919). The most prominent examples of tau PTMs include phosphorylation, truncation, nitration, glycation, and glycosylation (Pevalova et al. 346–53). When these modifications are disturbed, they can play a serious role in the

pathogenesis of various neurodegenerative diseases (Blagov et al. 6954). The most prominent of these, and most commonly associated with common tauopathies, is phosphorylation. In normal tau, the protein is phosphorylated for many important functions. For example, it regulates microtubule binding following phosphorylation, and its affinity for microtubules decreases allowing disassembly and reorganization. These processes are necessary for axonal growth and synaptic plasticity (Jahan et al. 359–72). Normal phosphorylation regulates tau’s involvement in axonal transport by affecting tau’s interaction with motor proteins and microtubules, all modulating axonal transport (Stern et al. 1079–87). Further, tau phosphorylation is implicated in neuronal plasticity through its prominent role in signaling pathways eminent to learning and memory formation (Rawat et al. 12841).

Although phosphorylation of tau is a standard PTM, in cases of pathology, hyperphosphorylation may contribute to the onset of neurodegenerative disease (Rawat et al. 12841). There are 40 phosphorylation sites on the protein tau that are associated with disease, all with their own unique functions (Kimura et al.). Specifically, hyperphosphorylated tau loses its ability to bind and stabilize microtubules effectively, thus leading to impaired dynamics of microtubules. As a result, free tau molecules may aggregate and form neurofibrillary tangles (NFTs) (Gong and Iqbal 2321–28). When NFT aggregates accumulate in neurons, various cellular functions are disrupted. In a broad sense, hyperphosphorylated tau may accumulate in both dendrites and synaptic terminals, interfering with neurotransmission (Rajmohan and Reddy 975–99). Recent studies have also shown that this all may contribute to neuroinflammation, including activation of microglia and astrocytes, and the propagation of tau spreading in a prion-like manner (Amro et al. 100242). In a broad sense, NFTs result in physiological impairments, apoptosis, and neuronal loss reflected as cognitive impairment. While the specifics of these characteristics differ in various tauopathies, these are the mechanisms contributing to tau pathology and the onset of neurodegenerative diseases (Sinsky et al. 9207). In a broad sense, neurodegenerative disorders are characterized by damaged neurons and a loss of neuronal connectivity, thus dramatically compromising affected individuals’ execution of daily tasks, often termed as dementia (Cunningham et al. 79–87). Table 1 below outlines the significant characteristics of CTE and AD, respectively.

Characteristic	CTE	AD
Isoform	Tau protein (3R and 4R)	Tau protein (3R and 4R), Beta-amyloid
Brain Region	Frontotemporal lobes, amygdala, hippocampus	Hippocampus, cerebral cortex
Cause	TBI and repetitive head trauma	Unknown

Phosphorylation Sites	Ser202, Thr205, Ser396, Ser404	Ser202, Thr205, Ser396, Ser404
Symptoms	Memory loss, confusion, impaired judgment, aggression, abnormal mood changes, progressive dementia	Memory loss, confusion, communication difficulties, abnormal mood changes, progressive dementia

Table 1: Comparative table detailing characteristics of Chronic Traumatic Encephalopathy, Alzheimer’s Disease, and Progressive Supranuclear Palsy. Table compares significant isoforms, affected brain regions, known causes of disease, tau phosphorylation sites, and associated symptoms.

Alzheimer’s Disease (AD)

Currently, over 55 million people worldwide are living with dementia, most of these individuals having been diagnosed with AD. Further, it has been projected that by 2050, this number will continue to increase to around 152 million cases globally (Alzheimer’s Disease International). This information is imperative because dementia, most frequently caused by AD, is currently the seventh leading cause of death, and a significant concern associated with aging (National Institute on Aging). Specifically, according to the Alzheimer’s Association, in the United States alone, it is estimated that 6.9 million Americans currently have AD (Alzheimers & Dementia). AD is defined as a progressive neurodegenerative dementia that impairs memory, thinking, and behavior, typically becoming severe enough to interfere with daily tasks (Kumar et al.).

I. Clinical Presentation

The clinical aspects of dementia that are associated with AD as a neurodegenerative disease are typically categorized into three stages of dementia: mild, moderate, or severe (Kumar et al.). The mild stage is associated with declined execution of functions, slight personality and behavioral alterations, altered sensory perception, and finally a loss in episodic, semantic, and implicit memories (Dickerson and Eichenbaum 86–104). The moderate stage of this progressive disorder entails neuropsychiatric loss of reading and writing skills, incorrect word substitution, aggression, irritability, delusion, and loss of long term memory. Severe stages are characterized by almost a complete loss of speech, a lack of emotion, intense fatigue, and a decline in muscle mass (Siddappaji and Gopal 357–89). Some additional characteristics of AD are psychiatric symptoms such as depression, anxiety, and hallucinations that can occur through any of these stages (Dickerson and Eichenbaum 86–104).

II. Neuropathology

As a primary precursor to AD it is necessary to understand its associated A β pathology. AD is typically characterized by two major molecular alterations: extra amyloid-beta (A β) deposits and interneuronal filaments of the tau protein (Gulisano et al. 611–31). This A β is formed through processing of the membrane protein known as amyloid β protein precursor (APP) (Bamford and Oguro-Ando 184–202). Next, β -secretase is able to shed the ectodomain of APP, which leads to the cleavage of the protein precursor (Hartmann). As a result, extracellular deposits of amyloid-beta peptides form, and are typically prone to aggregation. Further, dominant missense mutations of the APP gene may contribute as early-onset cues, ultimately leading to the formation of longer, more aggregation prone forms of the protein (Itoh et al. 3139–51).

Further evidence supporting the significance of A β aggregation in AD pathogenesis comes from various studies using mouse models. In AD, A β plaques are distributed in different regions of the brain, correlating to the stages of the disease (Hampel et al.). Through mouse models, it was noticed that initially, plaques are deposited within the neocortex, which is highly responsible for higher order brain functions such as sensory perception and cognition (De Sousa et al.). As the disease progresses, plaques may be spread to the hippocampus and other parts of the brain depending on the severity of the disease (Targa Dias Anastacio et al.). While these plaques can be seen in various other conditions, it is apparent that this pattern of prevalence in these particular two brain regions are specific to AD (Hampel et al.).

Following the formation and aggregation of amyloid β protein and its associated plaques, tau plays a subsequent role. More specifically, the aggregation of the amyloid β protein has been observed to induce further pathological changes associated with tau such as hyperphosphorylation and its aggregation (Gulisano et al. 611–31). In AD, when tau is hyperphosphorylated as a response to A β aggregation, intracellular aggregates may form paired helical filaments (PHF), which are also components that are unique to AD (Rawat et al. 12841). When tau detaches from microtubules it forms the tangles known as NFTs. While this is a characteristic of prominent tauopathies, in AD specifically, the patterns of NFTs are a characteristic specific to AD. Additionally, another component of AD pathology that differentiates AD from other tauopathies is that it follows a distinct pattern of NFT spread (Sinsky et al. 9207).

Specifically, this unique pattern of distribution known as Braak staging, characterizes different progressive stages of Alzheimer's disease based on the NFTs and their associated spread into the memory circuit (Nelson et al. 1–14). Stages I and II are limited to transentorhinal regions of the brain, whereas stages III and IV differentiate involvement in the limbic regions such as the hippocampus. Finally, stages V and VI characterize neocortical involvement with tau NFTs. These stages are significant to AD pathology, because they are currently the most tell-tale sign of AD pathology that can be diagnosed in autopsy (Braak et al. 389–404).

III. Tau Specific Characteristics of AD

In various tauopathies, a differentiating characteristic of their unique disease pathology is the distribution of specific isoforms (Cherry et al.). However, in AD, all six isoforms are present in PHFs and NFTs, but the relative amount of specific isoforms and the modifications that they undergo may differ depending on the state of the disease's pathology (Kolarova et al. 1–13). Specifically in AD, all isoforms of tau are hyperphosphorylated, reducing their affinity for microtubules. This also accounts for the impaired neurotransmission, memory retention, and formation (Rawat et al. 12841). Furthermore, while in some neurodegenerative diseases such as progressive supranuclear palsy, there is an observed difference between the ratios of the 3R and 4R isoforms, in AD, both isoforms aggregate to form the neurofibrillary tangles (Buchholz and Zempel).

Although the ratio of 4R and 3R isoforms are less significant to AD than other neurodegenerative diseases, it can still serve as an important marker of the disease's pathology (Holper et al. 7307–7). With the proper associated technology, changes in the ratios of tau isoforms or modifications occurring post-translationally to the protein tau have recently been observed to be a promising biomarker for AD and other similar diseases (Bellier et al. 1573–85). These observed changes in cerebrospinal fluid or blood can serve as biomarkers for AD diagnosis and tracking progression (Salvado et al.). Thus, understanding the molecular mechanisms of various neurodegenerative diseases and their associated pathological characteristics is a promising development aimed towards developing targeted therapies and potentially counteracting disease progression (Zhong et al.).

IV. PTMs and Cellular Response

Observing the molecular mechanisms, it is apparent that PTMs of tau isoforms are crucial to the progression and pathogenesis of AD (Guo et al.). Knowing that all isoforms of tau in AD are hyperphosphorylated, it is important to recognize phosphorylation as a PTM. In AD, phosphorylation occurs at polar amino acid sites such as threonine, serine, and tyrosine residues. More specifically, the sites include Ser202, Thr205, Ser396, Ser404, and Ser422 (Kimura et al. 179). Responsible for phosphorylation, the enzymes glycogen synthase kinase-3 β (GSK-3 β), cyclin-dependent kinase (CDK5), and microtubule affinity-regulating kinase (MARK) are prominent PTM regulators in AD pathogenesis. For example, GSK-3 β is a major kinase that is responsible for phosphorylating tau at numerous of its associated sites (Fukiyama et al. 6941–57). Overactivation of this enzyme specifically is associated with abnormal tau phosphorylation. Since the activity of GSK-3 β is regulated by phosphorylation, it is inhibited by phosphorylation at Ser9, yet activated at Tyr216 (Sayas and Avila 721). CDK5 is typically regulated by its regulatory subunit p25. When p25 accumulates, tau is abnormally phosphorylated and contributes to the hyperactivation of CDK5 (Kimura et al.). Mark kinases specifically affect microtubule dynamics by phosphorylating sites that are critical for proper microtubule binding (Timm et al.). However, again, the increased activity of this kinase

contributes to hyperphosphorylation, and ultimately disrupts microtubule stability, therefore mechanisms of neuronal transport (Gong and Iqbal 2321–28).

Aside from hyperphosphorylation, two other PTM that are often associated with AD are acetylation and glycosylation (Zhong et al.). In acetylation, histone acetyltransferases (HATs) are responsible for adding an acetyl group to the lysine residue of a protein. This PTM is known to cause the loss of function of tau protein (Park et al.). Furthermore, acetylated tau is more prone to aggregation in the form of paired helical filaments and NFTs (Cohen et al.). Because increased levels of acetylated tau are prominently associated with neurotoxicity and cognitive deficits, interestingly, modulations of the mechanisms that both activate and inhibit acetyltransferases are being explored as a therapeutic strategy for AD (Min et al. 1154–62). Next, the glycosylation addition of sugar molecules to N-linked amino acid terminals affect the solubility of tau, influencing its ability to form aggregates (Alquezar et al.). Therefore, the combined effects of these enzymes are critical in AD pathogenesis through promoting the aggregation of hyperphosphorylated tau isoforms, ultimately contributing to cellular and cognitive decline.

Chronic Traumatic Encephalopathy (CTE)

Chronic Traumatic Encephalopathy (CTE) is characterized as a progressive neurodegenerative disease resulting from repeated head trauma (Inserra and DeVrieze). The most common groups diagnosed with CTE are athletes, most commonly American football players (Safinia et al. 34–48). According to post-mortem studies of American football players' brains, researchers identified signs of CTE in 87% of the 202 subjects studied (Boston University). This finding is pertinent because CTE is associated with concussion and other brain injuries that fall under the classification of traumatic brain injury (TBI) (Graham and Sharp 1221–33).

I. Clinical Presentations

The clinical presentation of CTE involves a variety of cognitive, behavioral, and motor symptoms. CTE is a disease characterized by neuronal death, therefore these symptoms ultimately contribute to a gradual cognitive decline (Gandy et al. 37). As CTE continues to worsen over time, increased neuronal death contributes to the shortened life spans of affected individuals (Lakhan et al.). Individuals with CTE may experience memory impairment that is characterized by issues with short term memory impairment. However, due to the disease's progressive nature, these may progress to more severe deficits. Other cognitive symptoms may be classified as executive dysfunctions that impair problem solving and attention deficits compromising the retention of focus in affected individuals (McKee et al. 350–64).

Another common presentation of CTE are mood disorders and physical symptoms. In addition to behavioral changes, affected individuals may experience depression, apathy, emotional instability, and aggression. Furthermore, behavioral changes may also be characterized as difficulties with impulsivity, irritability, and potentially socially inappropriate behavior depending on the severity of the disease's pathology (Antonius et al. 313–22). Individuals typically experience symptoms such as tremors, rigidity, and bradykinesia (slowed movements).

Problems with balance, coordination leading to unsteadiness may also be prominent in CTE cases (Montenigro et al. 304–17).

II. Neuropathology

In accordance with the ionic imbalance experienced as a primary response of TBI, the sudden increase of calcium ions within neurons activates various calcium dependent kinases, which catalyze the phosphorylation of various proteins (Atkins et al. 1507–18). The specific kinase pertinent to the onset of CTE pathology is the calcium dependent kinase known as Glycogen Synthase Kinase-3 β (GSK-3 β). GSK-3 β is directly responsible for catalyzing the phosphorylation specifically of the protein tau (Fukiyama et al. 6941–57). Researchers have concluded that the molecular modifications of the protein tau are one of the major prominent pathological characteristics of the onset of CTE (Halicki et al. 12556–56).

The pathology of CTE is often described through four stages, known for their progressive symptoms (Inserra and DeVrieze). Interestingly, many parallels may be drawn between the stages of AD and CTE. Stage I of CTE is characterized by mild cognitive impairment of memory and attention span in addition to slight behavioral changes. Stage II is classified through more extensive pathology in the frontal cortex (Castellani et al. 493–99). The tau pathology tends to spread in a prion-like manner, spreading to the superficial layers of the cortex (Alyenbaawi et al. 1487). Stage III is characterized as widespread tau pathology throughout the entire frontal-temporal cortices. This includes the hippocampus, amygdala, and entorhinal cortex. Stage IV is defined as severe tau pathology that affects the majority of neural regions spanning to the brainstem and spinal cord. Furthermore, extensive cortical atrophy and white matter changes are evident (McKee et al. 350–64). Focusing on the molecular response of TBIs, concussions contribute to a complex molecular runaway inflammatory cascade upon initial injury (Tonny et al.). To simplify this cascade, the first responses that typically occur immediately following TBI are mechanical damage and cellular disruption.

III. Mechanical Damage and Cellular Disruption from TBI

Concussive hits automatically lead to axonal shearing, a process by which axons are shredded and torn. This results in the immediate compromise of the structural integrity of the neuron, therefore impairing efficient neurotransmission (Bruggeman et al. 31–44). Additionally, these injuries may also immediately disrupt the cell membrane, contributing to an immediate ionic imbalance between calcium and potassium ebbing through the neuron. This imbalance also affects the ability of the neuron to generate an action potential, or in other words, the electrical conduction of a neural signal is compromised (Grider et al.).

These early molecular mechanisms serve as primary responses to TBI, which then initiate an additional signaling cascade of secondary responses including the disruption of the blood-brain barrier, and the release of neurotoxic substances. In its normal function, the blood-brain barrier (BBB) is a semi-permeable vascular membrane that tightly regulates molecular movement between the blood and the brain (Chodobski et al. 492–516). When this

system is disrupted as a result of TBI, the permeability of the BBB increases, which consequently impairs the barrier's ability to protect the brain's central neurons. This consequence of TBI potentially increases the possibility of neurotoxic substances entering the brain without regulation (Archie et al. 1779). Furthermore, upon injury, the primary response of an ionic imbalance triggers an unnecessary release of glutamate in the brain, which ultimately triggers apoptosis (Krishnamurthy and Laskowitz). Thus, due to disruption of the BBB, the brain is more prone to the cell death that may result from the random influx of glutamate in the brain (Boyko et al. 5897).

In accordance with the ionic imbalance experienced as a primary response of TBI, the sudden increase of calcium ions within neurons activates various calcium dependent kinases, which catalyze the phosphorylation of various proteins (Atkins et al. 1507–18). The specific kinase pertinent to the onset of CTE pathology is the calcium dependent kinase known as Glycogen Synthase Kinase-3 β (GSK-3 β). GSK-3 β is directly responsible for catalyzing the phosphorylation specifically of the protein tau (Rawat et al. 12841). Researchers have concluded that the molecular modifications of the protein tau are the most prominent pathological characteristics of the onset of CTE (Cherry et al.).

IV. PTMs and Isoforms

In CTE, there is an observed predominance of 4R tau isoforms. While both 4R and 3R isoforms undergo hyperphosphorylation, the predominance of 4R isoforms is a significant classification of CTE specifically (Cherry et al.). The NFTs formed in CTE additionally are composed of 4R and 3R isoforms, typically found around small blood vessels and at the depths of subpial regions (McKee et al. 350–64). The accumulation of these isoforms and 4R predominance is responsible for contributing to microtubule destabilization and cellular toxicity to both neurons and glial cells (Mietelska-Porowska et al. 4671–713).

Upon the initial translation of the protein tau, it must undergo specific post translational modifications (PTMs) (Haj-Yahya and Lashuel 6611–21). One specific PTM of the protein tau is phosphorylation. When GSK-3 β is activated, tau protein may be phosphorylated at many different sites all with different regulatory functions (Cheng et al. 1650–70). However, when GSK-3 β is overactivated by the excessive amount of calcium ions absorbed as a result of TBI, tau protein may become hyperphosphorylated at multiple sites, counteracting the regulatory nature of the PTM in its normal state (Bartolome et al. 1421). Repeated TBI continues the overactivation of GSK-3 β , therefore continuing the cycle of hyperphosphorylated tau being produced as a result of the PTM (Cheng et al. 1650–70).

Aside from the abnormal PTMs of the protein tau, another common molecular change resulting from repeated TBI is neuroinflammation. When the brain initially experiences trauma, microglia and astrocytes are released as an initial immune response (Davidson et al. 8816–39). When GSK-3 β is activated, tau protein may be phosphorylated at many different sites all with different regulatory functions (Sayas and Avila 721). However, when GSK-3 β is overactivated by the excessive amount of calcium ions absorbed as a result of TBI, tau protein may become

hyperphosphorylated at multiple sites, counteracting the regulatory nature of the PTM in its normal state (Bartolome et al. 1421). Repeated TBI continues the overactivation of GSK-3 β , therefore continuing the cycle of hyperphosphorylated tau being produced as a result of the PTM (Sayas and Avila 721).

When tau is hyperphosphorylated, it detaches from microtubules and forms abnormal tau aggregates known as neurofibrillary tangles (NFTs) (Gong and Iqbal 2321–28). Hyperphosphorylation as a PTM negatively affects both the normal functioning of the protein tau and proper functioning of the neuron itself (Zhong et al.). When tau detaches from microtubules, it is unable to maintain the structural integrity of the neuron, therefore impairing axonal transmission. As a result, the protein tau begins to aggregate in the neuron (Robbins et al.). These abnormal aggregates are also known as NFTs, which continuously impairs intracellular transport to the point of cell death. This process is only further reinforced and repeated through further TBI (Avila). Thus, this process perpetuates a neurodegenerative cycle of neuroinflammation and abnormal cellular responses.

Conclusion

This research compared the molecular mechanisms of Alzheimer's Disease and Chronic Traumatic Encephalopathy. While these neurodegenerative tauopathies are seemingly different in both onset and some aspects of clinical presentation, it is crucial to understand the molecular mechanism governing these diseases. Identifying similarities in these two disparate diseases provides further insight into understanding and treating tauopathies on a larger scale, through identifying fundamental pathological characteristics. Thus, potential therapeutic targets for tau pathology in Alzheimer's Disease and Chronic Traumatic Encephalopathy include inhibiting kinases like GSK-3 β and CDK5 in order to prevent tau hyperphosphorylation. Research into the logistics of this implementation should be conducted in order to further inform this prediction. Additionally, using small molecules and antibodies to block tau aggregation, while an ambitious therapeutic goal, could prove promising in future treatments of tauopathies. Finally, modulating neuroinflammation with anti-inflammatory or immune-modulating therapies among disease onset or early symptoms may prove as a target for treating tauopathies in the future.

Understanding the molecular mechanisms of Alzheimer's Disease and Chronic Traumatic Encephalopathy—specifically associated with abnormal tau aggregation and cellular response—holds significant promise for advancing knowledge of neurodegenerative diseases. As done in this review, identifying the unifying molecular characteristics of these tauopathies can lead to the development of broad preventative measures, offering a proactive approach to reducing disease incidence. Thus, future research should expand upon these principles. However, identifying the differences between the molecular features of each tauopathy may aid in facilitating earlier and more accurate diagnoses. Therefore, while it is important to identify molecular similarities between tauopathies, future research should also investigate the differences in order to derive treatment methods for specific tauopathies.

However, identifying the differences between the molecular features of each tauopathy may aid in facilitating earlier and more accurate diagnoses. While research has previously been conducted following this approach, emphasis in future and continual research should be placed on exploring the differences between tauopathies in order to begin identifying distinct treatment methods for distinct and individual tauopathies.

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How the Loss of Religious Piety and a Social Revolution of the Lower Class Accumulated to Economic Growth in the Post Industrial Era By Junyang Zheng

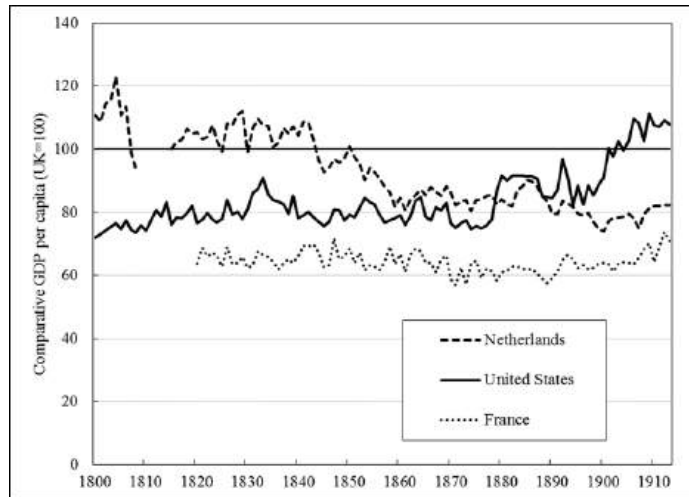
After centuries of stagnation, a period of sustained economic development followed the commencement of the later years of the 18th century. Whilst the growth could be directly linked to the rise of industry in the age of industrialization, it is rather its social side effects that proved most consequential in the rapid development of economic power. Through a combination of growing class struggle and gradual erosion of traditional values, the economic order of pre-industrial Europe became increasingly undermined by a revolutionary new wave of modernism contributing to economic growth. In his book, Charles A Beard remarked that “the Industrial Revolution has two phases: one material, the other social; one concerning the making of things, the other concerning the making of men.”¹ While the rise of machines nursed the beginning of an economic boom, this essay analyzes the fundamental changes to the humane society that more directly contributed to an era of unprecedented growth in the economics of western civilizations in the immediate aftermath of the industrial revolution.

Kicked started by the revolutionary mechanics of James Watt’s steam engine at the height of 18th century England, a plethora of extraordinary feats of human ingenuity transformed the British Isles into a land of industry. From the ashes of the declining agrarian economy rose an emerging class of industrialists vying for prestige and wealth. Aided by the Laissez-faire policies of the British Parliament, the rising power of the bourgeoisie began to challenge the traditional power base of hereditary aristocracy in British society. The prevalent concept of Noblesse Oblige among the nobility represented a genuine contemptment of the British upper class in regards to monetary transactions and its subsequent consequence of economic growth. As characterized by historian John O. McGinnis, the aristocrats of the 17th century, though recognizing the importance of businessmen, viewed them as “money grubbers who lacked (an) appreciation for the higher things in life.”² McGinnis’s characterization is also evident in the broader scope of the European continent, with France in particular remaining influenced by pious factions opposed to usury, a key component for the workings of a functional banking system.³ In a sense, the social rigidity and piety of pre-industrial Europe hindered the explosion of economic growth made possible by the emerging bourgeois class who lacked the same appreciation for traditional norms as their more conservative aristocratic counterparts.

It could be argued from a certain perspective that the era of industrialization instituted a trend of growing moral decay in correlation with the faltering significance of the traditional gentlemen. Perhaps most vividly described in Fitzgerald’s *Great Gatsby* of the rise of gilded age materialism in post-industrialized America, this negatively perceived change in social dynamics nevertheless spurred positive growth in the economic sector. Religious opposition from the Catholic and sects of Protestant churches spearheaded the conservatives, crediting from the New Testament which preached that “the love of money is the root of all evil”⁴. In the Netherlands, however, the Calvinist faith fostered the beginning of a capitalist boom stemming from Max Weber’s theory of the protestant work spirit, while the economies of Catholic western Europe

stagnated. Well the prosperity of the Dutch could not be attributed solely to religion, it offers an interesting case study. The shifting of trade routes in the early 17th century allowed for a blossom of banking and trading power that destabilized the religious authority of the Dutch social conservatives. In the face of the rising faction, religion gave grounds. The Dordrecht synod of 1574 that advised against participation of businessmen in religious ceremonies translated to its subsequent overruling in the declaration of the Dutch estates in 1658, where it is decreed that “no church had the right to deprive any banker of participation in the communion service because he was a banker.”⁵ The economic prosperity of the growing merchant class meant growing opposition to the clergy, a trend amplified by the industrial revolution. The profit of industrialization led to open government sponsorship, a shift from the policies of pre-industrial Europe aiming at the appeasement of the clergy. The success of industry further marked the rise of secularization across all classes of society, a trend observable from an analysis of the literature of the age⁶. A reason for such development could be found in the faltering significance of events deemed divine will. For agrarian Europe, much economic profit derives from agricultural labor, which in turn is dependent on the weather for which men held little control. The people turn to religion in prayers for good weather and a plentiful harvest in hopes of economic prosperity. The industrial world is much different, with the primary method for economic gain rooting in logistics and capitalist investments. Indeed, the scientific growth of the age left religion scrambling for a place in the hearts of the common man, a trend that correlated with economic growth in the American experiment across the Atlantic.

Much like their counterparts across the Atlantic, the traditionalist puritan societies of Massachusetts gradually evolved to the glamor of wealth in New York and Boston. The gilded age, as labeled by author Mark Twain, glorified the very concept of wealth in the age of capital materialism. In his work “Democracy in America”, French writer Alexis de Tocqueville criticized the superficial nature of gilded age American society, writing: “I know of no country, indeed, where the love of money has taken stronger hold on the affections of men⁷.” Yet, despite Tocqueville’s perceived moral high ground over the Americans, the gilded age’s frank glorification of wealth correlated with a decade of American economic prosperity and growth, overtaking traditional European economic heavyweights of the French and the Dutch in the decades following 1880⁸.



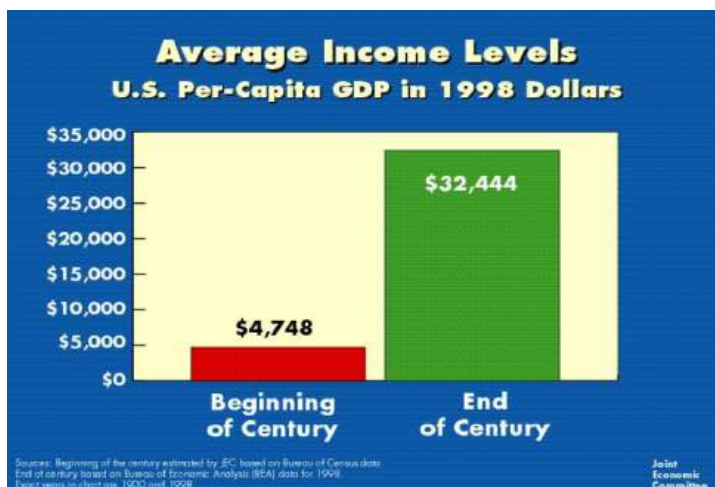
(Figure 1)

In the decades since the civil war, American industry exploded from the fractured ruins of war to the forefront of international economics at the dawn of the 20th century. The declining influence of the quasi-aristocratic American gentry made way for the rise of industrialists and an increased obsession with wealth⁹. Indeed, Tocqueville’s criticism is well founded. At the height of gilded age, wealth became a socially accepted form of status, and religion was forced to conform with the episcopalian Bishop William Lawrence even concluding that “Godliness is in league with the riches”¹⁰. The blunt pursuit of wealth in America governed by little concern for morality became a case of proven economic success, and the American model lured Europeans to follow in varying degrees¹¹. Industrialization is an era of economic growth no doubt, but it is also a period of growth for all aspects of society. The improved healthcare and infrastructure meant the sick no longer reverently prayed to god for a miracle but rather fervently ran for a doctor. In many ways, the religious hold of pre-industrial Europe stemmed from fear¹², but recent advances replaced god with man as the workers of miracle. In response, religion evolved, taking a backseat with a marked rise in atheism and the acceptance of more modern values in its operation corresponding to an era of economic prosperity increasingly unrestricted by religious taboos^{13 14}.

As industrialists rose to positions of power left vacant by the declining clergy, they became increasingly entangled in a class conflict with the struggling poor¹⁵. Despite the idealist transcendentalism of Emerson and a few proven cases of success, the position of the lower class remains very much unchanged in the centuries since the invention of the steam engine. What has changed, however, is who now holds the whip. Indeed, in the words of Irish playwright Oscar Wilde, the new age of industry has ushered in “the slavery of the machine” while the misery of the poor remains¹⁶. Another building block for the economic explosion of the 18th and 19th century came as a continuation of the historical class struggles that plagued the history books for centuries, only this time the struggle saw the emergence of a new ideology in a changing historical context vying for a tangible social revolution.

Here we must draw a distinction between the socialism of Wilde and Marx with socialist movements of Western Europe and America. While its systematic implication saw failure in the communist states in the east¹⁷, the growth of social unionism saw significant progress in the economic situation of the poor¹⁸. The reactionary response to the rise of industry saw a marked rise in the voice of the lower class, with the emergence of the Labor party in Britain and the progressive movement in America breaching national politics with agendas aligning outside of the ruling minority. The historical context of the new world differs greatly from pre-industrial Europe, where the economic and political spheres of the nation blended together in the form of a strong government. Protests in face of economic hardship are often viewed as an attack against the political power of the state and faced with military opposition, as seen by the prosecution of the hungary French starved of economic prosperity in the early stages of the French revolution. The industrialization process and the rise of capitalism separated industry from the ruling elite, and at the turn of the 19th century, unions and marches for economic betterment were met with cases of reluctant compromise and signs of genuine progress¹⁹. The economic revolution of the 18th century effectively gave the proletariat more potential for change with the shift of industrialists overtaking government agents in the running of the economy, with reduced power for prosecution compared to centuries prior.

The emergence and success of labor and progressivism correlated with a marked increase in the economic situation of the general population. The minimal wage and a reduction of work hours allowed for leisure time and consumer spending that boosted the economy, contributing to the establishment of a consumer culture aided by the steady growth of the middle class. In short, the pressure of the counter-reaction to the economic hardship of the industrial working class forced a change in the socio-economic system of the west untouched before the 18th century. The resulting impact could be seen with an analysis of economic data from America, with a noticeable increase in both economic potential and general standard of living in the decades following the explosion of the socialist progressive era²⁰.



(Figure 2)

Work Time Needed for the Average Worker to Buy Selected Products

Product	Beginning of Century	End of Century
Half gallon of milk	56 minutes	7 minutes
One-pound loaf of bread	16 minutes	3.5 minutes
Hershey chocolate bar	20 minutes	2.1 minutes
Three-pound chicken	2 hours 40 minutes	14 minutes
Pair of Levis jeans	9 hours 42 minutes	3 hours 24 minutes
100 kilowatt hours of electricity	107 hours 17 minutes	38 minutes
3 minute coast-to-coast phone call	90 hours 40 minutes	2 minutes

Source: Myths of Rich and Poor, Michael Cox and Richard Alm, 1999.
Items are for various years near the beginning and end of the century.

Joint Economic Committee

(Figure 3)

In a more economic analysis, the increase in household hold income leads to a corresponding raise in consumer spending, as postulated in by the keynesian consumption function (figure 4), where a direct relationship is derived between real disposable income and total consumer consumption.

$$C = A + MD$$

where:

C = consumer spending

A = autonomous consumption

M = marginal propensity to consume

D = real disposable income

Figure 4²¹

Basing our argument on the basic concept of supply and demand, the increase in spending power correlates to growth in aggregate demand, contributing to uniform growth across all sectors of the economy²². The effect of the labor unions, though more impactful in some places than others, paved the way for the modern economics of the 20th century. While the industrial revolution spurred economic growth at its infancy, its subsequent social upheaval is perhaps

more significant for its role in facilitating the unprecedented economic growth of the modern age in the years following the end of the 18th century.

While the industrial machines facilitate the beginning of an economic boom, it's rather the social movements from which it spawned that developed the modern economic landscape, establishing the basis for the economic boom and rampant growth of the middle class after the end of the 18th century. The erosion of the old moral standards of the nobility with the rise of capitalist pursuit of wealth led to the rise of both industrialists and a countermovement of social unionism in response. The ensuing class struggle allowed for greater access to wealth and leisure spending for an expanding middle class, accumulating in the creation of a rapidly growing modern economy unimaginable in the stagnant economics of pre-industrial Europe.

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The Effects of Gang Presence and President Bukele's Mano Dura Policy on the Salvadoran Economy By Mara Ungureanu

Abstract

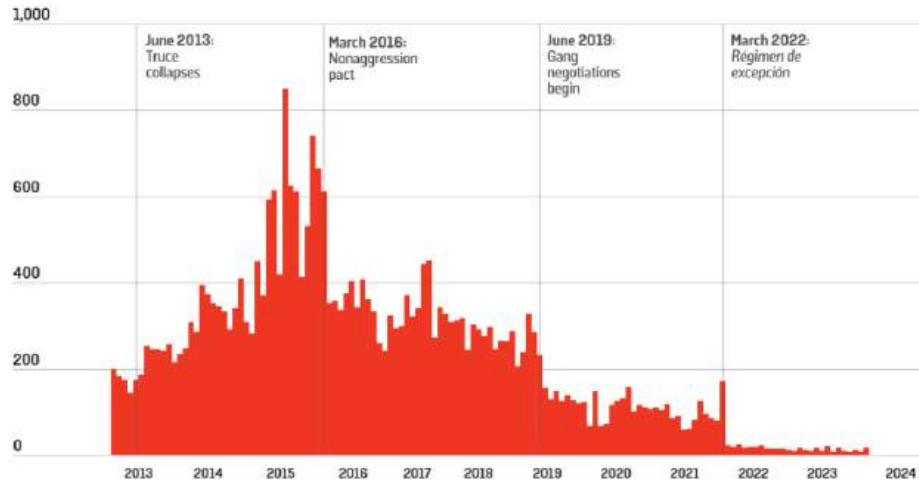
After decades of oppressive gang violence, El Salvador has entered a new era under President Nayib Bukele, who has implemented his unique version of mano dura government policy to eradicate the gangs and bring about leaps in public safety. These improvements have come at a cost; Bukele's solution has involved the suspension of several constitutional rights, human rights violations, and questionable uses of power.

This paper analyzes and orders by magnitude the economic effects of both past gang rule and current mano dura policy using the framework of the Cobb-Douglas production function. I conclude that gangs had an overwhelmingly negative effect mostly because of extortion practices and decreased business confidence and sentiment, despite the minor benefits of informal employment by gangs. Secondary negative effects were also exerted by blows to education and increases in violence-fueled outward migration. My conclusion in regard to Bukele's policy is more nuanced; positive effects – increases in public safety, boosts to business participation and confidence – are mostly short-run, and its negative effects – declines in education, deteriorating rule of law – are exerted over the long run. Therefore, Bukele's mano dura may disincite El Salvador away from long-term growth if mitigating action is not taken soon. At the conclusion of the paper, I provide policy suggestions, namely increased focus on addressing the root causes of gang participation with investment in education and job programs coupled with increased fairness and democracy within government and law enforcement.

Introduction

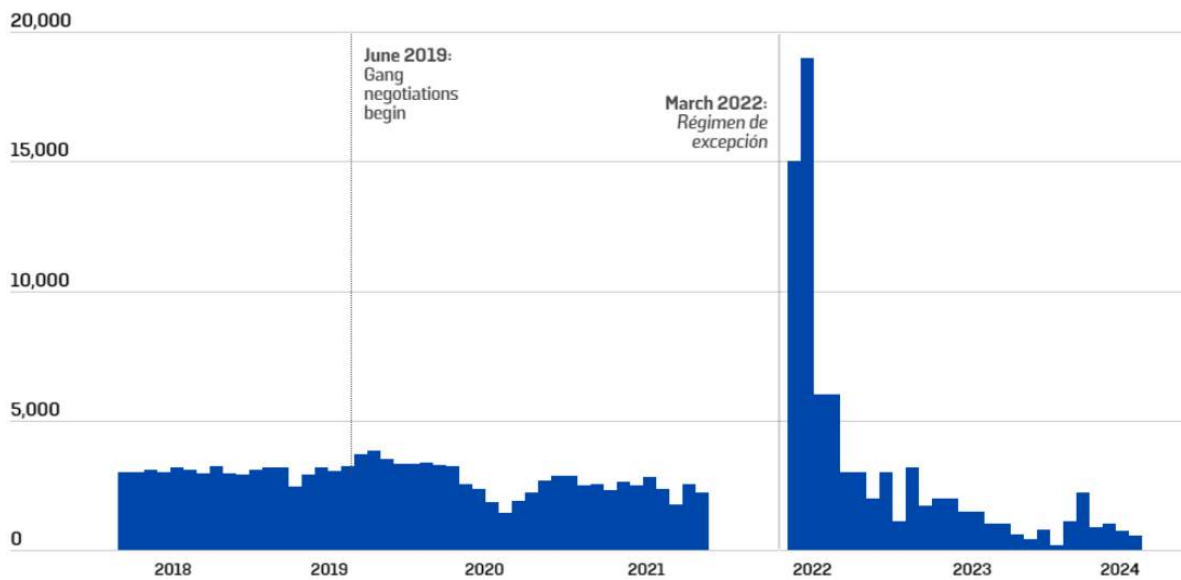
El Salvador was once known for having the highest murder rate in the world, shown at its peak of 103 homicides per 100,000 inhabitants in 2015 (Statista). El Salvador's two most prominent gangs – Mara Salvatrucha or MS-13, and Barrio 18 – were largely to blame. However, since the election of current Salvadoran president Nayib Bukele in 2019, the nation's homicide rate has plummeted, and its gang crisis has been declared over. The sudden decrease in homicides is shown in Figure 1. The country's transformation is the result of Bukele's vastly popular mano dura, or iron fist gang policy, which includes the régimen de excepción, or state of emergency, as a cornerstone. Since the policy's implementation, El Salvador's incarceration rate has skyrocketed to over 1,000 prisoners per 100,000 inhabitants, 2% of its adult population, steadily taking a new spot as one of the most incarcerated countries in the world (Statista) (World Prison Brief). El Salvador's official detentions are shown in Figure 2. Bukele's version of mano dura has provoked numerous alarm bells from humanitarian and watchdog agencies, as well as domestic protests.

Figure 1
Official Homicides by Month in El Salvador Since 2013



Note: Sourced from Foreign Policy. Note the drastic drop upon régimen de excepción, or state of emergency.

Figure 2
Official El Salvador Detentions by Month



Note: Sourced from Foreign Policy. Data missing from October 2021 to March 2022

Previous research has examined Salvadoran gang violence through an economic lens (Amaya), but not yet alongside Bukele’s ongoing gang policy. NGOs such as Amnesty International and Human Rights Watch have conducted in-depth investigations of Bukele’s policy and drawn conclusions about its human rights implications, but not its economic impact. The recent nature of

Bukele's groundbreaking policy explains the lack of economic literature analyzing it; this paper seeks to draw from economic research on mass incarceration in the United States, the effects of a strong rule of law, and other relevant topics to draw conclusions about Bukele's *mano dura*, a novel event.

Bukele's actions, and the widespread gang violence that preceded it, touch the lives of every inhabitant of El Salvador, as well as many Salvadorans living outside its borders. In 2018, International Crisis Group (ICG) reported that "every Salvadoran knows someone who was shot dead by gang members or someone who pulled the trigger." The overwhelming majority of Salvadorans living in the United States during years of high gang violence in El Salvador sent money home to affected family members experiencing poverty exacerbated by extortion practices. How these remittances from family members in the United States are spent, and how badly they are needed, is thoroughly intertwined with the threat of gang activity in El Salvador – mainly, extortion. Apart from Salvadorans, this issue is relevant to all countries in the Northern Triangle; Honduras and Guatemala have been similarly impacted by intense gang violence, and now after Bukele's proclaimed success, his *mano dura* model is being heavily imitated by President Xiomara Castro of Honduras, among other neighboring leaders, meaning the model's economic consequences will take place not only in El Salvador, but in the greater Northern Triangle region.

Speaking more broadly and in ideological terms, this issue's relevance extends even beyond the Northern Triangle to encompass the globe. Many have described the recent transformation in El Salvador as a trade – giving up civil liberties in exchange for safety – and one that has been met with incredibly popular approval (McArdle). This 'trade' poses a question to democracies everywhere about how much they might be willing to sacrifice, and what constitutes democratic consent. Martin Luther King, Jr. wrote in his Letter from Birmingham Jail that "injustice anywhere is a threat to justice everywhere" to highlight the interconnectedness of society regarding socioeconomic injustices. The economic consequences of current events in El Salvador will exert a ripple effect extending far beyond terms of trade and foreign exchange; they have the potential to change the way societies around the world view the trade-offs between fairness and order.

This paper seeks to investigate both beneficial and detrimental economic effects of El Salvador's historic gang presence and its more recent anti-gang policy under President Nayib Bukele. The first section will focus on El Salvador's historical gang violence, first providing background and then diving into an economic analysis. The second section will explore Bukele's *mano dura* policy, outlining his actions thus far and then examining their economic implications. This paper will seek to provide policy suggestions aimed at maximizing Salvadoran welfare. Economic analysis will be conducted through the lens of the Cobb-Douglas production function.

Background on Gang Presence

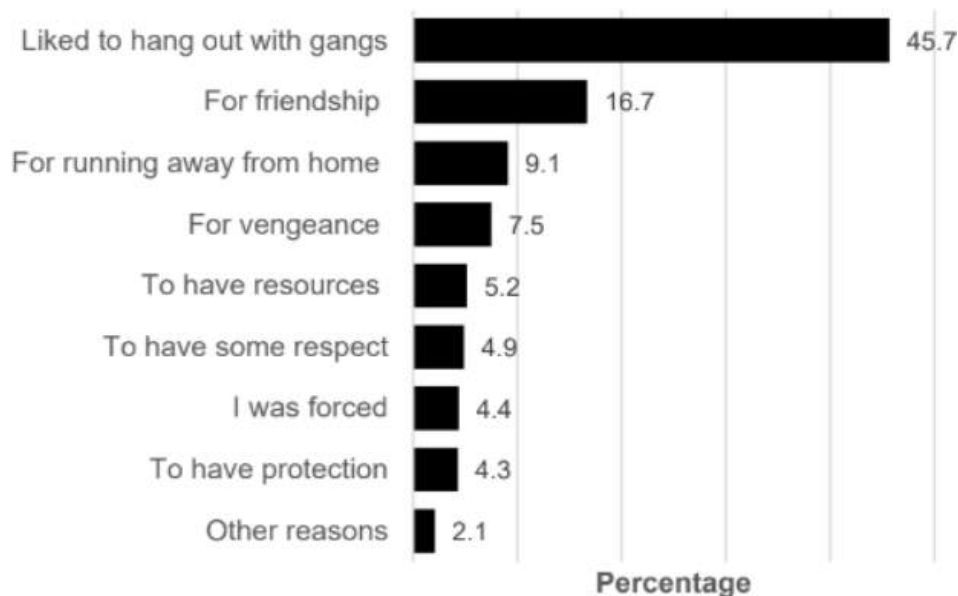
The two main gangs that operate in El Salvador are Mara Salvatrucha (MS-13) and Barrio 18. Both gangs are US exports from Los Angeles, California, and emerged during the 1980s. MS-13 was formed to defend marginalized and segregated Salvadoran communities, who had immigrated to the US during the Salvadoran Civil War of 1980-1992 and found themselves living in predominantly Mexican neighborhoods rampant with gang activity (Insight Crime) (Martinez). Barrio 18, on the

other hand, was formed by Mexican immigrants and then, upon witnessing an influx of Salvadoran, Guatemalan, and Honduran immigrants during a time of violence and civil war in the Northern Triangle, the gang began recruiting immigrants of various nationalities (Insight Crime).

So, how did MS-13 and Barrio 18 come to dominate El Salvador if they originated in the US? During President Bill Clinton's immigration crackdown in the 1990s, the passage of new policies such as the US Illegal Immigration Responsibility Act and the 1990 Immigration Act resulted in the deportation of more than 12,000 US residents with criminal records to El Salvador between the years of 1996 and 2002 (WOLA). Many deportees had little to no connection to their birth country, having immigrated at an early age. Furthermore, they arrived in a post-war El Salvador that was ill-equipped to handle a criminal influx and ill-informed of this influx by US authorities. The Chapultepec Peace Accords, which ended the Salvadoran Civil War, did little to relieve poverty, improve infrastructure, restructure society, or provide economic opportunity; they were primarily designed to finalize the war and reform politics (Martinez) (United Nations Institute of Peace). Thus, deportees reentering El Salvador during this time found themselves with few opportunities for a life outside of crime, and gang involvement skyrocketed alongside homicide rates.

The primary activity of both MS-13 and Barrio 18 in El Salvador is extortion, described as the "economic engine" behind both gangs and related violence. Targets range from informal vendors such as owners of streetside fruit stands to large enterprises such as bus lines; "la renta" touches everybody, and pushes many towards migration, business closure, and bankruptcy. In 2016, extortion payments totaled \$756M USD, equal to 3% of Salvadoran GDP, and it was estimated that over 70% of businesses were extorted in areas with gang presence (World Bank) (Martinez et al.). Despite the high levels of income both MS-13 and Barrio 18 receive from their extortion operations, it is vital to note that gang participation and participation in related extortion rackets does not pull gang members out of poverty. Instead, extortion activity creates a subsistence gang economy, where the money received is sent to gang leadership and then spent on the funerals of deceased gang members, gang member legal fees, paying bail, and similar expenses that fail to significantly improve the socioeconomic status of gang members. Generally, youth joining gangs are not seeking financial benefits or a lifeline out of poverty; they join to have pride in their identity and to feel like they belong to a group after growing up in broken families (ICG). A survey conducted by researchers at Florida International University in 2016, the results of which are shown in Figure 3, found that 62.4% of Salvadoran gang member respondents joined the gang either because they liked to hang out with other youth and gang members or because the gang provided them with close friends and a sense of brotherhood, with only 5.2% of respondents citing resources like money as their motivation for joining (Cruz et al.). Evidently, extortion operations do not leave either side economically better off, as gang members remain in poverty, and their victims are impoverished too, losing large chunks of their incomes. Everybody loses.

Figure 3
Reasons to Join the Gang According to 2016 Survey (in percentages)



Note: Sourced from *The New Face of Street Gangs: The Gang Phenomenon in El Salvador* by Jose Miguel Cruz et al.

When gang violence is severe and a gang’s grip on an area is strong, life becomes restricted and fearful. In many rural parts of El Salvador which had been “left behind” by the government, the gang was the only ruling power that remained and exerted full control over employment opportunities, business hours of operation, and similar factors. ICG reports, “Every Salvadoran knows someone who was shot dead by gang members or someone who pulled the trigger,” and many avoided public places and walks down the street (ICG). Streets were deserted after dark, and community soccer fields were the frequent sites of shootouts, so they went unused (Martinez). A homicide rate of 103 per 100,000 inhabitants in 2015 meant senseless killing, with women and children frequently caught in crossfire. Crossing the wrong street, refusing to be a gang member’s girlfriend, and having family visitors without asking permission from the gang were all reasons for which someone could be killed (ICG).

Gang Presence and El Salvador’s Economy

Economic analysis will be conducted through the lens of the Cobb-Douglas production function, $Y = A \cdot K^\alpha \cdot L^{1-\alpha}$, which decomposes the factors of production into labor (L) and capital (K). In addition, A is the total factor productivity (TFP), and alpha (α) is capital’s elasticity with

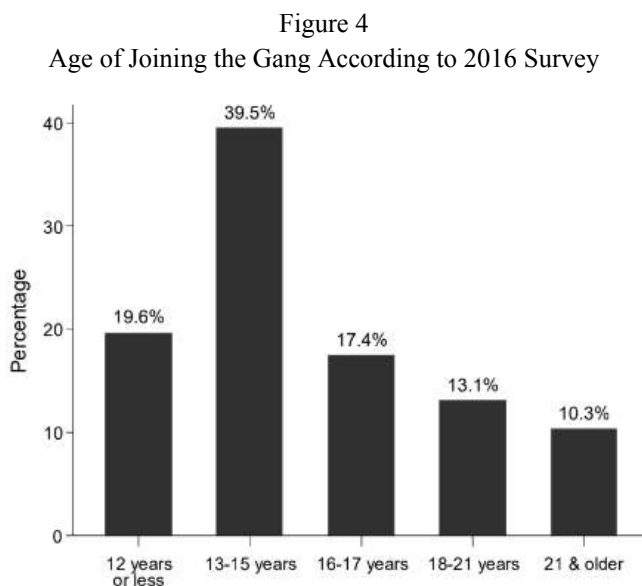
respect to output. The Cobb-Douglas production function is well-suited for use in this paper because its separation of factors allows us to determine the negative or positive contribution of gang presence or mano dura gang policy. Additionally, the function provides for an analysis that attempts to order positive and negative effects by magnitude and determine the net effect on Salvadoran production and welfare.

The most obvious negative economic effect of gang presence in El Salvador is the money that is lost by businesses large and small to extortion. Estimates place the percentage of businesses in the country that were being extorted in 2019 at 70%, with estimates of gang extortion revenues as high as \$31.2B USD (Zaidi). Daily extortion promises, or rentas, range from \$2 to \$3 for small businesses like fruit stands to \$5 to \$10 for medium-sized businesses and as high as \$20 for large businesses and distributors such as bus companies (Avelar) (Zaidi). Having to pay la renta to avoid certain death represents significant losses in wages for workers, and significant losses in profit and potential investment for business owners. Funds that could have been used to expand business operations – hiring more workers, buying new capital – instead go towards paying la renta. Indirectly, the fear and insecurity experienced by business owners because of extortion means they are less likely to take risks like investing in capital or expansion, even if they can afford to. Directly or indirectly, extortion by gangs negatively affects capital accumulation, decreasing the K factor in the Cobb-Douglas function. Capital accumulation has been repeatedly proven to be vital for economic growth because of its positive effects on productivity. Thus, the money lost to extortion by business owners exerts a large negative effect on Salvadoran Gross Domestic Product (GDP). On the worker's side, a business owner squeezed by la renta cannot pay him any more than a small wage and contributes to his perpetual poverty and financial insecurity. By preventing higher wages, extortion negatively impacts a worker's welfare and that of his family. Extortion also hinders job creation; businesses being extorted cannot afford to hire additional workers.

Perhaps a more abstract but still-significant negative economic effect of Salvadoran gang presence lies in the resulting decrease in business participation and business confidence. A strong gang presence such as the one experienced by El Salvador in previous years dampens business participation and confidence because, when gangs become as powerful as they were in El Salvador, they act as the dominant institutions, or powerholders in the country. This held true especially in poorer and more rural areas of El Salvador, where the state exercised little to no control and MS-13 and Barrio 18 readily filled the vacuum with their own presence, effectively taking over the role of law enforcement and local government in those areas. Even in urban areas, they rival or even surpass local government and law enforcement in terms of power and grip on civilian life. ICG quoted an MS-13 member as saying, "If a bus driver who pays us" -- or in other words, is extorted by the gang -- "gets robbed in his bus, he comes to us to solve the problem, not the police" (ICG). Thus, gang presence at its peak took the role of the ruling and dominant institution of El Salvador. Unfortunately for El Salvador's economy, Salvadoran gangs lack the qualities found by Acemoglu, Johnson, and Robinson in their paper *Institutions As a Fundamental Cause of Long-Run Growth* (2005) to be encouraging to economic growth. In particular, the attributes mentioned in the paper of broad property rights enforcement, self-constraint, and few rents being captured are majorly lacking: gangs

routinely confiscate houses for their own uses, which clearly violates property rights, capture rents as their main activity and source of income, and generally display little to no self-constraint (ICG). Because these attributes are lacking in the country’s dominant institutions, the gangs, the economy is disinclined towards growth. Similarly, weakened business participation and confidence lowers both capital investment (K in the Cobb-Douglas function) and labor participation in the workforce (lessens L), decreasing national GDP and leaving more Salvadorans jobless and economically worse off.

A secondary but notable negative economic effect of gang presence observed in El Salvador lies in resulting reductions in school attendance. The strong historical presence of MS-13 and Barrio 18 in El Salvador reduced school attendance through two main avenues: youth dropping out of school to become gang members, and families deeming the commute to school too unsafe for their child to attend because of gang activity (Hernandez). The first avenue can be proven with a statistic; a 2016 survey illustrated in Figure 4 reported that about 60% of gang members had joined the gang when they were under the age of 16, and therefore still school age (Cruz et al). The second avenue can be proven through case files and interviews that tell the story of young people found dead walking down the block, less than one kilometer away from their homes (NPR). Reduced school attendance and education will lower El Salvador’s human capital (L) in the long-run, and therefore lower Salvadoran GDP. Education is essential to any nation’s future growth, and its absence is a detriment to welfare.



Note: Sourced from *The New Face of Street Gangs: The Gang Phenomenon in El Salvador* by Jose Miguel Cruz et al.

Gangs exert a further negative impact on El Salvador’s L factor by spurring outward migration. The Council on Foreign Relations reported in 2022 that gang violence was “at the center of the region’s migration,” and International Crisis Group reported that in 2017 alone, 296,000 Salvadorans were displaced by gang violence (CFR) (ICG). Most who leave the country fleeing gang violence seek asylum in Mexico or the US – in 2020 and 2021, Salvadorans were the fourth largest group encountered by Customs and Border Patrol at the US-Mexico border and made up the plurality

of Central American immigrants, many citing the dangers and threats to their lives because of gang violence as their reason for migrating (Isacson) (Ward, Batalova). US immigrants from El Salvador are “especially likely to be of working age – 18 to 64” as 88% are in this age group compared to only 60% of people born in the US (Cohn, Passel, Gonzalez-Barrera). Thus, it can be concluded that gang violence in El Salvador has led to a significant loss of working age people, shrinking El Salvador’s labor force and decreasing its L factor.

A final negative economic impact resulting from gang presence arrives after dark: decreased consumer spending and economic activity because of fear of being outside at night, or for some, at any time of day. NPR tells the story of a schoolgirl, Aby, who began shutting herself in the house after the disappearance of her friend, not leaving except to go to school (McEvers, Garsd). Many more young people and adults do the same after dark, which is when gangs do most of their killings and violent activity (ICG). These self-imposed lockdowns out of fear for safety represent a loss in potential nighttime economic activity, or in many cases such as Aby’s, daytime economic activity. Small businesses like street food stands would have continued to make sales after sundown if it was safe to do so, but instead they close before dark for their own safety and because of a lack of customers, who have similar safety concerns. This loss in consumer spending decreases Salvadoran GDP.

Although gangs are by no means a positive economic force in El Salvador, they may offer employment outside of the formal economy (Seals). By offering an alternative source of employment for youths, especially during a period when it is difficult to find jobs, gang presence may increase El Salvador’s L factor. Typical jobs performed by young people associated with the gangs include collecting la renta, acting as lookouts, and running errands (ICG). People employed by the gangs may be less likely to migrate because of their employment; however, it’s important to note that upon any type of conflict or refusal between the person employed and the gang, that person will then become more likely to migrate out of concern for their safety (Duran). Although the Cobb-Douglas production function does not directly include any concept of unemployment, the jobs provided by gangs can be thought of as an increase in the workforce (L), since the more posts that are offered by the gangs, the more people are arguably able to enter the production function.

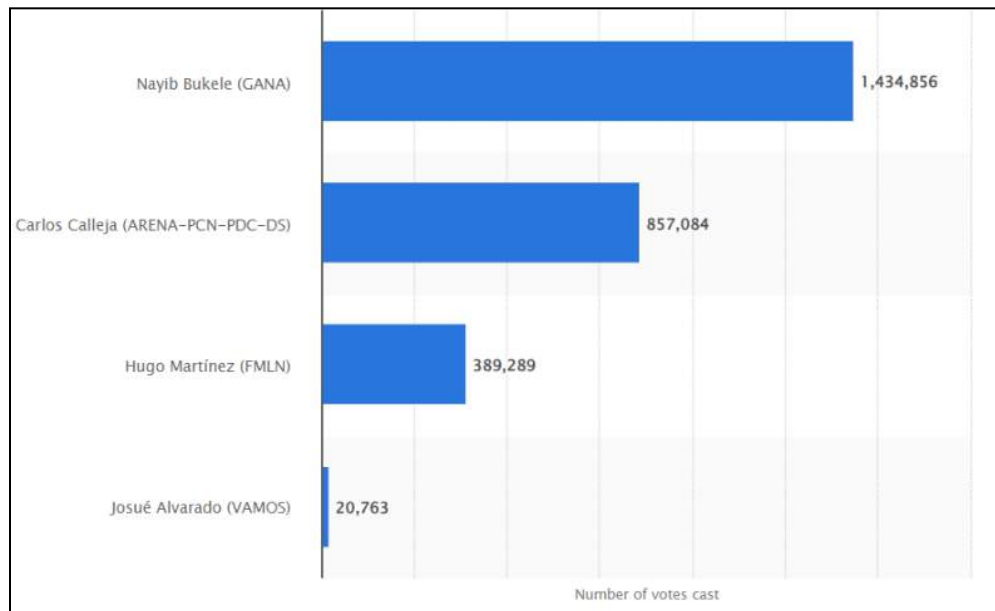
At the same time, it must be acknowledged that gangs do not lift their members or those they employ out of poverty (Martinez). Instead, gang activity creates a subsistence economy where the money generated through extortion rackets is spent on maintaining the gang and not on growth or improved welfare. Money brought in is typically sent up to leadership, and then used to pay for funerals, legal fees, and similar expenses that do not significantly improve the welfare of members and those employed. Any remaining money is spread thinly among gang members, who do not enjoy a significant improvement in their living conditions compared to before they joined the gang. Furthermore, although gangs provide informal employment and therefore increase the L factor, those employed are performing tasks and services that largely do not benefit Salvadoran industry or commerce. In fact, renta collectors and similar posts harm Salvadoran industry by impeding the flow of commerce and decreasing revenues. In summary, although gang presence may increase the Salvadoran workforce and add to its L factor, this provides only a marginal economic benefit, since

the work being done can detriment industry and commerce, and any conflict between gang employer and employee is likely to spur violence and encourage migration.

Background on President Bukele and his Mano Dura Gang Policy

President Nayib Bukele emerged victorious from the national Salvadoran elections of 2019 as a member of the GANA party, garnering more votes than either candidate from El Salvador’s two major parties, ARENA and FMLN, which had dominated Salvadoran politics since 1989. The votes cast in the presidential election of 2019 are shown in Figure 5. His Nuevas Ideas platform included promises of reduced crime and gang violence, improved infrastructure and job creation, and a reduction in corruption, which has stained the country’s politics for decades. An extremely popular candidate, Bukele won 53% of the popular vote – a clear majority (Statista Research Department).

Figure 5
Number of Valid Votes Cast in the 2019 Presidential Elections in El Salvador, by Candidate



Note: Sourced from Statista.

In El Salvador, presidents serve for 5 years, meaning Bukele was up for reelection again in 2024. Despite El Salvador’s constitutional ban on consecutive presidential terms, Bukele ran for reelection under his Nuevas Ideas party, and subsequently won a supermajority (Reuters). His huge margin of victory is shown in Table 1. Bukele remains ultra-popular, with his latest approval rating reaching 91% (Statista Research Department). Bukele had run on a platform of continued mano dura policy and promises of improvement in the economy and in infrastructure.

Table 1
Distribution of Votes Cast in the 2024 Presidential Elections in El Salvador, by Candidate

Nayib Bukele (Nuevas Ideas)	84.65%
Manuel Flores (FMLN)	6.4%
Joel Sanchez (ARENA)	5.57%
Luis Parada (Nuestro Tiempo)	2.04%
Other	1.34%

Note: Sourced from Statista.

What exactly is Bukele’s *mano dura*? A hardline, no-tolerance policy aimed at eradicating El Salvador’s gangs. It’s important to note that *mano dura* is not Bukele’s invention and has been attempted by past leaders with limited success. Strengthened police and military forces had managed to fill jails with gang members or gang associates, but prisons then became centers of recruitment, regrouping, and organization for extortion rackets. Arguably, gangs were strengthened by these past attempts.

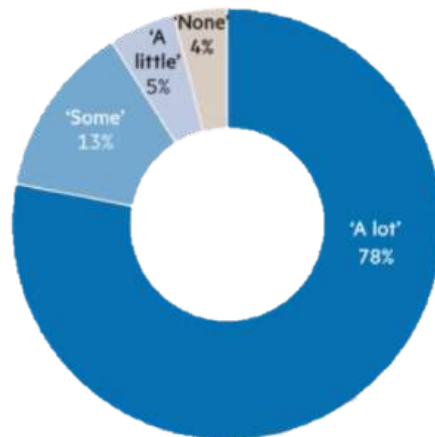
The centerpiece of Bukele’s version of *mano dura*, which has been declared a public safety success even by his political opponents, is his thirty-day *régimen de excepción*, or state of emergency. While the state of emergency is in effect, Salvadorans face a suspension of several constitutional rights: the right to freedom of association, the right to presumption of innocence, the right to legal counsel, the right to a fair trial, the right to be told the reason for one’s arrest, the prohibition of torture and discrimination, due process, freedom of assembly, and more (Roy) (Amnesty International). Cristosal also reported that under the state of emergency, arrests require little to no evidence to take place. Designed with the intention of use as a temporary measure, it was first approved by a Bukele-aligned government on March 27, 2022, and has since been renewed 29 times and been in effect for over two years (Aleman). The suspension of constitutional rights has sparked criticism from the UN, Human Rights Watch, WOLA, Amnesty International, Cristosal, and other watchdog groups.

Beyond suspending constitutional rights in the name of public safety, Bukele’s policy has also eroded access to reliable information and governmental checks and balances. His removal of several government officials, including supreme court justices, has left him a government where he faces little to no opposition, and is able to pass his *mano dura* measures without friction. Critics point this out as a sign of a deteriorating democracy and a rise in authoritarianism; Bukele himself once had an X bio that read “World’s Coolest Dictator.” Additionally, information that is meant to be public by law, such as information related to government management and the assets of public

officials, has since become classified (WOLA). Bukele’s government is therefore heavily lacking in transparency and accountability.

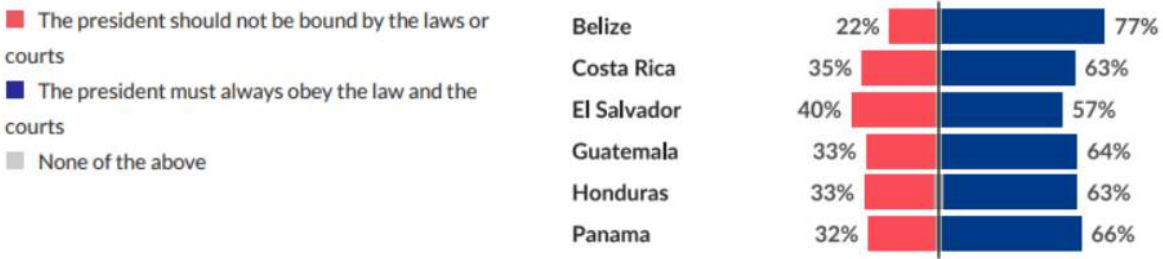
While concerns about democracy and human rights are being voiced, many Salvadorans are celebrating positive changes in their daily lives that have transpired since the dramatic reduction in gang violence. Their support for Bukele’s crackdown is shown in Figure 6, where 78% of respondent agree with Bukele’s mano dura measures “a lot.” Furthermore, Salvadorans say they can accept a president who ignores laws and courts, as shown in Figure 7, where Salvadorans are outliers in the percentage of Central Americans who hold this opinion as of 2023.

Figure 6
2022 Poll in El Salvador: Agreement with the Measures Taken Against Gang Members



Note: Sourced from CID Gallup Poll of 1,200 adults, 12-18.

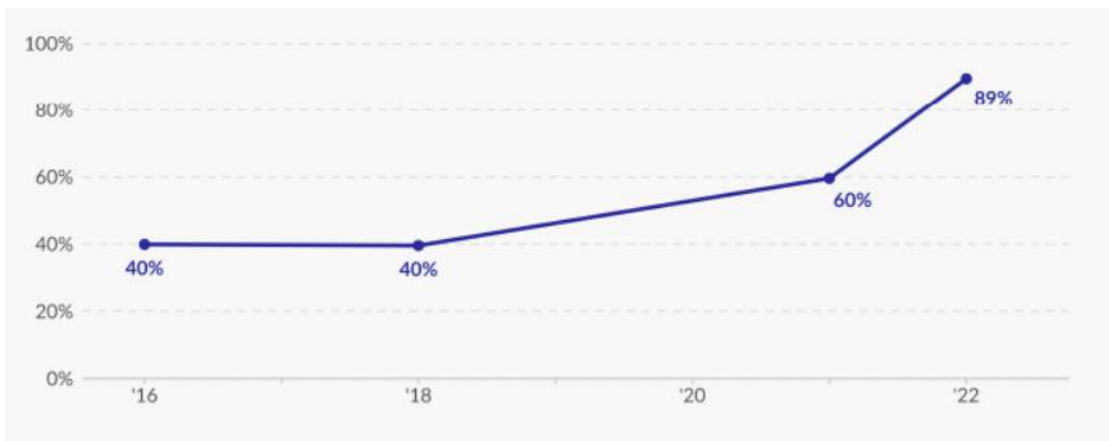
Figure 7
2023 Survey: Should the President be Bound by the Laws or the Courts?



Note: Sourced from WJP.

The reasons for this widespread support and acceptance are obvious. Public soccer fields that lay empty for years because they were the locations of shootouts now host soccer clinics and pickup games that run until after dark (Martinez et al). Neighborhoods that were once ghost towns after sunset now see residents sitting outside their homes playing cards until late at night. Families that were previously divided by gang territory lines can now visit each other in peace, without having to ask the ruling gang for permission. Small businesses who no longer must pay extortion promises are bringing in greater revenue. Bukele boasts that El Salvador is now the safest country on the continent. The leaps in public safety he flaunts are demonstrated in Figure 8, where an increasing percentage of Salvadorans feel safe walking in their neighborhoods at night.

Figure 8
Perceptions of Security in El Salvador Over Time: Percentage of Respondents Who Reported Feeling Safe or Very Safe Walking in Their Neighborhood at Night



Note: Sourced from WJP.

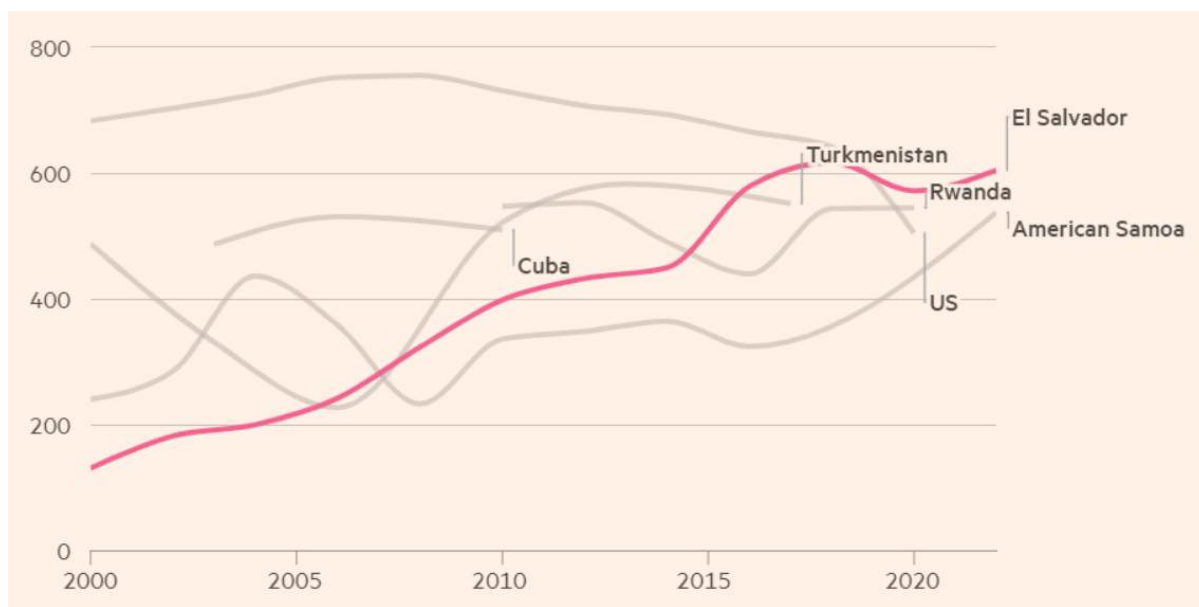
Despite high approval ratings for both Bukele and his state of emergency, not every Salvadoran is celebrating. The elimination of constitutional guarantees such as those to a fair trial and to due process have resulted in thousands of arbitrary arrests; Amnesty International reports 78,000 as of February 2024 (Miranda). Furthermore, Human Rights Watch declares 1,000 children have been convicted during the state of emergency as of July 2024. A girl at a bus station standing next to a stranger may be arrested along with him; a parent, girlfriend, or neighbor of a gang member may be arrested for ‘unlawful association’; a teenager in a poor neighborhood may be arrested for fitting the profile of someone a gang might have recruited (Mohor). Parents and spouses will exhaust all methods to locate and free their loved ones after they are detained, but often run into dead ends or the unfortunate news that their loved ones have passed away in state custody. Despite the construction of Bukele’s brand-new mega prison, the Terrorism Confinement Center, El Salvador is experiencing prison overcrowding of approximately 148% (Amnesty International). In fact, Bukele’s new prison is deliberately designed to overcrowd (Murry, Smith). Poor conditions, unrestrained state violence, and the mixing of gang members from rival groups has led to at least 261 deaths inside of overcrowded state prisons as of July 2024 (Cristosal).

However, prisoner deaths at the hands of security guards or other prisoners are no longer counted in El Salvador’s homicide numbers since a change implemented by Bukele, further contributing to his lack of transparency and giving the impression that there is less violence in the country than there really is. Critics accuse Bukele of swapping gang violence for state violence. Several reports of torture and mass graves have surfaced, many involving children under 18, who are often unlawfully held together with adult prisoners (Human Rights Watch).

Bukele’s Mano Dura Gang Policy and El Salvador’s Economy

Since the implementation of Bukele’s mano dura and the state of emergency, El Salvador has become the country with the highest incarceration rate in the world at over 1% (Insight Crime) (Mohor). El Salvador’s soaring prison population count is depicted in Figure 9, in which El Salvador has surpassed nations like Rwanda and Cuba to claim the highest prison population per 100,000 people in the world. Largely young men, El Salvador’s large prison population represents a loss in potential workers, lowering the nation’s L factor and having a negative effect on GDP (Kearney). This negative effect is exacerbated by the fact that Salvadoran prison systems have no programs preparing prisoners for reinsertion into society, meaning current prisoners are less likely to rejoin the labor force even after serving their sentences (Aleman). Moreover, a high incarcerated population, despite reportedly not receiving humane treatment, still weighs heavily on the existing debt and budget deficits.

Figure 9
Prison Population per 100,000 People, by Nation

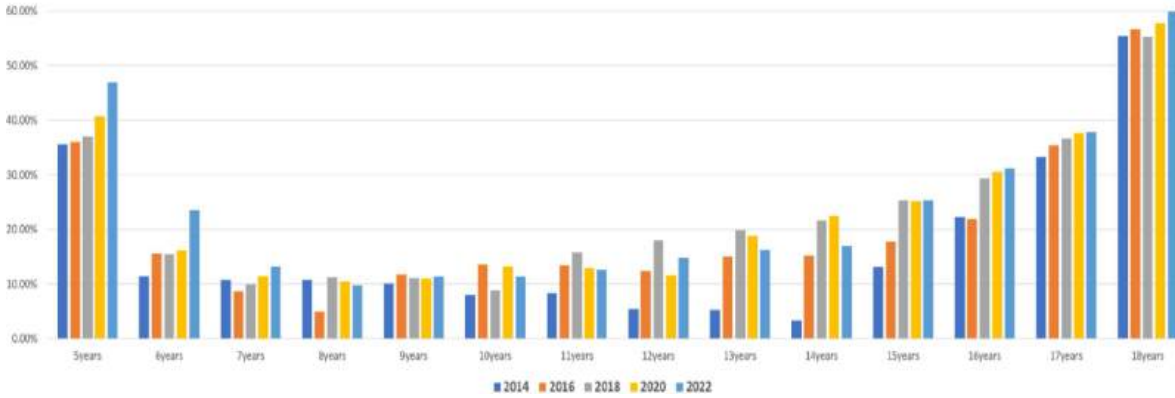


Note: Sourced from Financial Times and World Prison Brief. El Salvador is represented by the pink line.

Bukele’s *mano dura*, on the whole, has been expensive. The mass incarceration and the mega prison that accompanied it was only made possible by a large-scale, costly expansion of El Salvador’s military and police. Funds were diverted away from social, educational, and job programs towards the military and defense budget in order to fill jails. When combined with a total lack of planning for prisoner reentry into society and job markets after serving their sentences, this weakening of social, educational, and vocational government programs seems to only exacerbate the root causes of gang involvement in El Salvador: unemployment, broken families and communities. It’s possible that El Salvador may see its gangs revived stronger than before. Even more likely, it’s possible that El Salvador is filling jails at the expense of future employment and production levels. Less educational and vocational programs will likely result in an overall lesser-trained labor force, diminishing El Salvador’s L factor, and therefore, its GDP. Figure 10 shows the percentage of Salvadoran children of a given age, 5 years to 18 years, who are not attending school, over the years 2014 to 2022. Bukele took office in 2020, and began his *mano dura* gang crackdown soon thereafter, and the preceding years were times of record high gang violence and activity. As one would expect, these gang-ridden years, especially 2018, displace high percentages of children who are at gang-joining age – 12 years and older – and not in school. However, the beginning of Bukele’s crackdown in 2020 did not signal much of an improvement. In fact, Figure 10 shows increasing percentages of children out of school when looking at ages outside or on the edge of El Salvador’s compulsory schooling years, which are 6 years to 15 years. Figure 11 reinforces this trend, showing the total number of students enrolled in school in El Salvador continuing to drop even after Bukele takes office and begins taking action to fight gangs. This negative trend despite improvements in safety and gang pressure can largely be attributed to Bukele’s lack of investment in education in favor of larger military and law enforcement budgets. The fewer children that attend school now, even outside of the mandatory years, the weaker El Salvador’s L factor will be when they come of age and

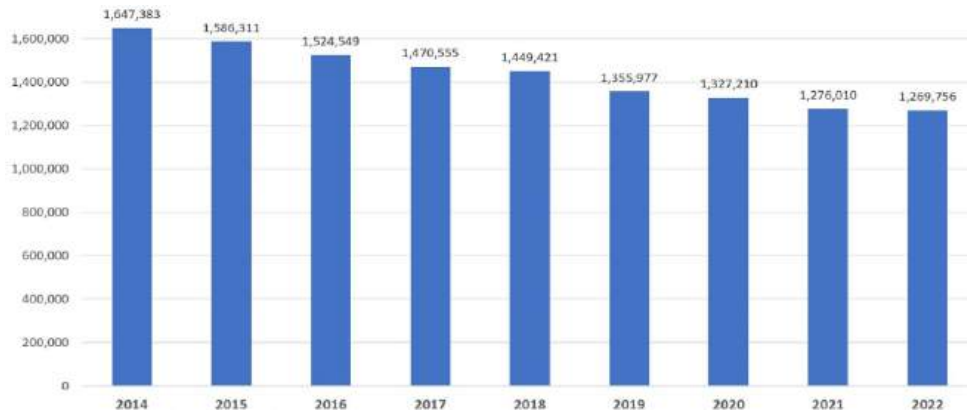
begin searching for employment.

Figure 10
Percentage of Salvadoran Children of a Given Age Who are Not Attending School



Note: Sourced from MINED.

Figure 11
Students Enrolled in Schools in El Salvador



Note: Sourced from MINED

Alerts and alarm bells from human rights watchdog agencies have sparked concern from El Salvador’s trading partners and those who send foreign aid, such as the US, who is El Salvador’s number one trading partner in and has sent billions of dollars in aid in the past 3 years. US Vice President Kamala Harris voiced Washington’s concerns about El Salvador’s democracy in 2021; if alarm from the US and other trading partners continues to grow, El Salvador could see its foreign investments and foreign aid vanish, negatively affecting business confidence in the process (Garriga) (ICG). However, as the US and Mexico, which is another of El Salvador’s top trading partners, both continue attempts to reduce the flow of migrants they encounter from the Northern Triangle and beyond, they may leave democracy concerns behind and prefer a president who has seen more than a

60% drop in migration since taking office (Goodman, Aleman). This decrease in outward migration is clearly beneficial for El Salvador's L factor, and favorable to an overwhelmed US immigration system, too. A former president of Inter-American Dialogue said, in regard to the US's goals and concerns, "Migration trumps everything else" and many American politicians credit Bukele with "address[ing] the root causes of migration" (Goodman, Aleman). Foreign investments and aid to El Salvador from the US and Mexico will depend on each nation's priority – humanitarianism, or reductions in incoming migrants?

Despite the massive decrease in outward migration that has occurred during Bukele's time in office, outward migration from El Salvador continues, although migrants' motives may be changing. Before Bukele's *mano dura* and the vast reduction of gang presence in El Salvador, violence was the primary reason behind migrant departures (World Bank). However, in 2023 and 2024, a different motive began to appear: fear of unjust incarceration (Mohor). Men and women who fear they may be profiled as gang members or associates, or who have family ties to gang members, fear being jailed for decades on the charges of 'unlawful association' or similar. Bukele has not completely ridded El Salvador of outward migration.

On the bright side, Bukele's *mano dura* has likely brought with it an increase in business participation and confidence. According to Acemoglu et al and their analysis of institutions conducive for economic growth, Bukele's government, in taking the place of gangs as El Salvador's ruling power, has fortified El Salvador's institutions and therefore increased business participation and confidence (Acemoglu et al) (ICG). Under Bukele's government, businesses can sell in whatever neighborhood they please, sell and buy from whoever they please, and cross old gang territory borders without fear (Moloney). Now that Bukele's government instead of gangs act as El Salvador's dominant institution, business conditions are more favorable and facilitate greater confidence and participation, increasing the A component of the Cobb-Douglas production function and leading to vital GDP growth.

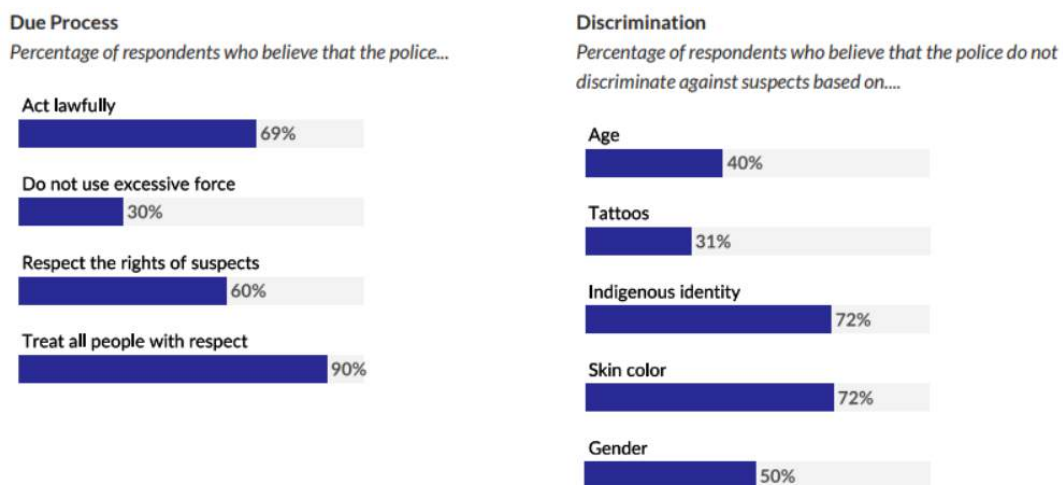
Additionally, Bukele can claim with some accuracy that his *mano dura* has restored rule of law in El Salvador because his government vastly increased protections against injustices committed by gangs. World Justice Project found that 9 in 10 Salvadorans felt safe walking in their neighborhood at night in 2023, which was an improvement of 29 percentage points since 2021 and 38 percentage points since 2013 (World Justice Project) (Pew Research Center). Furthermore, according to the 2023 report, Salvadorans were confident that police help them feel safe (93%) and resolve community security problems (90%) (World Justice Project). When citizens feel safe in their communities and confident that police will maintain order, rule of law is improved. A restoration or improvement in rule of law leads to capital accumulation, leaps in education, and greater research and development. Thus, within the Cobb-Douglas production function, all factors improve when rule of law improves – A, L, and K all increase. Therefore, Bukele's improvement to Salvadoran rule of law should have an outsize effect on Salvadoran GDP.

On the other hand, community safety and police effectiveness are not the only factors that are used to measure rule of law. Constraints on government powers and fundamental rights are also vital components, and Bukele fails to score as high in those categories (World Justice Project).

Considering the suspension of many constitutional rights during the state of emergency, including but not limited to due process, as well as reports of unlawful treatment of detainees and prisoners, Bukele’s government seems to score poorly in the ‘fundamental rights’ category outlined by the World Justice Project (WJP). His removal of several government officials to craft a government that is more favorable to an aggressive *mano dura* agenda worsens his ‘Constraints on government powers’ score, especially when examined alongside his unconstitutional reelection and misuse of state of emergency, which was intended for temporary use. However, it’s important to remember that the vast majority of Salvadorans supported Bukele’s reelection and continue to support the state of emergency.

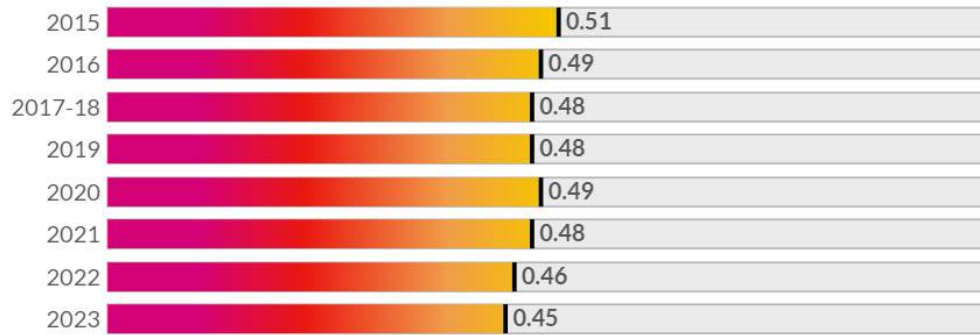
Further undermining El Salvador’s rule of law, police behavior during the state of emergency and gang crackdown is reportedly discriminatory and excessively forceful. Figure 12 shows that Salvadoran survey respondents are not confident that their police do not use excessive force or discriminate on the basis of age, tattoos, or gender (WJP). Young men with tattoos are the most stopped by police who assume them to be gang members because of their age, tattoos, and gender. This type of discrimination, excessive force, and lack of checks and balances and fundamental rights all combine to exert a net negative effect on El Salvador’s rule of law according to the World Justice Project, which has decreased El Salvador’s Rule of Law Score by 0.06 points, as shown in Figure 13. 0.04 of those 0.06 points were lost while Bukele has been in office.

Figure 12
2022 Salvadoran Survey Respondents on Law Enforcement Due Process and Discrimination



Note: Sourced from WJP General Population Survey 2022. Note the low percentages of respondents who believe police do not use excessive force, do not discriminate on the basis of age, tattoos, or gender. Young men with tattoos are the population most targeted by the police for fitting the profile of a gang member; having tattoos was the reason behind many arrests during the first days of El Salvador’s state of emergency (NPR).

Figure 13
El Salvador Rule of Law Score Over Time, 2015-2023



Note: Sourced from World Justice Project. Note that the score has decreased by 0.06 since 2015, and that 0.04 of the 0.06 point loss have been lost since Bukele’s inauguration in 2020.

Regarding rule of law in El Salvador, Bukele has brought improvements in the categories of criminal justice, and order and security alongside clear downgrades in the categories of fundamental rights and power constraints. Thus, the outsize effect that either an improvement or deterioration of rule of law would have, since rule of law affects every component of the Cobb-Douglas production function, is largely absent. Instead, opposing changes work to keep economic effects of improvements or deterioration of rule of law largely arbitrary, with slight decreases in El Salvador’s Rule of Law Score being cause for concern regarding the long run but not doing much to dampen current positive sentiment.

Conclusion

This paper sought to analyze both the positive and negative economic effects of past gang presence and current gang policy in El Salvador, and sequence them in order of magnitude to provide insight for improvements to policy.

In regard to past gang presence in El Salvador, its net economic effect was overwhelmingly negative despite the informal employment gangs provided to populations who may have otherwise been unemployed. The money lost by businesses large and small to extortion exerted the greatest economic impact alongside dampened business confidence and participation. Weaker but still significant effects were exerted by reductions in school attendance and violence-fueled outward migration.

In regard to Bukele’s *mano dura* policy, its net economic effect is more difficult to conclude, but seems to point towards long-run harm unless action is taken in the short term. The diversion of funds away from programs for education and jobs in order to support new jails and a military expansion could spell danger for the long run future of El Salvador’s labor market. The suspension of rights under Bukele’s state of emergency, and other indicators of democracy’s deterioration in El Salvador that human rights organizations have expressed concern about, have the potential to endanger the foreign investment the country has been trying to attract and dampen business confidence and participation in the long-term. Outward migration may rebound to past levels in the long run if fears of being unjustly detained continue to increase. Gang presence may also rebound in the long run if measures are not taken to prevent their strengthening in prison settings.

In the short run, however, El Salvador enjoys levels of safety it has not seen in decades, and a boost in business participation and confidence as a result. The near-total elimination of extortion rackets frees up money that businesses may use to expand, pay higher wages, create jobs, or invest in capital. El Salvador's short-term economic growth is highly likely, as Bukele promised in his presidential campaign.

To build sustainable economic growth that lasts beyond the short run, El Salvador should concentrate on the root causes of gang participation: unemployment, poverty, and limited education that transforms into limited employment opportunities. The current state of El Salvador's budget and policy foreshadows a stale job market, a low labor force participation rate, and, upon the end of current prisoners' terms, a wave of people reentering society without a semblance of readiness. History dictates that youth in El Salvador without a job and without an education are significantly more likely to join a gang. The government should allocate more funds for job training, education, and reintegration for those imprisoned instead of putting the same funds once used for education towards incarcerating as many people as possible. Various experts have concluded that rehabilitation of gang members is an essential step towards lasting reductions in violence, but Bukele's version of *mano dura* accomplishes almost nothing in this regard (ICG). Prisoners who haven't received any reintegration support are likely to turn to gang participation upon release, or at the very least, find themselves unemployed and unqualified.

Additionally, Bukele and the Salvadoran government should strive to operate in a fair justice system with more sophisticated policing efforts (ICG). Internal checks on security forces and the further development of 'checks and balances' to restrain any individual member of the police or military's power are advisable. An immediate improvement in the nation's defense of human rights is needed to ensure future foreign investment and economic growth. Bukele's government should alleviate prison overcrowding and reinstate due process, as well as all other fundamental rights that have been suspended during the state of emergency. In 2029, at the end of Bukele's current term, El Salvador should facilitate a peaceful transfer of power and strive to cultivate a government of diverse opinions and increased transparency.

Future research may choose to analyze Bukele's *mano dura* in hindsight, reporting on long-term effects and economic outcome. El Salvador's economic state in the coming years will largely be a direct result of Bukele's current and recent actions. Only time will tell if El Salvador's current, relatively positive climate will extend into the long run or turn sour as Bukele's failure to invest in the more distant future reveals itself.

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Therapy Use of AI-Powered Visual Art and Music for Neurodivergent Individuals

By Syed Armaan Bashir

Introduction

Over the last few years, there has been an increased incorporation of AI in art and music therapy, leading to positive outcomes for those diagnosed with ASD and Down syndrome or other forms of neurodivergence. The mentioned technologies are of significant benefit because they provide customized and engaging therapeutic sessions that enhance creative boundaries and teach fine motor control and emotional regulation. AI provides real-time response and modification to therapy sessions according to client responses, leading to more effective interventions (Williams et al. 4). However, there are new concerns, such as ethical questions related to data privacy and little or no empirical evidence for the efficacy of AI-based therapies. This paper discusses how AI art and music therapies might help neurodivergent persons, what opportunities these treatments possess, and what limitations they have for better quality of life and increased social inclusion of such citizens.

Art Therapy and AI

Art therapy has been accepted for years as a treatment. Those who may not be able to use art therapy verbally have an opportunity to express themselves and communicate. Neurodivergent learners with ASD or Down syndrome can express themselves creatively through art and enhance their motor coordination, vestibular/proprioceptive sensitivities, and affect regulation (Brancatisano et al. 4). AI further amplifies these because of the individualized approaches used by an AI platform. For instance, AI algorithms can interpret an individual's gesture or facial movement while painting to inform the therapists in real-time. Such feedback assists in modifying the therapy session regarding its utility and interest during the session.

Furthermore, the art platforms built using Artificial Intelligence technology can create exercises in art based on the person's choices and subsequent reactions, starting from a more superficial level and escalating to more complications that build up the skill set (Hu et al. 4). This kind of interaction ensures that the patient is at liberty to make their own decisions that will enable them to be as independent as possible and express him or herself since most patients with neurodivergence have challenges receiving and responding to standard communication methods.

Music Therapy and AI

In the same way, music therapy, as the use of music as an instrument of therapy, focuses on the cognitive, emotional, and social aspects of human beings. Self-awareness concerning the sensory system is shared among neurodivergent individuals, and as such, music therapy aids in enhancing these aspects (Khalid et al. 950). AI enhances music therapy by designing and generating musical experiences through which the patient-therapist partnership can relate to the latter's sensory processing system.

Music therapy apps for individuals are specific applications that can adapt to a person's conditions in terms of tempo, tone, and level of comprehension in real time. For example, using

music with autism spectrum disorder, patients might improve gradually by increasing the volume or the rhythm of the sounds while keeping the atmosphere in the room quiet (Zhou and Lee 6). With the help of pulse, oxygen level, blood pressure, and other factors, music therapy can be more individualized and fun, thus staying in the therapy frame and helping the patient.

Applications and Case Studies

Several applications of AI have already revealed promising possibilities for their use in the therapy of neurodivergent persons. For example, Brain Power combines Google Glass with AI programs to help ASC users identify and understand people's facial expressions and thus interact effectively (Hayashibara et al. 5). The other is Infinite Flow, which is an integration of AI and dance, enabling the performance of flow arts to the disabled and non-disabled persons, affirming the value of togetherness. In addition, there are AI composers such as Aiva Technologies that have been designed to generate music pieces for coping with anxiety and stabilizing the mood of persons with Autism spectrum disorder (ASD) and Down syndrome. Such applications demonstrate AI's capacity to support people with neurodivergence by utilizing their art and melody talents.

Benefits and Challenges

Art and music employed in combination with AI have been found to have advantages when used to address neurodivergent individuals' needs, but some issues must be taken into consideration.

Benefits:

Personalization: Artificial intelligence allows therapy sessions to adjust to the patient's preferences, the degree of sensitivity to certain stimuli, and cognitive functions. Still, using the data collected from users, the AI algorithms can adjust the interventions offered in real-time, thereby providing therapy in the most suitable ways as mandated by the numerous requirements it has to meet.

Accessibility: AI-based digital platforms and virtual environments provide equal chances for therapy to people in developing or other tremendously inaccessible areas (MacIntyre et al. 3). This aspect is essential to guarantee that people with neurological disorders can receive treatment regardless of area limitations that may hinder such service.

Engagement: AI promotes interactive and immersive tools that improve patients' interest and participation in therapy. Techniques that involve artificial intelligence include using prompts created by artificial intelligence or developing virtual reality exercises that would keep the patient's attention and even help them maintain that attention throughout the session.

Challenges:

Ethical Considerations: A significant issue of privacy is brought up with regards to the collection and utilization of highly confidential personal information in therapy with the use of Artificial Intelligence (MacIntyre et al. 4). The following ethical issues happen and need strong measures to protect user's data and transparent policies in handling such data.

Validation:

However, one should remember that AI-based therapies, when applied to the neurodivergent population, should undergo empirical confirmation of the promised benefits. It is, therefore, necessary to assess the benefits of research by its ability to establish long-term outcomes, the applicability of the results across different populations, and the success in relation to conventional treatments.

Integration: Training of therapists for AI technologies along with implementing new technologies into existing therapeutic models present some of the compatibility issues, along with general issues such as a threat to the therapeutic alliance (MacIntyre et al. 5). Another issue that is very important to address is whether the implementation of AI will increase or decrease interactions between professionals.

Conclusion

In conclusion, the implementation of AI in visual art and music for the therapy of neurodivergent persons, including those with ASD or Down syndrome, has excellent potential. Thus, focusing on that, AI helps make therapeutic experiences more effective and enjoyable due to individualization, constant feedback, and flexibility. It is expected that with the development of technology shortly, this concept of using AI in art and music therapy will open new heavens for growth and development, not only for the neurodivergent population. Thus, constantly developing AI-based therapy for neurodivergent people is a promising prospect for their support and further enhancement of their quality of life through artistic activities.

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Meal Timing and Alzheimer's Disease By Grace Beck

Introduction

AD (Alzheimer's disease) is a mysterious, multifactorial, incurable, fatal neurodegenerative disease that affects more than 4 million people in the United States. Predominantly a disease of old age, it affects one in ten people over 65 and more than half of the population over 85 (Nolan H. Et al., 2001). Despite 80 years of research, a review has concluded that "The pathogenesis and etiology of AD remain an unknown territory" (Chinthapalli K. et al., 2014). The 'amyloid hypothesis' suggests that the presence of plaques composed of insoluble fibers in the brain accounts for AD symptoms, but this theory is contentious. "Doctors gradually began to recognize the disease, but the truth is, doctors are not certain that these plaques cause AD, and doctors are unclear about what usually causes those plaques" (Leibing A. Et al., 2021). Such limited understanding renders AD incurable.

The amyloid hypothesis has been the primary focus of pharmacological research interventions for several years. However, interventions only showed efficacy in the early stages of the disease. Mild to moderate AD patients treated with an experimental drug, named monoclonal antibody, showed no cognitive improvement after eighteen months (Leibing A. Et al., 2021). Researchers attributed this to the notion that AD begins long before symptoms appear; one study found that the level of amyloid-beta ($A\beta$), the key component of the amyloid plaque, is altered 25 years prior to the onset of symptoms. However, what renders AD impossible to cure is that early identification of AD remains challenging, with up to 25% of participants in a phase III trial being misdiagnosed. No biomarker yet demonstrated a "high probability" of disease (Chinthapalli K. et al., 2014). Consequently, many researchers have shifted their focus from developing curative treatments to emphasizing early lifestyle intervention. Italian researcher Stefano Sensi noted, "The failure of the Phase 3 aducanumab trial (AD drug candidate) clinical trial is another warning that the field must take a different approach. "Some authors have already called for a rejection of the amyloid hypothesis ... we need to remind ourselves that a third of AD cases are strongly dependent on modifiable factors like low education, midlife hypertension, midlife obesity, diabetes, physical inactivity, smoking, and depression" (Leibing, A. Et al., 2021).

A growing body of evidence suggests an association between dietary habits, one of the most important modifiable lifestyle factors, and the risk of AD. Several studies have indicated that dietary restriction, whether caloric or intermittent fasting, may offer protective benefits against AD (Kim W. Et al., 2022). However, no research has yet examined the effect of the timing of meals on AD development. The timing of meals could be a critical factor that requires appropriate investigation for its association with disease development as abnormal meal times are becoming prevalent in modern society. With the advent of affordable artificial lighting and industrialization, modern humans began to experience prolonged hours of illumination every day, leading to extended food consumption compared to our ancestors before the 18th century. This contemporary lifestyle has allowed people not only to work after sunset but also to eat

during the night, promoting abnormal timing of meals. In industrial societies, nearly 10% of the workforce performs night-shift work: permanent night work, rotating shifts, or irregular schedules. Approximately 10% of the workforce in industrial societies performs night-shift work, which is associated with various diseases, including diabetes, obesity, and neurodegenerative disorders (Mattson P. Et al., 2014). However, the relationship between meal timing and AD has not been examined. The increasing prevalence of individuals failing to align their eating patterns with the circadian rhythm and the insufficient understanding of its correlation with AD underscores the imperative need for research into how meal timing may influence the development of AD. This research is essential for advancing lifestyle interventions that could mitigate this risk. Given that AD is a multifactorial disease where prevention is crucial, identifying additional lifestyle factors would allow more effective strategies in reducing the risk of AD.

In hopes of finding a potential lifestyle factor that could contribute to the prevention of AD, this ex post facto correlational study examines how meal timing varies between AD patients and individuals without the disease. By doing so, this study aims to illustrate a potential association between meal timing and the risks of AD, thus identifying predictions of the effects of certain meal timings on the development of AD.

Operational definitions:

Meal timing

For this study, “meal timing” refers to whether meals align with the circadian rhythm. The two most important aspects of circadian rhythm are food consumption during daylight hours and fasting for hormonal maintenance.

Eating during the Bright time

Brightness is a crucial factor in eating habits because oscillators that control the circadian rhythm in the central nervous system (CNS) are synchronized by changes in light and darkness. In other words, the eating behavior of animals and humans is strongly related to the daylight cycle (Nutric Recommendations et al., 2004). In this study, the brightness was measured through the participants’ perception of past brightness during meal times to ensure accuracy, as brightness depends on seasons and individual days rather than specific times.

Fasting

Fasting is necessary for cellular maintenance and alignment with the circadian rhythm as it allows glucose conservation and repair of damaged cells. Hormonal maintenance is achieved through fasting, which is abstaining from food for a certain period of time (Lesica A. Et al., 2017). Although there are various types of fasting, this study measured fasting based on a 16-hour period, as outlined in a study on meal timing aligning with the circadian rhythm (Charlot, A. Et al., 2021).

2. Literature Review

The biological block

To assess the potential relationship between meal timing and AD, it is essential first to understand meal timing's importance to health. From a chronobiological perspective, humans are diurnal (active during the day) and are programmed to rest and mobilize blood glucose during the night (Lowden A. Et al., 2010). Human ancestors aligned their cellular schedules around food consumption during daylight hours and fasting during the dark. Oscillators in the CNS, synchronized by the changes in light and darkness, largely determine the temporal organization of eating. During nocturnal sleep and fasting, metabolism is designed to mobilize endogenous blood glucose for the brain cells and to save blood glucose by a decreased uptake in muscle cells- a phenomenon called insulin resistance. Hence, the absence of fasting during nocturnal periods leads to an increase in insulin resistance, accounting for the correlation between nocturnal eating and type 2 diabetes and metabolic syndrome (Nordic Nutrition Recommendations 2004., Et al., n.d.). During the day, cellular processes are scheduled for food intake-related activities such as digestion, insulin release, and glucose storage. Conversely, at night, cells engage in maintenance activities, such as waste removal and repair (Lesica, N. A. Et al., 2017). Available blood glucose levels are reduced at night, leading to glucose conservation (Lowden A. Et al., 2010). As a response to the schedule, liver cells adapt by releasing substantial quantities of glucose during the night and minimal quantities during the day. This pattern forms what is known as circadian rhythm, wherein cells perform different functions based on the time of the day. These activities are regulated by the brain's internal clock, which organizes cellular functions by responding to the cyclic patterns of light and darkness (Lesica, N. A. Et al., 2017). Disruption of the circadian rhythm can impair cellular function and lead to significant metabolic alterations.

Effect of meal timing on metabolic changes: insulin levels

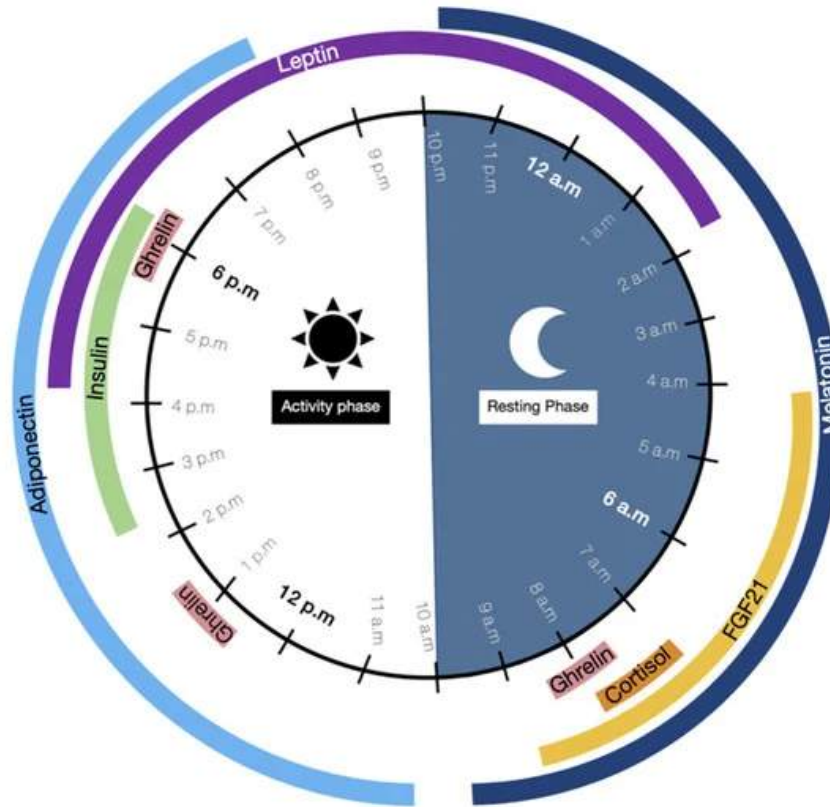


Figure 1: Time-of-day-dependent patterns of circulating hormone levels variation (Charlot, A. Et al., 2021).

The circadian rhythm gets disrupted when meals are consumed at night when cells are typically inactive in glucose processing. It causes a substantial release of glucose both day and night, thereby increasing overall glucose levels (Lesica, N. A. Et al., 2017). Postprandial blood glucose levels (Blood glucose levels after meals) usually increase and then return to baseline as insulin is released. The same pattern applies to insulin secretion as shown by participants in a simulated shift work study exhibiting elevated postprandial insulin response after a late evening (22:30 hours) meal (Lowden, A. Et al., 2010). Research by the American Physiological Society has shown that glucose and insulin responses vary significantly with the time of day, regardless of the duration of fasting. Evening meals result in approximately double the glucose response and up to 50% higher insulin secretory responses than morning meals (E Van Cauter et al., 1992). Nocturnal eating itself elevates insulin levels regardless of the fact that the same content is consumed as usual as cellular processes adhere to their diurnal rhythm of releasing substantial quantities of glucose throughout the day and adjusting to nighttime eating by increasing glucose release. As seen in Figure 1, during the afternoon from 2 to 6 p.m. and especially at the 4 to 5 p.m. peak, insulin is produced by pancreatic islets and induces metabolic changes. Food consumption should not occur during the insulin peak because it induces fat storage, nor should it occur at night (Charlot, A. Et al., 2021). Meal times around late afternoon and night render an increase in insulin. Thus, the optimal meal time for food intake to prevent an elevation of insulin levels would be morning to early afternoon.

Effect of fasting on metabolic changes: Inflammation and obesity

The circadian rhythm schedule involves fasting to regulate glucose levels and eliminate metabolic waste. Consuming meals late at night, followed by an early morning intake, can lead to cellular stress and accumulation of metabolic by-products, overwhelming the body's capacity to process glucose and fats efficiently. With an accumulation of partially metabolized fats and glucose, inflammatory processes are likely to occur. A study with mice demonstrated that those fed randomly throughout the day and night exhibited typical obesity-related issues, including inflammation and insulin resistance. In contrast, mice fed only during their active daytime hours maintained health comparable to those on a natural diet (Lesica A. Et al., 2017). The study suggested that disrupting cellular rhythms through consistent consumption in the absence of breaks for maintenance could lead to weight gain and other issues, including inflammation and insulin resistance, despite having the same caloric intake as daytime eating. Fasting as a practice in the management of several diseases, including diabetes, obesity, cancer, or neurodegeneration, is becoming increasingly popular. However, the effects of fasting are heterogeneous as some studies have shown no improvement in risk indicators for cardiovascular disease, and some have shown only short-term improvements. Nevertheless, internal cellular processes highlight the importance of hormonal maintenance in inhibiting inflammatory processes.

Human studies have also shown that night shift workers are more likely to be overweight or diabetic, correlating meal timing with weight gain (Lowden, A., Et al., 2010). Accumulating evidence from systematic reviews and meta-analyses suggests the association of overweight and obesity with cognitive decline and increased risk of AD (Ezzati A. Et al., 2023). Moreover, it is known that metabolic factors like insulin resistance or low-grade inflammation might contribute to age-related cognitive impairments (Witte A. V. Et al., 2009). Understanding the relationship between these metabolic changes and AD could elucidate the unknown relationship between AD and meal timing.

AD and its relationship to metabolic factors

Research has consistently found correlations between insulin levels and AD. Elevated insulin levels have been reported in AD patients' blood and cerebrospinal fluid (the fluid that cushions and carries nutrients to the brain) (Nolan H. Et al., 2001). Improved insulin sensitivity, which reduces the need for high insulin levels, has neuroprotective effects, while increased peripheral insulin levels promote cognitive impairments and AD (Witte, A. V. Et al., 2009). Insulin receptors are highly concentrated in brain regions that degenerate during AD, such as the choroid plexus, the olfactory bulb, the cerebral cortex, the structures of the limbic system, and the hippocampus (Chinthapalli, K. et al., 2014). Excessive insulin leads to neuronal death through excitotoxicity, as seen with chronic N-methyl-D-aspartate (NMDA) receptor activation in AD brains. Memantine and NMDA glutamate receptor antagonists were initially developed to treat diabetes by potentially lowering insulin levels to block this chronic activation without disrupting normal transmission (Chinthapalli, K. et al., 2014). A group of researchers found that both amyloid-beta ($A\beta$) and insulin are broken down by insulin-degrading enzymes in the human brain. Excessively high insulin levels in the rat brain completely blocked IDE from working

correctly. Therefore, they hypothesize that excessive amounts of insulin would cause a build-up of A β plaques in brain tissue (Nolan H. Et al., 2001). These researches suggest a cause-and-effect relationship between high levels of insulin and AD, indicating that meal timing might influence the risk of developing the disease by disrupting daily rhythms and increasing insulin levels. Inflammatory processes, which are likely to occur without regular maintenance, have also been implicated in the pathogenesis of AD (Lourida, I. et al. 2013). Overall, such associations suggest a significant association between the risk of developing AD and disturbances in daily rhythms caused by nocturnal eating without proper maintenance. However, no research has studied the association between AD and meal timing. Researchers state that AD exists approximately 25 years before the onset of symptoms, as indicated by an altered state of A β levels in studies (Chinthapalli K. et al., 2014). Thus, this study studied whether there is a relationship between an individual's meal time in the past and their risk of developing AD in the future.

Gap in Research

Previous studies have found metabolic effects from meal timing that have shown potential links to AD. Meal timings that are misaligned with the circadian rhythm have been shown to elevate insulin levels, render inflammatory processes, and cause obesity-related complications. These metabolic disturbances due to misaligned meal timings have been proposed as potential pathogenesis of AD. Studies have been done extensively on meal timing and metabolic factors, as well as between metabolic factors and AD; however, no research has explored the direct relationship between meal timing and AD when those studies evidently indicate a potential relationship. Therefore, this study can contribute to the literature by examining the potential connection between AD and meal timing if meal timing is associated with whether or not one develops AD. Considering the fact that AD starts developing approximately 25 years before the actual onset of symptoms such as memory loss, this study investigates individuals' meal times in the past to see if it is directly related to their risk of developing AD in the future. This study can provide information on whether or not the past meal times of AD patients differ from those of AD individuals and further predict if certain meal timing would have a higher risk. The insights gained from this study would add to the potential prevention strategies for AD. Since AD is a multifactorial, incurable disease, this study is essential for identifying new preventative approaches.

3. RESEARCH DESIGN AND METHODOLOGY

Research Design

As seen in the literature review, AD develops approximately 25 years prior to the onset of symptoms. Thus, it is essential to examine past meal times before the onset of symptoms. Given the inability to predict people who have AD in the future, the researcher conducted an ex post facto study. An ex post facto enables the researcher to compare the differences between Non-AD individuals and AD patients' past meal times. Thus, the researcher could predict the causal relationship between meal timing and AD. This ex post facto study was done by collecting data *after the fact*, meaning the data was collected from those already diagnosed with AD. The ex

post facto research adopts several aspects of an actual experiment but is non-experimental– it makes comparisons between individuals who belong to different groups (in this case, the two groups are AD patients and Non-AD individuals) thus, the researcher could predict and cause and effect relationship. (Akinlua, S. 2019). An ex post facto study is most appropriate in the case of examining the relationship between AD and meal timing, as it explores whether unhealthy meal timing could lead to AD without manipulating the meal timings of individuals. Conducting experiments that manipulate meal times to assess the direct impact on AD development would be ethically questionable as it has the potential to harm participants. Even though a strong correlation is found, it doesn't mean that specific meal timings *cause* AD. Despite the fact that a causative relationship couldn't be concluded, correlation can be assessed, which can demonstrate a prediction of a potentially modifiable lifestyle risk factor in AD development that could be developed by further research. Additionally, a survey was conducted because a question regarding their brightness needed the participant's memory of the brightness of their usual meal times as it varies from individual days and season to seasons. This aspect of the study was crucial to understanding the context of meal timing beyond the plain clock times. Moreover, this study was approved by the IRB.

Sampling

The method of data collection and sampling consisted of four steps. First, the researcher acknowledged that to match the AD groups and non-AD group ages, the researcher needed a full sample of AD patients first. Between January and March of 2024, the researcher listed AD facilities and hospitals and sought people around her for personal connections with individuals diagnosed with AD. Second, for people the researcher knew, the researcher printed out the questionnaire (Appendix 5) with the consent form physically and provided caregivers with questions on the questionnaire directly. Fifteen people were from the researcher's personal connections, such as the researcher's Grandma and mom's friend. Etc. Third, for the sake of qualifying a minimum sample size of 30 to establish the suggested sample size to assume a statistical relationship, the researcher went to a Dementia Facility in Gwangjin-gu, Seoul, and asked patients and caregivers in the waiting line to physically inquire for their contribution to answering the survey questions. Since the researcher acknowledged that not everyone was willing to volunteer, the researcher offered a 5 thousand-won coupon for patients and caregivers in line. AD patients and caregivers had to sign the consent form, but the researcher instructed only the caregiver to respond to the questionnaire. The caregivers were ensured that time would be consumed and that their past knowledge of the patient specifically would be asked. Lastly, after gathering 30 responses for meal timing for AD patients, the researcher calculated the mean of the age of AD patients in order to match Non-AD patients' ages with AD patients and thus make the two groups comparable as AD is influenced heavily by age. Non-AD individuals' datas were collected in the same way AD patients' datas were collected to once again make the groups comparable so that they both have similar environments. Fifteen Non-AD individuals were sampled with connections to the researcher, and Fifteen Non-AD individuals were asked in the welfare center for elderlies in Gwangjin-gu, Seoul. Snowball and convenience sampling were

utilized to gather AD patients, a type of nonrandom sampling where the subjects are easily accessible to the researcher, whether based on geographical nearness, time availability, or willingness to participate. (Etikan et al, 2016). Considering the possibility of AD patients having difficulty recalling meal times due to their symptoms and the potential for exacerbating their condition through inquiring about past information about themselves, caregivers who had lived with them in the past before symptoms appeared responded to the questionnaires. This approach allows for more accuracy since AD patients may have difficulties recalling past meal times before the onset of symptoms. Moreover, surveying AD patients raises ethical concerns as the nature of the questionnaire regarding past information could be confusing or distressing for AD patients.

Questionnaire Design

This study's questionnaire (Appendix 5) was constructed to measure the two prerequisite components of circadian alignment: Fasting and Brightness. The questionnaire was structured in the following manner: First, the demographic information, including age and sex, was used to see potential factors that may influence the result of the study. Subsequently, three multiple choice questions on meal times when they consumed their first, second, and third meals before their onset of symptoms were asked to assess the participants' past meal times before their onset, as AD starts long before symptoms appear. Meal times were divided into categories for approximate meal times rather than exact ones. Specifically, "early morning" meant approximately (6 am to 8 am) and "late morning" (9:00 am - 11:00 am), "early afternoon" (12:00 am - 2:00 pm), "late afternoon" (3:00 pm - 4:00 pm), "evening" (5:00 pm - 7:00 pm), "late night" (8:00 pm - 5:00 am). Although AD starts approximately 25 years before the onset of symptoms, mealtimes were asked before the onset, with the note to consider meal times approximately 25 years. The reason for this choice was because specifically asking 25 years before may confuse the participants and also because there are no evidence that AD starts exactly 25 years before the onset of symptoms. Thus, the questionnaire aimed to ask about approximate past meal times. Because there may be variations from person to person, one of the multiple choices also was "Did not typically eat a third meal," and another question was asked if they had an additional meal beyond the third meal and if that was during the dark. The question allowed the researcher to analyze if one's usual meal times consisted of dark meal times and also one's full meal time duration. The four questions on meal timings allowed the researcher to measure the participants' fasting duration. Meal times were inquired as asking direct questions about their fasting duration may introduce nonresponse bias and an absence of response due to uncertainty (Sedgwick, P. Et al., 2014). Their perceptions of brightness in their frequent meal times were asked for their first, second, and third meal times as the brightness itself couldn't be analyzed through meal times due to differences in individual days.

To measure if the participant's times consisted of the two essential components of circadian rhythm, the participants responses were categorized into groups. Participants who responded dark for either of the four questions, including the one that asked about extra meals during darklight hours, were categorized as Dark(nocturnal) diners, and the rest of the

participants were categorized as Bright(diurnal) diners. In measuring whether participants' meal times consisted of fasting or not, criteria of approximately 16 hours of fasting, fasting time of 14h to 18h was used, which is suggested as “time-restricted feeding” that aligns with the circadian rhythm (Charlot, A. Et al., 2021). According to the data, all except Em-Ev, which is 6-8 am to 6-7 pm, approximately getting at most 14 hours done was the only group who didn't get fasting done, others, for example, Im- la (late morning to late afternoon 9-11 am to 3-4 pm) which got at most 17 hours of fasting, was all included as ones that did fasting. Although the participants responded that they ate their meal from early morning to early afternoon, they were marked as a non-fasting category if they responded yes to the question of eating during the dark. Only participants who responded that they only ate from evening to late night would have been marked as a fasting category when they answered yes to the same question. However, no one fell into this category for the context of this study. Questionnaires were constructed in two separate languages, English and Korean.

Statistical Analysis

Since the study's predictor variable is the meal times of the individuals, which was converted to categorical variables, and the outcome variable is categorical (presence of AD), a non-parametric test was chosen. This is useful because non-parametric doesn't require the sampling distribution to be expected. Given the complexity and potential variability in dietary habits and AD, the normality of the sampling distribution can't be ensured. This data was analyzed using 2x2 contingency tables that separate each subject into one of four categories based on two factors, each with two possibilities (fisher's exact test) for the categorical variables to see the hypothesized association between the two: AD and meal timing. Fisher's exact test is a nonparametric test for categorical data but can be used when the chi-square test cannot, such as small sample sizes. This test is used when comparing 2 data sets that create a contingency table and tests the association between the two criteria (Winters, R., Et al., 2010).

4. Results

Demographic Information

The first three questions on Participants' Demographics (age, sex, ethnicity) in the survey were taken to understand any possible factors that could influence whether the participants have AD or not. Each question helps provide a basis for a better understanding of which individuals are at more risk than others. The data table below includes information for each participant's demographic question answered.

Figure 2. AD patient's and Non-AD participants' demographic information

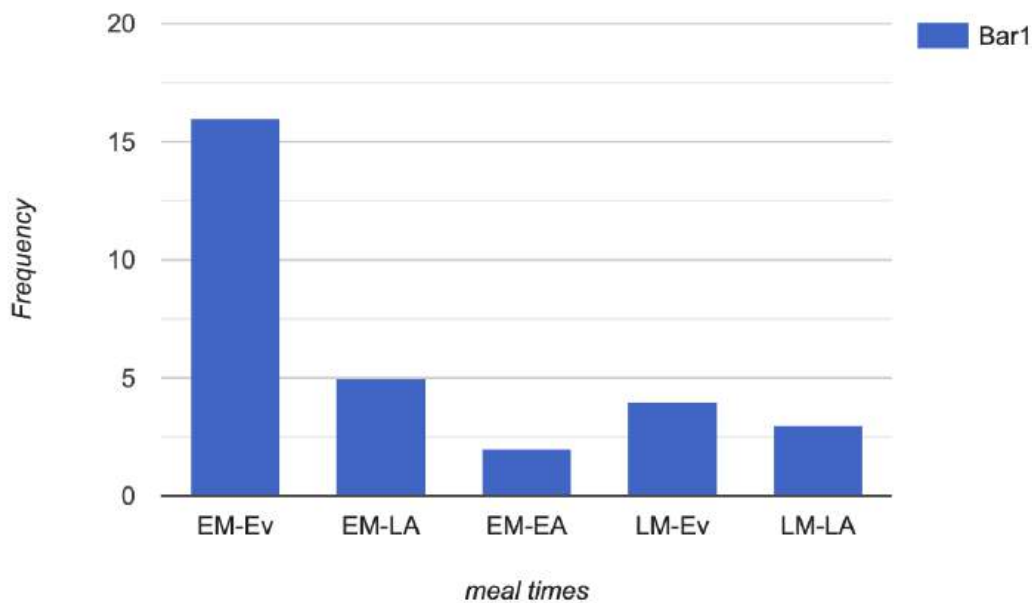
AD patient's and Non-AD participants' demographic information (n= 60)

	gender		age		Ethnicity	
	male	female	mean	standard deviation	Caucasian	Asian
AD group	11	19	85.4	7.2	1	29
Non-AD group	15	15	84.55	6.4	0	30

Meal time results: Fasting

As seen in the literature review, one of the important links between meal timing and AD is related to fasting. Cellular maintenance is crucial for removing accumulated glucose and fat, as observed in rodent studies in which the absence of fasting increases inflammation (Lesica, N. A. Et al., 2017). The inflammatory processes are suggested as the pathogenesis of AD (Lourida, I. et al. 2013). However, asking for the participants' fasting time may result in non-response bias and potential inaccuracy. Thus, the approximate meal "times" were asked to calculate the duration of their fasting hours. Below is a figure of participants' approximate eating window or meal time duration based on their responses to three questions regarding meal time categories for their first, second, and third meals.

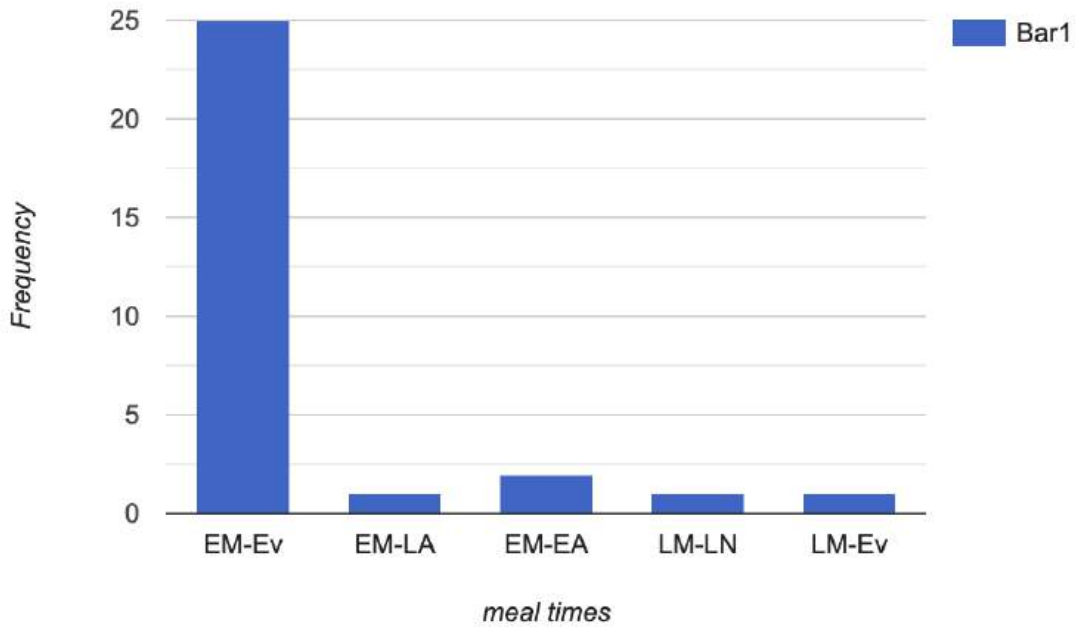
Figure 3. AD patient's approximate mealtime window (bar graph)



Note. Em to Ev meant early morning to evening (6~8 am to 6~7 pm), EM to LA meant early morning to late afternoon (6 ~8 am to 3~ 4 pm), Em to Ea meant early morning to early afternoon (6~8 am to 12~2 pm), Lm to Ev meant late morning to evening (9-11 am9-11 am to 5-7 pm), Lm to La meant late morning to late afternoon (9~11 am 9~11 am to 3~4 pm).

Since all groups except em-ev approximately showed fasting for at least 14 hours, all of them except the EM to EV group were categorized into fasting factions. Next, the respondents who responded yes regarding the question if one ate additional meals/snacks during dark after their third meal were categorized into the nonfasting group.

Figure 4. Non-AD individual's approximate meal time window (bar graph)



For Non-AD individuals, a group of LM-LN, meaning meal duration from late morning to late night, was present. Thus, all except the Em-Ev and Lm-Ln groups were categorized as fasting groups. Non-AD individuals who responded yes to the question regarding additional meals or snacks during the dark were also categorized to nonfasting groups. Figure 4 outlines the data of groups.

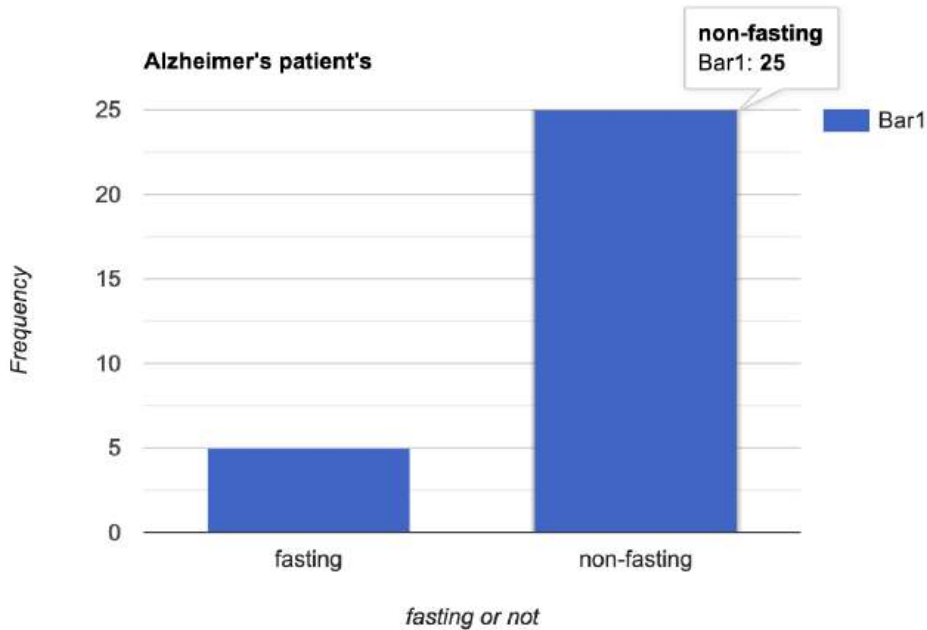
4.3 Meal time results: Fasting results

Figure 5. Frequencies of AD Patients and Non-AD individual's fasting frequency (two-way table)

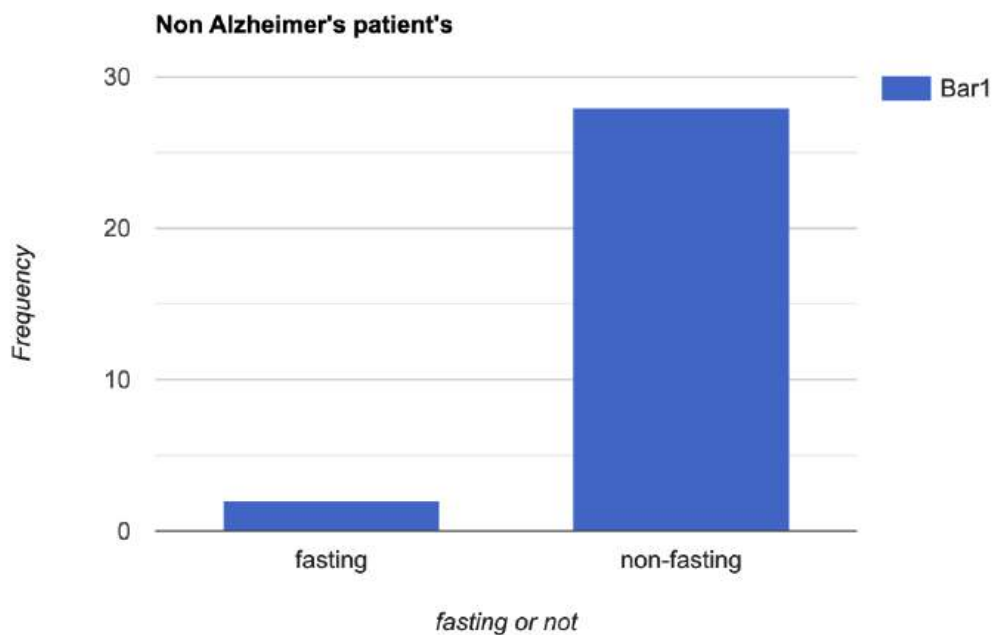
	AD patients	Non-AD individuals	Total
Fasting	5	2	30
Nonfasting	25	28	30
Total	30	30	

Note. From the graphs above frequencies, respondents were categorized as fasting or nonfasting, depending on their group.

Figure 6. Frequencies of AD patient's fasting frequency (bar graph)



Note. Non-fasting individuals for non-AD were 25, and Fasting individuals were 5.
Figure 7. Non-AD individuals fasting frequency



Note. There were 28 non-fasting individuals for non-AD and 2 Fasting individuals.

Meal time analysis: Fasting

When comparing figures 6 and 7, more presence for fasting was shown for AD patients; however, the overall data was not significantly different between AD patients and Non-AD individuals. A Fisher’s exact test was conducted to see the association between individuals who

fasted in the past and those who didn't and individuals who have had AD and those who don't have AD (Appendix 8). P- value, the probability of observing the same difference in fasting and nonfasting between two groups, AD patients and Non-AD individuals, when assuming that the fasting phase is not correlated with the presence of AD was 0.424, meaning that it wasn't significant. Since the P-value of 0.424 is more than the alpha, which is 0.05, the null hypothesis failed to be rejected. No convincing evidence has been found that AD and Fasting are associated.

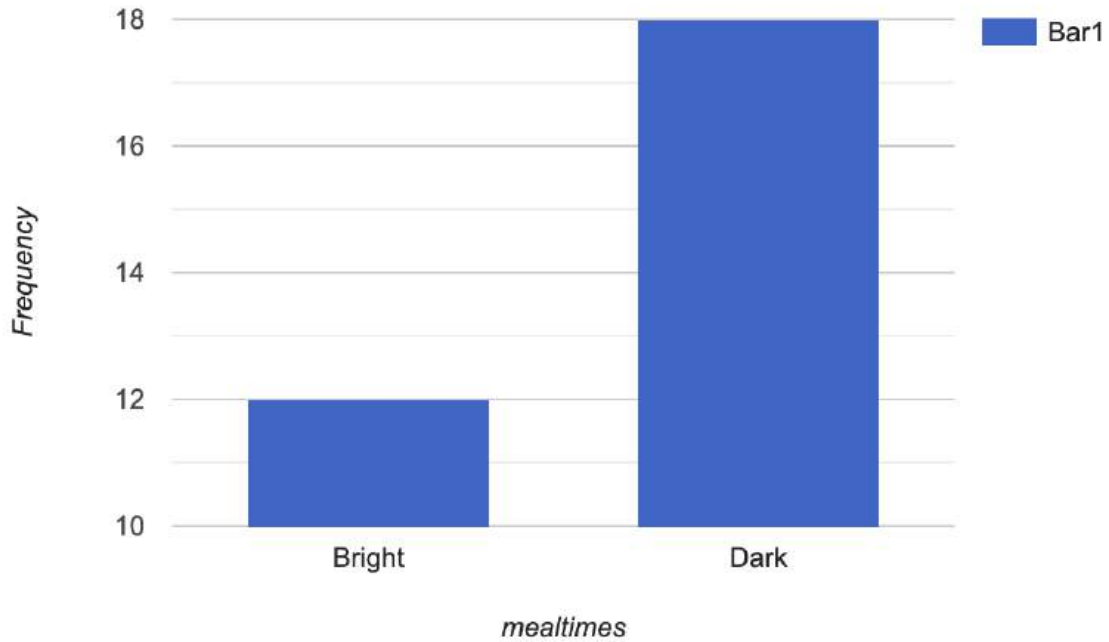
Meal time results: Brightness

The literature review suggests that humans are biologically predisposed to eat during daylight. However, the precise times couldn't account for the brightness of meal times due to sunrise and sunset variations influenced by seasonal changes and daily fluctuations. Therefore, to address the brightness of meal times, participants were asked to recall the light conditions during their meal times rather than using quantitative data of meal times. The survey included multiple choice responses such as "Dark," "Becoming brighter," and "Becoming darker." For analysis purposes, any response indicating light presence, including "Becoming brighter" and "Becoming darker," were categorized as Bright groups. Furthermore, since individuals may have additional meals or snacks after their third meal, participants were asked about post-third meal consumption during nighttime. If answered 'yes,' the responses were counted as 'Dark' category consumption, as outlined in Figure 8.

Figure 8 Meal timing results: Frequency of individual's nocturnal/ diurnal diners

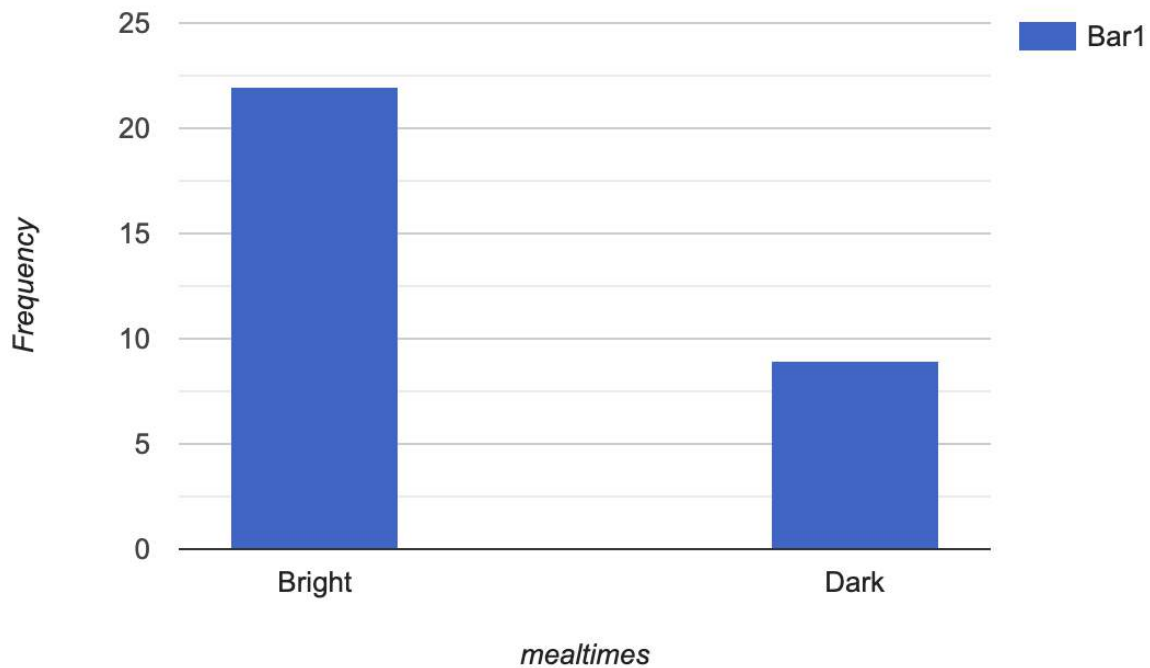
	AD patients	Non AD individuals
Bright (diurnal)	12	22
Dark (nocturnal)	18	9

Figure 9 Frequencies of AD patients nocturnal/ diurnal diners. (bar graph)



Note. More (18) people for AD patients ate during the Dark than those who ate during the Bright (12).

Figure 9 Frequencies of non-AD individuals nocturnal/diurnal diners (Bar graph)



Note. There were fewer (9) people for non-AD individuals who ate during the Dark than those who ate during the Bright (21).

Meal time Analysis: Brightness during daily eating times

As seen in Figures 8 and 9, a notable proportion of AD patients had refrained from eating during daylight hours, while many Non-AD groups ate during daylight hours. The same test, Fisher's exact test, was conducted to see the association between an individual's mealtime brightness and one's presence of AD (Appendix 9). P-value, the probability of observing the same difference in eating during bright and dark between two groups, AD patients and non-AD individuals, when assuming that brightness during meal times is not correlated with the presence of AD, was 0.0370. Since the P-value of 0.370 is lower than the alpha of 0.05, the null hypothesis could be rejected. There is statistically significant evidence of an association between eating during the dark and the presence of AD.

Discussion

As identified in the result analysis, no significant association was observed between presence of fasting and the presence of AD. However, brightness during daily eating times had a statistically significant relationship to the presence of AD. Since the p-value, the probability of obtaining the results of the study that a substantial portion of AD patients having nocturnal food consumption when assuming that no association exists is actual was smaller than alpha, it was concluded that there was an association. Consistent with previous studies, results from this research suggests that food consumption during different light hours has a relationship to the development of AD. As seen in the literature review, eating during bright could be related to a lower risk of AD through several potential mechanisms, including high levels of insulin, insulin resistance, and obesity. The brightness phase in meal times has been associated with improved insulin sensitivity and lower levels of insulin as well as a low risk of obesity (Lowden A. Et al., 2010). The pleiotropy of the advantageous effects of food consumption during daylight hours supports the biological plausibility of its potential role in reducing AD risk. This research identified the first potential relationship that hasn't been explored in AD and dietary habit research. Therefore, this research serves as a first indication of a possible connection between meal timing and AD. Moreover, this could be an implication for nocturnal eaters, including shift workers, for their risk of AD. More research could be done specifically on nocturnal eaters with AD, as there have been other researches, as stated in the introduction, on the correlation between shift workers and common metabolic diseases. (Mattson, M. P., Et al., 2014)

Verifying hypothesis

The original hypothesis in this study posited that presence of AD would be correlated with both important components of meal timing, presence of fasting and brightness. Though there was, in fact, a significant association between light hours in the presence of AD, there wasn't a significant association between fasting. Looking back at the literature review, the results of this study contribute to the ongoing debate of whether fasting or brightness is essential in regulating the circadian rhythm (Lesica A. Et al., 2017). This study indicated that fasting is just a phenomenon that comes with aligning to the circadian rhythm as individuals naturally abstain from eating during the dark. However, fasting alone, in the absence of appropriate light exposure, may not be sufficient. The biological clock, which historically regulated by human ancestors is primarily synchronized by light changes, suggesting that circadian rhythms are

predominantly determined by light. (Lowden, A. Et al., 2010). Physiological evidence, such as the oscillator in the central nervous system that determines the temporal organization of eating synchronized by the changes in light and darkness, also suggests that circadian rhythm is determined solely by brightness. Thus, this study's result adds to the research field that not fasting according to the circadian rhythm does not increase the risk of AD. These results also connects back to studies that found the effects of fasting being heterogeneous. One of the reasons that could account the controversial results of the fasting practices is that fasting solely do not consider the importance of matching the food intake timing with the circadian clock (Charlot, A. Et al., 2021). Thus, fasting may only show beneficial effects when aligned with the circadian clock. Further research has to be done on the beneficiary effects of fasting.

Limitations

It is worth noting, however, that several factors may affect the extent to which the results could be generalized. First, although the researcher didn't limit the scope of her study, the researcher got responses from the majority of Korean elderly. As the study couldn't account for all the AD patients of different ethnicities and people living in other places, the result could not be generalizable to the global population. Although there was one white or caucasian participant, it was an extremely small representation of that population, thus the generalizability could only be limited to Koreans. As a correlational study was conducted for ethical reasons, it is difficult to draw from these results whether or not eating only during the bright times daily impacts the presence of AD in one's future life. The result of the study could potentially have occurred because those who tend to eat during the bright days had a healthy lifestyle and thus didn't develop AD. This study's results don't tell the causation of AD; instead, they suggest a potential connection that could be studied even further. Nonetheless, the current ex post facto research has particular strengths: it can help analyze a cause based on an effect being studied, and the researcher's opinion does not affect the result of the research. (Akinlua, S. Et al., 2019). As the nature of AD being multifactorial, multiple factors including low level education, midlife hypertension, midlife obesity etc. all are potential variables that may affect the result of the study; however to reduce the complexity of the questionnaire and potential distress that might bring to the participants, the study didn't include questions on all the different factors. Therefore, this paper was unable to assess the effects of different potential factors that may have affected the result of this study.

Further implications

This study contributes to understanding the correlation between AD and meal timing, addressing the gap in existing research. However, as stated in the limitations, the implications may predominantly apply to Korean people. Further research utilizing diverse populations and observation methods is necessary to broaden the implications of these preliminary findings. Other researchers can widen the scope and obtain meal time data for not only Koreans but also in diverse settings. It could be scientifically meaningful to those with the time and money to observe people for several years could conduct a longitudinal study to track different aspects of people's lifestyle such as exercising and other factors which could potentially add up to AD

while tracking meal times as well. Moreover, research could be done to add to the literature review, the effect of disruption in the circadian rhythm to the insulin seems to be correlated with the degeneration of AD; further research can see the varying severity of AD patients with the correlation to their eating environment (bright or dark). The result of this study also raises the question of which of the factors of circadian rhythm is more important, fasting or brightness. Further studies should be conducted examining the biological processes of insulin levels or inflammatory processes in the relationship to cognitive processes of AD.

Considering an increase in nocturnal eating and a rise in AD these conclusions could have global effects and advance our understanding of the mysterious disease. The results of this study would allow predictions for Korean people to what extent their meal timings may be associated with their risk of AD in the future, further suggesting dietary implications for prevention of AD. Moreover because modern people tend not to eat according to the circadian rhythm, this study would further allow people to acknowledge their risky behaviors and allow more effective strategies for AD prevention. Given that there wasn't research on the direct relationship between meal timing and AD, this study could indicate the first step of the study of the two. In this study, it was found in the Korean elderlies a link to developing AD, further study could expand and see more applicable results that could help individuals prevent AD.

Overall, because the first study investigating the association of AD and meal timing was done, more helpful lifestyles are added to the field. This study contributes benefits to the field of AD prevention as AD is a multifactorial disorder in which early-life prevention efforts are crucial.

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Video Upscaling and Enhancement Utilizing Super Resolution Performed by Artificial Intelligence By Krishay Bhole

Abstract

Recent advancements in AI-driven super resolution have led to significant improvements in overall image quality. This paper expands the range of super resolution's abilities by discussing a method to upscale the resolution and enhance the quality of a live video feed in real time using several pretrained AI models, such as EDSR, ESPCN, FSRCNN, and LapSRN. Through experimentation, areas in which improvements were made include edge quality, pixelation artifacts, and overall image status. When the Mean Squared Error (MSE) loss function was used to evaluate pretrained model performance, there was a 15.84% maximum improvement in the quality of the images when compared with bicubic interpolation. We also perform an analysis of the models covering improvements to be made, limitations present, and steps for additional experimentation.

Introduction

Super resolution is an image processing technique that enhances an image by generating additional details beyond the original pixel information using artificial intelligence. Super resolution algorithms are not equivalent to bicubic interpolation, which is the standard algorithm for image upscaling. Bicubic interpolation involves using weighted averages of neighboring pixels to increase the size of an image without creating new details or improving image quality to the same extent as super resolution. Interpolation reveals pixelation artifacts when the output image is upscaled, making the image perceived to have reduced clarity. On the other hand, super resolution improves the perceived quality of the image when upscaled using deep learning tactics. Unlike bicubic interpolation, there is little to no loss in perceived quality, and the average pixel density often increases.

While super resolution has been explored, there has not been significant involvement in applying the technology to videos. Super resolution-based video enhancement is a useful tool in medical imaging, security and surveillance, and autonomous vehicle object detection; each field of study would only benefit from increased video quality. Therefore, the focus of this paper is to investigate the topic of upscaling a low-resolution video feed into a stream that is more comprehensible and detailed.

The employed technique is to take a single frame from a video feed and apply super resolution before outputting the frame in a separate window. The frame rate of the video has been reduced to decrease the need for computational resources. We employ pretrained super resolution models to be the backbone of our program. Additionally, we analyze each model's strengths, weaknesses, and areas for improvement in subsequent sections.

Methodology

In set intervals of time, a frame is selected from the continuous video feed. We employ intervals to select frames to minimize the need for processing power. Using the model that was

initially loaded using OpenCV's deep neural network super resolution module [6], we then perform super resolution on the desired image. Bicubic interpolation is simultaneously performed with the same loaded image for comparison and analysis. Both images are outputted for reference. In all tests, the same image is used for consistency across tests. The pretrained models used are EDSR [3], ESPCN [4], FSRCNN [1], and LapSRN [2]. Initial results from the pretrained models without DnCNN are shown [Figure 1]. Significantly visible changes are highlighted with red outlining.

One more addition to the super resolution framework is a denoiser named DcDNN [5] which reduces pixelation artifacts. The denoiser is particularly valuable when the video feed contains human subjects or when the subjects are obscured by background interference. The denoiser is used right before applying super resolution. However, this may result in reduced edge quality, so the quality of improvement provided by the denoiser is subjective. Therefore the decision to incorporate the denoiser is based on the quality of the input video feed and whether noise is initially present.

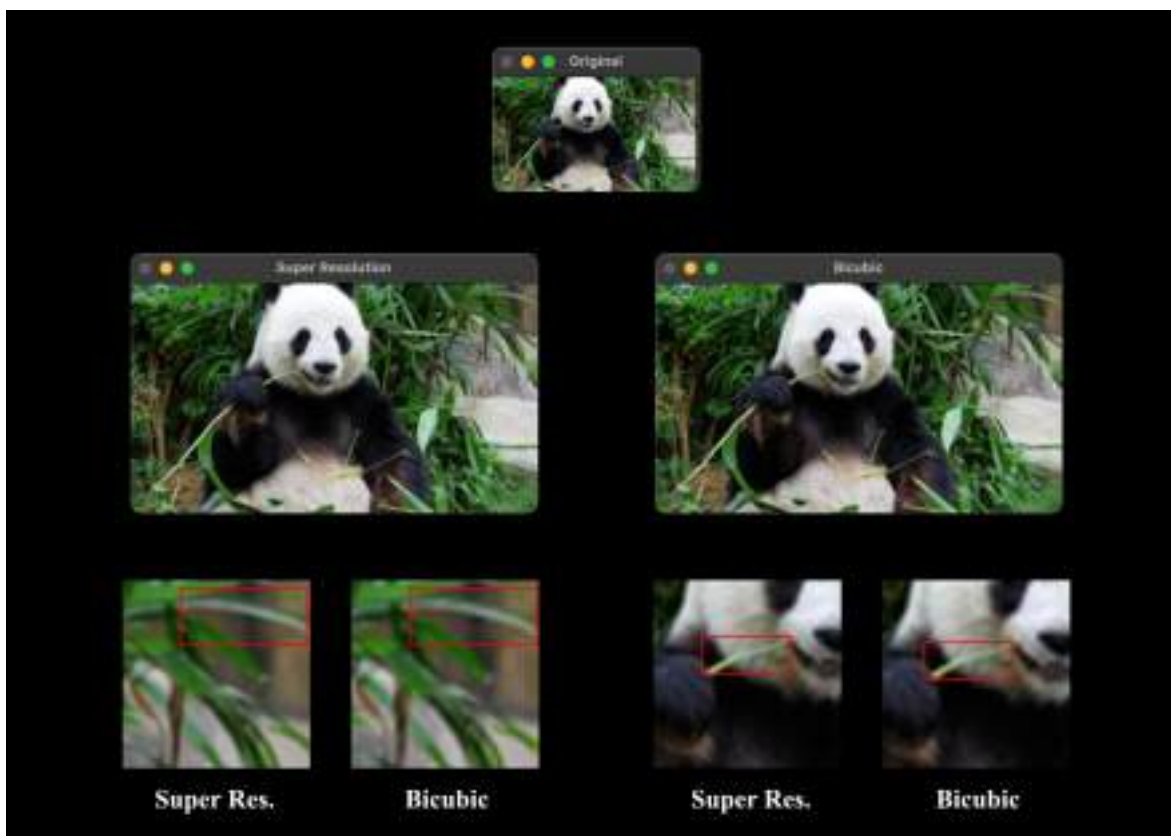


Figure 1: Initial results from EDSR for a 195x110 image of a panda.

Results

All models, except EDSR, used up approximately the same amount of time to apply super resolution to the image [Figure 2].

EDSR and LapSRN appeared to have the most significant improvement based on the quality of the images shown [Figure 3]. However, a quantitative data analysis is necessary to confirm these claims. This analysis can be done with the use of loss functions, which compare multiple images with certain algorithms. The loss function used was Mean Squared Error (MSE), which is defined as follows:

$$MSE = \frac{1}{mn} \sum_{i=0}^{m-1} \sum_{j=0}^{n-1} [I(i, j) - K(i, j)]^2 \quad (1)$$

$$= x \quad (2)$$

MSE is performed on a 2D grid with $m \times n$ dimensions. For each point, the difference is computed between an actual value $I(i, j)$ and a predicted value $K(i, j)$. Subsequently, the sums of the squared of these differences are summed across all data points before then being divided by the total number of points to get an average. Performing MSE results in a whole number output, x , such that $x \in [0, \infty)$ (equations 1 and 2). A larger x indicates less similarity. This metric is particularly useful in model analysis and evaluation because it provides a single scalar value that will provide the overall change in our new image.

To find the "percentage improvement" between two images, a specific metric is used involving MSE. This metric consisted of finding the MSE between the low-resolution image (LR) and the super resolution image (SR) along with the MSE between the LR and the resulting image of bicubic interpolation (BC). Mean squared error

Average speeds of super resolution models from five trials upscaling a 195x110 image to a 390x220 image on an M1 chip.

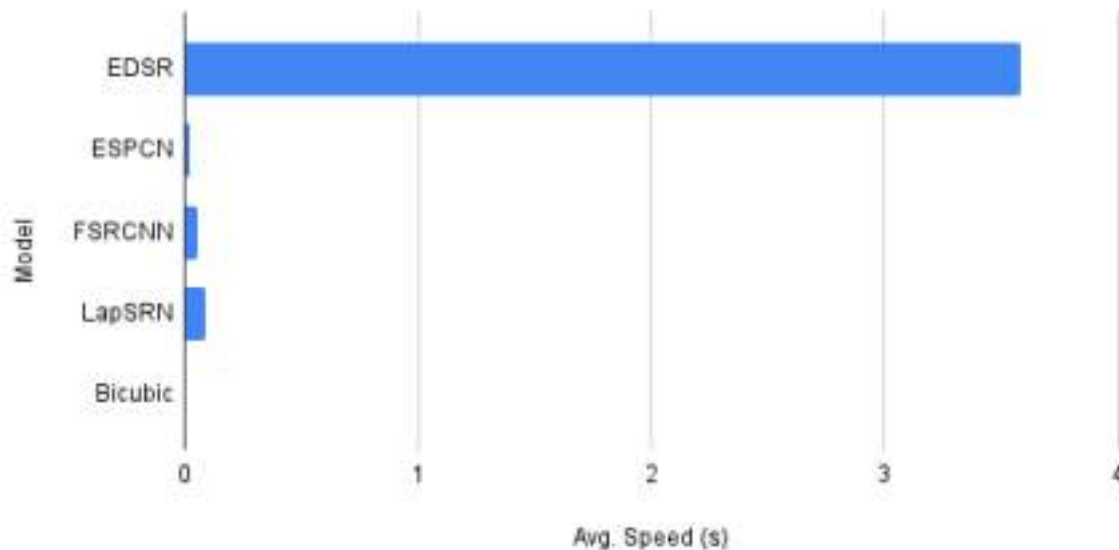


Figure 2: Comparing the average computation time for the pretrained models.

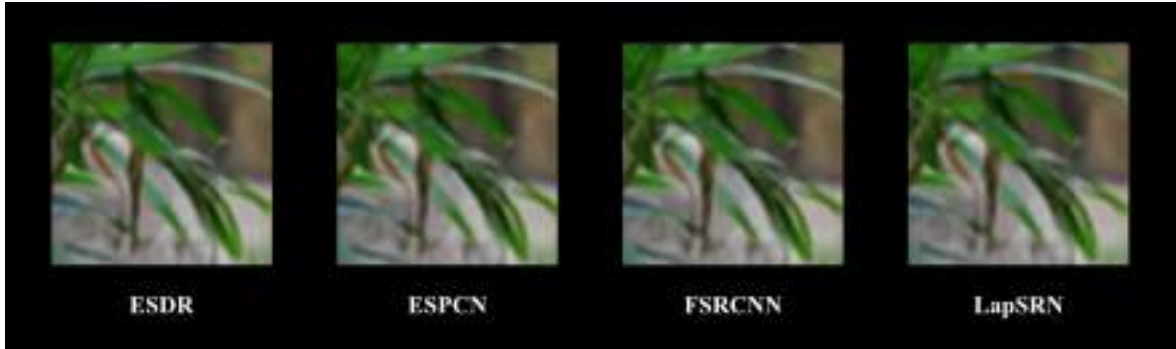


Figure 3: A closer look at the four different pretrained models.

is represented as a function $MSE()$ that takes in two images for parameters and outputs a whole number. After obtaining MSE results, an equation similar to relative error was used; the result is an expression for IMP, which is the improvement as a percentage (equation 3).

$$IMP = \left| \frac{MSE(LR, BC) - MSE(LR, SR)}{MSE(LR, BC)} \right| * 100\% \quad (3)$$

This metric allows for simple comparison between the different super resolution models that we utilized. Calculating the percentage improvement for each model with the image of the panda provided the results shown in Figure 4. Based on MSE, LapSRN demonstrated the greatest magnitude of improvement at 15.84%, followed by EDSR at 10.58%. ESPCN and FSRCNN exhibited 5.09% and 1.24% improvements respectively. This confirms our initial claims of LapSRN and EDSR being the best performing models based on super resolution image output.

Percentage improvement on a 195x110 image upscaled by a scale factor of two by several super resolution models.

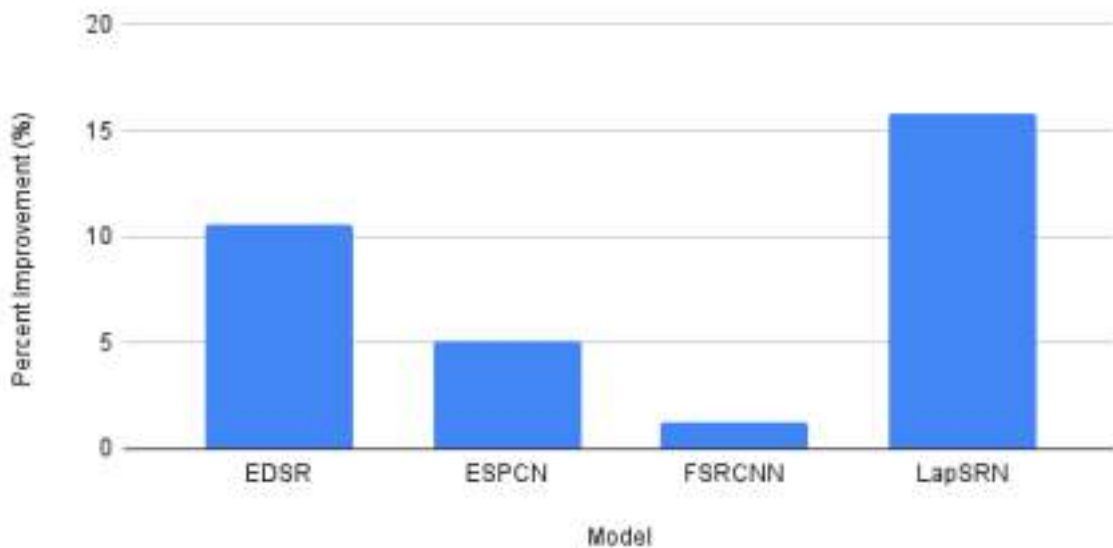


Figure 4: Comparing the average percentage improvements for the pretrained models.

With these results, LapSRN was chosen for applying super resolution to the video, as it was one of the faster models and had the greatest percentage improvement. EDSR was the runner-up in terms of percent improvement, but its computational speed was slow; the video feed would significantly lag and frames per second would be low if EDSR was used.

After utilizing DcDNN to reduce noise and subsequently implementing pretrained models for super resolution, the final output of the technology described in this paper resembles the one depicted in Figure 5. The figure shown stems from a live video feed.

Discussion

In this study, we thoroughly explored the pre-trained model approach for upscaling and enhancing low resolution videos.

The main limitation of the models was processing speed. The computation time reduced the frames per second rate to approximately 0.73 FPS when performing super resolution on a single frame at a time. Nevertheless, there are visible changes, signifying that the method properly improves the quality of the video. As shown in Figure 5, the original image visually indicated more pixelation artifacts. The results from the methods explained in this paper are demonstrated in the two images located on the right side of the figure. Having lowered the

4



Figure 5: Final comparison between omitting and using a denoiser for super resolution.

visibility of the pixels, they feature clearer details. The denoiser smooths out any edges and removes extra noise if present in the image, but some loss of detail is present.

Areas where improvements need to be made include utilizing different loss functions to objectively analyze model results, such as adversarial and perceptual loss, which are beyond the scope of this paper. They may introduce new ways in which different pretrained models show their own strengths. Another place for improvement lies in the efficiency of

super resolution algorithms. With our testing, we were only able to achieve a low frame rate, which would be less practical for real-world use. Optimizing model architecture, leveraging better hardware, and exploring other super resolution processing techniques are all categories for further investigation. Another possible technique is to analyze groups of frames and possibly even generate frames using AI to increase frame rate. A CNN approach is a possible route to explore, as well. While the initial CNN model training needs to be extensive to cover a wide variety of videos, the result is bound to be impressive due to the recent increased potential for CNNs.

Conclusion

Super resolution for video serves as a powerful device that can be utilized in surgical medicine, security and surveillance, or other fields of interest. Even though computer imaging and super resolution is constantly evolving, there has not been much work devoted towards using such tools in video and film. This paper marks the initial step towards a series of advancements in a field with great potential for enhancing video quality. As technology continues to advance, the integration of super resolution in the realm of video and film holds promise for revolutionizing visual content and raising the bar for quality in many industries.

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A Comparative Analysis of Microfinance in Bangladesh and South Africa By Anish Chowdhury

Abstract

This research paper is an inquiry into why South Africa's microfinance system has seen a lackluster performance compared to the nation of Bangladesh. Using the findings of previous literature, I developed a nine-point rubric based on density, governmental support, and internal management to examine a broad comparison between the two nations' microfinance systems. From this rubric, Bangladesh scored a seven out of nine, while South Africa scored a five out of nine. Major differences that the rubric highlighted between the two countries included internal management and population density. This paper aims to lay the groundwork for a broad metric that can be used to analyze a variety of countries' quality of microfinance systems.

Background:

Introduction: After the 1971 Bangladesh Liberation War, Bangladesh had an abnormally low GDP per capita of about 113.60 USD (Basu, 2021). Comparatively, the World Bank reported that neighboring India's GDP per capita was over triple the size, at 361 USD ("GDP per capita (current US\$) - India," 2024). Today, the GDP per capita of Bangladesh has increased by over 2,300% to 2,687 USD, overtaking India ("GDP per Capita Bangladesh," 2024). But it is not just GDP per capita; it is also the literacy rate which has jumped from 18% to 81%, and also life expectancy, which has increased from 45 to 74 ("Life Expectancy at Birth, total (years)," 2024). The development of Bangladesh as a nation is due to a plethora of factors, including the introduction of the garment industry and export-oriented industrialization. Many prominent scholars such as Nobel laureate Amartya Sen, believe non-governmental organizations (NGO) microfinance institutions have helped industrialization and development (Sen, 2023). Microfinance is defined by former Senior Advisor of the World Bank, Ira W. Lieberman, as a loan given to people and small businesses who normally do not have access to loans under the status quo (Lieberman et al., 2020). Microfinance institutions are often abbreviated as "MFI" (Lieberman et al., 2020). Prevalent MFIs include the Grameen Bank, Bangladesh Rural Advancement Committee (BRAC), Pokisha, and the Association for Social Advancement (ASA).

In an interview I conducted with the Senior Director and Head of Microfinance of BRAC Bangladesh, Arinjoy Dhar, he explained that most microfinance involves people who live in poverty applying for loans ranging from 50-500 USD (Chowdhury, 2024). With these "micro-loans", they can develop their skills by buying capital such as yarn, a food cart, or livestock (Chowdhury, 2024). From the profits of these ventures, the borrowers can pay back their loans with an interest rate of a maximum of 24% (Naren, 2022). This method and structure have been widespread among Bangladeshi microfinance institutions, including

Grameen Bank, which have uplifted five million people out of extreme poverty between 2000 and 2010 (Khandker & Samad, 2014).

The United Nations explains that although Bangladesh continues to face many problems, which is emphasized by its low human development index of 0.610, it has surpassed its expected growth in all metrics over the past fifty years (“New directions for human development in Bangladesh,” 2023). The Industrial Development Leasing Company points to Bangladesh in 2021 having a 98% repayment rate with regards to microloans within the country (Rakeen, 2021). The same method Bangladesh attempted has been tried in other parts of the world including South Africa. However, South Africa has failed to implement microloans successfully. As apartheid ended in 1990, many in South Africa were optimistic that the country’s economic output, specifically in black communities, would improve dramatically. This is demonstrated by the fact that banks such as the Small Enterprise Foundation (SEF), Kuyassa Fund, South African Micro-Finance, and Marang Microfinance were established in South Africa post-apartheid (Rees & Volker, 2013). However, life expectancy did not increase at all, while economic growth remained stagnant and has seen a decrease of about 15% over the last ten years (“World Bank in South Africa,” 2024). Yet, the same principle ideas of microloans were used to increase economic productivity within the most economically disadvantaged communities. The vast majority of these efforts have failed and have often hurt financially disadvantaged people, causing 40% of South African micro-loan borrowers to be unable to repay their loans (Rees & Volker, 2013). The difference in loan repayment between the two nations leads to the following question: why has Bangladesh’s microfinancing system seen a much more dramatic level of success than South Africa's?

Literature Review:

To get a better understanding of the metrics regarding why certain microloan systems have been regarded as more successful, it is critical to look at past studies. Several studies have addressed how density has impacted microloaning systems. An earlier investigation looked at a case study within Niger in which Jose Perdosa, an official at the United Nations Development Program, and Quy-Toan Do, an official at the World Bank, looked at whether or not density impacts the success rate of microloans (Perdosa and Do, 2006). In the study, they compared 191 loan applicants and 163 corresponding clients and tested whether population density impacts loan repayment, the ability of banks to track loan repayment and the industry loan recipients who are asking for loans. The study found that areas of low population density are more likely to ask for loans in the agricultural sector (Perdosa and Do, 2006). In addition, both make the point that as distance increases the ability to monitor repayment loans has decreased significantly (Perdosa and Do, 2006). However, the research paper also makes the point that microloan recipients in low-density areas have “limited outreach” and therefore would “not be able to deliver enough cash to interest MFI” (Perdosa and Do, 2006). Another study conducted by Annabel Vanroose, a professor at Stellen Bosch University in South

Africa, concluded that high population density leads to greater success and attracts a greater possibility of microfinance institutions (Vanrose, 2008). In general, areas that have “high population density lowers the operational costs of serving microfinance” (Vanrose, 2008). Both the studies conducted by Perdosa and Vanrosa indicate that population density has an impact on the success rate of microfinancing. However, both studies fail to address other potential factors such as competition and governmental regulation that impact microfinancing’s success.

Studies have shown that a country's openness to competition is vital regarding microfinancing (Nina et al., 2022; Wondirad, 2020). Comparative research was conducted by assessing 67 studies done about specific microfinance institutions and concluded that “competition would positively impact MFIs” (Nina et al., 2022). Competition is determined to be crucial because institutions are incentivized to provide better loan terms and better service to gain borrowers. These positive impacts on microfinancing include, “better portfolio quality, broader client base, and stable profit” (Nina et al., 2022). Another study, in which a mathematical formula was developed to test the effectiveness of microfinance competition in India, concluded that competition spurs “MFIs to reach out to the very poor segment of the population and helps them to be sustainable” (Wondirad, 2020). Nina and Wondirad’s studies suggest that healthy competition must be maintained within the microfinance industry for a successful microfinancing system to exist. Therefore, if a single microlending institution dominates a country, the probability of success will be significantly lower. Although both studies were able to explain the private sector's necessary components adequately, they failed to mention the success of the public sector.

In addition to competition, it is necessary to review the impact that governments have on microfinancing systems. While examining case studies from South America, Asia, and Africa, Cécile Lampu concluded that “the state must offer a conducive regulatory and economic framework to allow private institutions, particularly microfinance institutions, to develop without constraint” (Lampu, 2000). Lampu also addresses that the public sector must have strong regulations by stating that “regulations should be disseminated and implemented beyond the strict enforcement of the regulatory frame” (Lampu, 2000). The reason why Lampu concludes that there must be governmental regulation when it comes to MFI is due to the possibility of ultra-high interest rates on the most vulnerable people in society who lack basic education in reading and mathematics. Through Lampu’s assessment, it is clear that the public sector must work carefully with the microfinancing sector to ensure the success of the MFI system overall. In a macroscale through all studies, it can be concluded that although MFI is dependent on uncontrollable factors such as population density, there are many controllable factors such as the competition of microfinance institutions that exist within a country, and governmental aid and regulation.

It is also important to review the factors within MFI organizations that fuel their “success”. After looking at multiple models of multiple failed microlending companies it becomes clear that microlenders need to have the ability to generate profit to allow themselves

to grow (Lieberman, 2012). For MFI to be able to scale and have the ability to expand its market base, “introducing new technologies, and developing new products all rely on MFIs commercializing, generating a profit, and producing an acceptable return on investment to attract private investors” is crucial (Lieberman, 2012). Although microloans attempt to uplift the most vulnerable in society, Lieberman points out something often overlooked, which is that most successful microlenders' main goal is to make a profit. Microfinance institutions, just like any other bank, employ thousands of employees. However, many employees are often overlooked and many successful microfinance institutions invest in training employees as there is a positive “relationship between training needs assessment and employee performance in Microfinance Institutions” (Neema, 2021). In addition to Neema (2021), coinciding studies have shown that millions of dollars have been mismanaged/ within microfinancing institutions in Ghana (Barnie et al., 2013). Employee performance in large microfinance institution banks which impact millions of people's lives is as critical as their mistakes in handling funds can lead to millions of dollars in losses. Therefore employee training is critical. Overall on a micro level, it is clear that microfinance organizations must attempt to make some type of profit while also maintaining quality through staff training and proper internal management. With all this information, it is clear that microfinancing success is rooted in three things: uncontrollable circumstances such as population density as Perdosa and Vanrose found, governmental regulation/ability to promote private sector competition as Nina, Wondirad, and Lampu found, and internal management as Lieberman and Neema found. With this information, the exploration of the research gap of a comparative study between Bangladesh and South Africa can begin. This paper aims to compare the effectiveness of microfinance systems in Bangladesh and South Africa by examining three critical factors: population density, governmental support, and internal management

Method:

To determine the metric of success in microfinancing systems, a rubric must be created to determine what makes microloans successful. As mentioned in the peer review, three major categories cause a microfinance system to be successful or a failure: density, governmental support, and internal management of a microloaning banking system. The following information was used to create the rubric below:

	One Point	Two Points	Three Points
Density	The country's density is well below the threshold necessary to sustain a proper market base to sell products in all commodities sectors.	A country has a relatively high population density of between 600-1500 people per square kilometer.	A country has a very high population density of at least 1500 people per square kilometer. In addition to having a high population

	Overall the ability for businesses to prosper is extremely limited due to the lack of ability to create enterprises.		density, there must be a notable market space that exists within rural communities
Governmental support	There are no governmental regulations that prevent interest manipulation in the country. In addition, the country's policies regarding the banking system do not promote competition within the country. In certain aspects, governmental regulations may hinder the success of microfinance	There is a set interest cap within the country that regulates the microfinance industry. However, laws are loosely enforced and some banks still manipulate consumers through debt hedging/unfair interest. The country neither hinders nor supports competition within a country.	The government has assigned a key emphasis on consumer protection and has tightly capped interest within the country. In addition, the country fosters governmental policies that allow competition among microfinancing institutions.
Internal Management	The most prominent microfinancing institution/institutions lack support within their system that allows loans to be delivered quickly and without hindrance. There is an emphasis on profit and “unethical” behavior does take place which places. Companies are not	The internal management within prominent microfinancing institutions ranges. Certain microfinancing organizations have a proper and secure structure that can deliver loans at a fast pace. In addition, these organizations do not manipulate consumers and set	These microfinancing organizations have a proper and secure structure that can deliver loans at a fast pace. In addition, these organizations do not manipulate consumers and set fair expectations. It is also critical for these prominent organizations to not give loans to

	doing proper vetting of loans which can be seen if majority loans are being used inappropriately by borrowers.	fair expectations. However certain microfinancing institutions lack support to deliver loans and take part in “unethical” behavior.	unfeasible ideas.
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Analyzing data regarding density is easy to acquire by looking at governmental censuses provided by both South Africa and Bangladesh’s governments. In addition to looking at governmental censuses, it is essential to examine where microloan distributions took place. Finding areas where microloans took place in South Africa can be acquired by auditing “the Small Prize Enterprise Foundation” and “Marang Financial Service Women’s Development Business” is essential in the study. In terms of Bangladesh, BRAC, Grameen, International Food Policy and Research Institute show the type of area where the majority of microloans take place. When reviewing a country's governmental support, one key category is to see if governments can regulate the percentage of interest that MFI can place on their client. In addition to interest, it is key to examine whether a nation can maintain a healthy competitive environment to exist in regards to the microfinancing system. A metric that shows whether a government properly takes care of their country is examining specific laws that exist within the country regarding microfinance, and evaluating laws regarding consumer protection which help prevent pricing manipulation. Finally, in regards to the third row of the rubric, it can be examined whether the five most prominent microfinancing institutions have been able to successfully maintain internal policy that minimizes the risks of loans being misused by borrowers. Percentage of success and failure of on-time repayment can serve as a quantifying metric for this paper. The rubric serves as a summary comparison of the overall microfinancing system that exists in both South Africa and Bangladesh. It can overall determine what has been working properly, and what has been failing in both countries. Once the rubric is scored, it becomes easier to compare the two countries and see what both can take away and learn from each other.

Results: Using the rubric that was made it could be determined what caused the difference between South Africa and Bangladesh. The following are the results of each country which caused the difference.

Results for Bangladesh

	One Point	Two Points	Three Points
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<p>Density</p>	<p>The country's density is well below the threshold necessary to sustain a proper market base to sell products in all commodities sectors. Overall the ability for businesses to prosper is extremely limited due to the lack of ability to create enterprises.</p>	<p>A country has a relatively high population density of between 600-1500 people per square kilometer.</p>	<p>A country has a very high population density of at least 1500 people per square kilometer. In addition to having a high population density, there must be a notable market space that exists within rural communities</p>
<p>Governmental support</p>	<p>There are no governmental regulations that prevent interest manipulation in the country. In addition, the country's policies regarding the banking system do not promote competition within the country. In certain aspects, governmental regulations may hinder the success of microfinance</p>	<p>There is a set interest cap within the country that regulates the microfinance industry. However, laws are loosely enforced and some banks still manipulate consumers through debt hedging/unfair interest. The country neither hinders nor supports competition within a country.</p>	<p>The government has assigned a key emphasis on consumer protection and has tightly capped interest within the country. In addition, the country fosters governmental policies that allow competition among microfinancing institutions.</p>
<p>Internal Management</p>	<p>The most prominent microfinancing institution/institutions lack support within their system that allows loans to be delivered quickly and without</p>	<p>The internal management within prominent microfinancing institutions ranges. Certain microfinancing organizations have a</p>	<p>These microfinancing organizations have a proper and secure structure that can deliver loans at a fast pace. In addition, these organizations</p>

	<p>hindrance. There is an emphasis on profit and “unethical” behavior does take place which places. Companies are not doing proper vetting of loans which can be seen if majority loans are being used inappropriately by borrowers.</p>	<p>proper and secure structure that can deliver loans at a fast pace. In addition, these organizations do not manipulate consumers and set fair expectations. However certain microfinancing institutions lack support to deliver loans and take part in “unethical” behavior.</p>	<p>do not manipulate consumers and set fair expectations. It is also critical for these prominent organizations to not give loans to unfeasible ideas.</p>
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Results for South Africa

	One Point	Two Points	Three Points
Density	<p>The country's density is well below the threshold necessary to sustain a proper market base to sell products in all commodities sectors. Overall the ability for businesses to prosper is extremely limited due to the lack of ability to create enterprises.</p>	<p>A country has a relatively high population density of between 600-1500 people per square kilometer.</p>	<p>A country has a very high population density of at least 1500 people per square kilometer. In addition to having a high population density, there must be a notable market space that exists within rural communities</p>
Governmental support	<p>There are no governmental regulations that prevent interest manipulation in the country. In addition, the country's policies</p>	<p>There is a set interest cap within the country that regulates the microfinance industry. However, laws are loosely</p>	<p>The government has assigned a key emphasis on consumer protection and has tightly capped interest within the country.</p>

	regarding the banking system do not promote competition within the country. In certain aspects, governmental regulations may hinder the success of microfinance	enforced and some banks still manipulate consumers through debt hedging/unfair interest. The country neither hinders nor supports competition within a country.	In addition, the country fosters governmental policies that allow competition among microfinancing institutions.
Internal Management	The most prominent microfinancing institution/institutions lack support within their system that allows loans to be delivered quickly and without hindrance. There is an emphasis on profit and “unethical” behavior does take place which places. Companies are not doing proper vetting of loans which can be seen if majority loans are being used inappropriately by borrowers.	The internal management within prominent microfinancing institutions ranges. Certain microfinancing organizations have a proper and secure structure that can deliver loans at a fast pace. In addition, these organizations do not manipulate consumers and set fair expectations. However certain microfinancing institutions lack support to deliver loans and take part in “unethical” behavior.	These microfinancing organizations have a proper and secure structure that can deliver loans at a fast pace. In addition, these organizations do not manipulate consumers and set fair expectations. It is also critical for these prominent organizations to not give loans to unfeasible ideas.

Bangladesh Scored a 7/9 while South Africa scored 5/9

Bangladesh explanation

The reason why Bangladesh scored a 3/3 in the category of density was because Bangladesh has a total density of 1,180 people per square kilometer (“Population & Housing Census, 2022). Beyond just simply being one of the most densely populated countries in the

world, Bangladesh in general has a highly dense rural population. An average village in Bangladesh has about 2000 people which is well beyond the average rural population when compared to the results of the world (Population & Housing Census 2022, 2022). In the second row, Bangladesh receives a 2/3 as Bangladesh has strict guidelines when it comes to regulating microfinancing. Bangladesh is one of the few countries in the world to have a direct committee that oversees all NGO governmental bodies: the Microfinance Regulatory Authority (MRA). According to the MRA's website, the maximum interest rate in Bangladesh is 24% for all microfinance institutions ("Microfinance in Bangladesh," 2023). In addition, the MRA includes restrictions that help protect consumers. On the MRA website, it reports that there must be a 15-day grace period with no punishment for the first installment payment, and there can not be more than 15 Bangladeshi Takka (0.14 USD) worth of application fees/service fees for clients ("Microfinance in Bangladesh," 2023). In addition, the microfinance authority performs annual inspections to ensure that clients are not getting manipulated and provides licenses to ensure this.

Despite all these protective measures undertaken by the government, under the rubric, Bangladesh still only receives two points out of three. In 2021, reports by reputable third-party NGO such as the International Monetary Fund and Asiatic Society of Bangladesh explained that many unregulated microfinance institutions formerly or currently engaged in debt hedging by giving large sums of money to poor farmers without auditing the feasibility of their ideas (Islam and Karim, 2019); the purpose of the MRA is to stop these type of incidents from occurring. On top of this, many small microfinance organizations were not properly audited by the MRA government reports within the last five years in the two Bangladeshi districts of Barishal and Khulna European Journal for Business and Management reported (Shohel et al., 2020). These inconsistencies of results in Bangladesh by both third-party and governmental audits is what caused it to get a one-point reduction.

Finally, in the third category regarding internal loan management Bangladesh received a 2/3. For this part of the study, I focused on the top four largest microfinancing institutions: the Bangladesh Rural Advancement Committee (BRAC), the Association for Social Advancement (ASA), Basic Unit for Resources and Opportunities of Bangladesh (BURO) and Grameen Bank. These four banks handle the majority of microloans within Bangladesh. In my interview with the Senior Director at BRAC, Arinjoy Dhar, he explained that BRAC has multiple offices and has a chain of command that directly audits every loan and transaction that takes place within the government (X, 2024). When reviewing his claim it became clear that multiple third-party sources including USAID and Stanford University determined BRAC's internal structural hierarchy as sufficient to do collaborative work in regards to developmental economics (Charlifue, 2020). The ASA and BURO have both been audited by third and published full reports on their website and have been found there to be a comprehensive internal system. However, Bangladesh only earned two points under the rubric due to government audits which allege major misuse of funds by Grameen Bank. In a scathing report, the government alleged that Grameen Bank's structure and work culture encouraged

employees to aggressively seek funds and management internally promoted aggressive collection practices (“Summary of the Review Committee Report on Grameen Bank,” 2023). This resulted in Grameen Bank’s founder, Mohammad Yunus, being charged with violating Bangladesh’s labor laws. Due to possible questionable practices this results in Bangladesh losing one point on the rubric.

South Africa explanation

South Africa received a score of 2/3 for density. South Africa's overall density as a nation is 48.72 people per square kilometer according to last year’s census. However, major urban centers such as Cape Town (1,530 people per square kilometer), Johannesburg (2,696 people per square kilometer), Petoria (1,100 people per square kilometer), and Durban (2,600 people per square kilometer) are quite dense (“Census 2022 Population,” 2023). That being said, a study done by Brigham Young University found that the majority of loans take place in rural farm areas (Baumann, 2005). Most of these rural farm areas have a population that ranges from 500 to 1000 people. For that reason, South Africa receives the “middle of the road” score of 2/3 for density.

For governmental support, South Africa receives 2/3 points. South Africa has a committee to regulate microfinance known as the Microfinance Regulation Council. The Regulatory Council on their website explains they have rules that regulate microfinance institution interest rates to 5% on monthly repayments (“Small Business Development”, 2015). In addition, the council also asked that all microfinance institutions ensure that all recipients of loans have an “understanding” of the type of loan agreement they are getting into but was vague on language (“Small Business Development”, 2015). This vagueness of the wording leads to ambiguity that results in two instead of three points. The International Monetary Fund explained that banks within South Africa tend not to follow the Microfinance Regulation Council's rules and continue to use deceptive practices (Gulde et al., 2006).

Finally, South Africa received a score of 1/3 for internal management of banks. Although key microfinancing agency websites suggest a rigorous internal management structure, scathing reports and government audits suggest that most microfinance institutions do not audit loan recipients properly. In most instances, as a UC Berkeley study found in South Africa, loans were given and used for personal expenses (“Rethinking Microfinance,” 2018). The IMF report cited above and a study done by Northwestern stated the majority of microloans are being misused for personal reasons instead of entrepreneurial ventures (Whittaker, 2008). Utilizing loans for personal expenses rather than an entrepreneurial venture, and the failure of microfinance institutions to have an internal system that would prevent this from happening caused South Africa to earn only one out of three points.

Filling in the gap

When determining the difference between South Africa and Bangladesh’s microfinance programs it becomes abundantly clear that the density and internal management

of Bangladesh have allowed there to be a much more competitive and well-functioning microfinancing system than South Africa. Population density is an uncontrollable factor which governments do not have direct control over. However, in the analysis done by South Africa and Bangladesh, I have also found that the government and the MFIs themselves handle the micro-loan market much more efficiently, with better auditing and incentivization of high-productivity uses for loans. When loans are used for entrepreneurial endeavors and investments in capital, they help an economy grow in the long run; when they are used for personal expenses, they are spent on immediate consumption and have little to no effect on the long-run growth of microfinancing systems. It is also possible that better management of the micro-loan system could lead to its deployment in higher-density areas of South Africa.

Bangladesh's framework, which includes high density and strong institutional integrity from both governmental oversight and good management of MFIs, has resulted in a much larger poverty reduction than what South Africa could achieve. However, while South Africa has the same level of governmental oversight in theory as Bangladesh, there has been little institutional integrity. South African microloans have regularly gone toward non-entrepreneurial efforts and have therefore failed to follow the targeted principles of microfinancing. Comparatively, although in Bangladesh, organizations such as Grameen Bank's integrity have been severely questioned, there have been third-party audits that found that major microlending organizations in Bangladesh are performing necessary oversight and primarily giving loans toward entrepreneurial endeavors. Microlending banks within Bangladesh have shown through interviews that oversight exists within the company which has allowed it to perform much better than South Africa. South Africa has shown no evidence of proper auditing existing within the company. The difference between Bangladesh and South Africa's internal management despite similar governmental structures could allow for future longitudinal studies that assess the long-term impact that specific policies have on micro-financing itself. The purpose of this study was to primarily address the large differences that exist between South Africa and Bangladesh which paves the way to researching micro differences that exist between the countries which leads to dramatic differences in results. One major takeaway from this study that may help is to ensure a better internal structure exists within microlending systems in South Africa. Seeing as South Africa's internal microfinance structure performed so poorly, implementing more stringent transparency requirements for loan terms may be helpful if not necessary.

Limitations and future areas of growth

Limitations of the study include the fact there was a lack of expert peer review to legitimize the methodology of this study. The methodology of utilizing a rubric is often limiting and can be a broad way to address a complex issue. One of the most challenging issues that existed in this research was finding a quantifiable way to address the governmental support and internal structure for microfinancing organizations. Although I was able to find studies that discussed the failure to utilize loans properly, it remains difficult to quantify

“internal management” and “governmental support”. In the future, this study could be improved by focusing on specific language rather than broadly generalizing and rating policies. Despite the fact the rubric was able to successfully give a broad overview of the difference between South Africa and Bangladesh, it is limited in its ability to address more nuanced differences between countries. In addition, this paper was not peer-reviewed by any expert in microfinance who could catch discrepancies. However, the rubric still contributes to serving as a broad generalized comparison between two country’s microfinance systems.

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Impact of Neutral and Negative Word Choices in News Headlines on Thai People's Perception of Environment News Stories By Dalida Milly Janevathanavitya

Abstract

In today's fast-paced media landscape, headlines serve as a contact point between audiences and news stories. The language used in these headlines is not merely to summarize, but to influence how the readers interpret the article as a whole. To determine the level of impact headlines have on the perception of news stories, two surveys were designed and sent to two randomized groups of 56 people from Bangkok, Thailand. One survey showed a headline with neutral language and the other showed a headline with negative language, and both asked the respondents to predict information presented in the article. To reduce bias from politics and social issues, the news topic selected was environment news.

Results of this research demonstrate that negative word choices lead to more polarized perceptions compared to neutral ones. Usually, negative language triggers more people to choose relatively negative answers. However, on some occasions, it results in polarization, with most people choosing very negative or very positive answers. Moreover, predictions made by respondents who read headlines with neutral language were closer to real-world values than those made by respondents who read headlines with negative language. This suggests that negative word choices can distort perceptions and highlights the importance of careful consideration before drawing conclusions from headlines.

Keywords: word choice, news headlines, perception

Introduction

Diction manipulation is everywhere, from billboards on the roads to advertisements on milk cartons. Notably, even factual content such as news stories dedicated to public information distribution are designed to evoke emotions. With abundant information available due to the rise of digital media, a reporter's task is not only to provide informative updates but also to make them eye-catching, at times at the cause of shifting the story's tone.

Understanding the impact of diction manipulation is essential in promoting journalistic integrity and fostering an informed public. The media has a powerful role in shaping public opinion (Happer and Philo, 2013), and it does so through language. Previous experiments have shown that even subtle changes in language, such as a single word in a metaphor, can significantly alter the interpretation of stories (Thibodeau and Boroditsky, 2011). This research builds on that foundation by specifically examining how neutral versus negative word choices in headlines sway readers' perceptions of news stories.

To effectively analyze how language affects perception, this study minimizes pre-existing biases by choosing a less polarizing news topic: environment. By using weather

patterns as the focal point, this research hopes that the impact on perception is primarily due to the differences in language used in the headlines.

Literature Review

Several theories are crucial to understanding the impact of word choice in news reporting, including schema theory. Schema theory proposes that readers interpret information based on pre-existing mental frameworks or schemas — shaped through personal experiences, social interactions, and cultural influences (Bartlett, 1932). These schemas can be triggered by certain words or phrases. For example, in a narrative where characters are described using words associated with heroism or villainy, readers may activate corresponding schemas that affect their judgments of characters' actions later on. This is often applied to news reporting, where players are connected to characteristics through word choice.

In addition, other biases can arise upon encountering each word. Semantic network theory suggests that minds have interconnected structures to help people make connections quickly and use prior knowledge to facilitate comprehension (M.R. Quillian, 1967)—which encourages the linkage of words and ideas. Quillian states that each concept: *a car, to drive, keys*, are nodes made up of around 10-100 neurons in a network in our minds. A node, when activated, can activate related nodes, making inferences. Essentially, each word people read will activate connected ideas, which can drive perception one way or another.

Specifically, negative words can impact perception more than positive words. Negativity bias explains that negative experiences evoke stronger reactions and are processed more thoroughly than positive ones (Rozin and Royzman, 2001). In other words, a single negative event can outweigh several positive events in shaping a person's overall perception and memory. This bias has evolutionary roots, as paying more attention to negative stimuli (eg. tigers, wildfires) historically increased chances of survival. As the media realizes the power of negativity, it puts out overly negative news coverage that sometimes does not match the reality of the world (Soroka et al., 2015).

Aware that language influences perception, many searched for methods to shape understanding by taking advantage of word choice. Today, professionals in mass communications often utilize this deliberate wording design to package and present information to the public to achieve a desired viewpoint. In particular, for environment news, the media tends to oversimplify, misrepresent, or dramatize research results (Lewandowsky et al., 2012) through diction. This may be because scientific details can be difficult to digest or uninteresting for news readers. For example, when writing about a study forecasting future global extinctions as a result of climate change, news reports made the consequences seem more severe and the timescale shorter than actually projected by the study (Ladle Jepson and Whittaker, 2005).

Within news reporting, headlines are effective devices to alter the impression, understanding, and memory of the news content. Research has shown that misleading

headlines, including those of factual articles, can influence the recall and interpretation of the article's content (Ecker et al., 2014). Incorrect perception due to headlines can cause people to have a permanently biased understanding of the news. Studies have shown that corrections rarely eliminate reliance on misinformation, even when people acknowledge and remember the correction. Multiple explanations exist for the continued influence effect. One is the formation of mental models, where people prefer a coherent but incorrect model over an incomplete one (Johnson and Seifert, 1994). Another is retrieval failure, involving source confusion or failure of strategic monitoring processes, which can cause misinformation to be remembered as credible (Ayers and Reder, 1998). Lastly, people resist being told what to think which makes retractions ineffective (Lewandowsky et al., 2012).

Research Methodology

Designing the Surveys

To determine the differences in people's perception of news stories when headlines use neutral versus negative language, two surveys were designed. The headline in Survey 1 used neutral language (eg. reports, unusual) whilst the one in Survey 2 used negative language (eg. warns, catastrophe). The news headlines within each survey were created for the experiment to avoid prior knowledge or bias from real news.

The headline in Survey 1: Scientists Report Climate Impacts as Unusual Weather Patterns Arise

The headline in Survey 2: Scientists Warn of Imminent Climate Catastrophe as Extreme Weather Escalates

Both surveys asked the same three questions:

1. From this headline, estimate the earth's annual temperature increase mentioned in the article.
2. From this headline, estimate the annual percentage increase of natural disasters mentioned in the article.
3. From this headline, estimate the number of annual deaths due to natural disasters mentioned in the article.

All answers were multiple choice with an option for "other, please specify." All responses were collected through Google Forms.

Sampling

In total, 112 people in Bangkok, Thailand were surveyed. 56 responded to Survey 1 and 56 responded to Survey 2. The respondents included individuals from a wide range of educational qualifications, incomes, and ages, to satisfy the purpose of this research. As for

age, in Survey 1, 9.1% of respondents are in middle or high school (approximately 13-18 years old), 1.8% are in university (approximately 19-22 years old), 67.3% are working age (approximately 23-60 years old), and 21.8% are retired (above 60 years old). In Survey 2, the numbers were 16.1%, 5.4%, 71.4%, and 7.1%, correspondingly. As for gender, in Survey 1, 67.9% of respondents were male, and 32.1% were female. In Survey 2, 55.4% of respondents were male, 42.9% were female, and 1.8% answered other/prefer not to say.

Survey Findings

Q1	0.05°C	0.10°C	0.15°C	0.20°C	Total
Survey 1: neutral language	10	15	8	23	56
Survey 2: negative language	3	12	8	33	56
Total	13	27	16	56	112

Figure 1: Table displaying responses to Q1 from Survey 1 and Survey 2 respondents

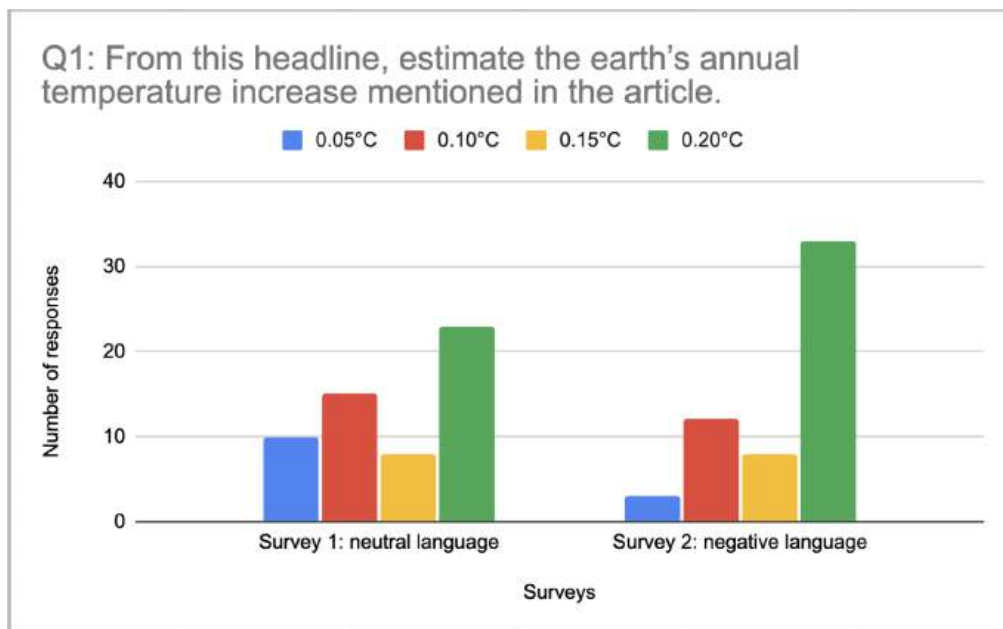


Figure 2: Graph comparing responses to Q1 from Survey 1 and Survey 2 respondents

In Q1, the results of Survey 1 and Survey 2 had similar trends but displayed a notable difference between the respondents' perceptions. The most popular prediction is the same in both surveys, which is 0.20°C or the highest temperature choice. However, the answers were more extremely skewed towards higher temperatures in Survey 2, as 58.9% of Survey 2 respondents chose 0.20°C, but only 41.1% of Survey 1 respondents did. It is also apparent that the lowest value is extremely unpopular in Survey 2, with only 5.4% choosing it. The average

of Survey 1 responses was 0.14°C and the average of Survey 2 responses was 0.16°C, which has a percentage difference of 13.3%.

Q2	5-10%	11-15%	16-20%	21-25%	Total
Survey 1: neutral language	18	21	11	6	56
Survey 2: negative language	13	12	16	15	56

Figure 3: Table displaying responses to Q2 from Survey 1 and Survey 2 respondents

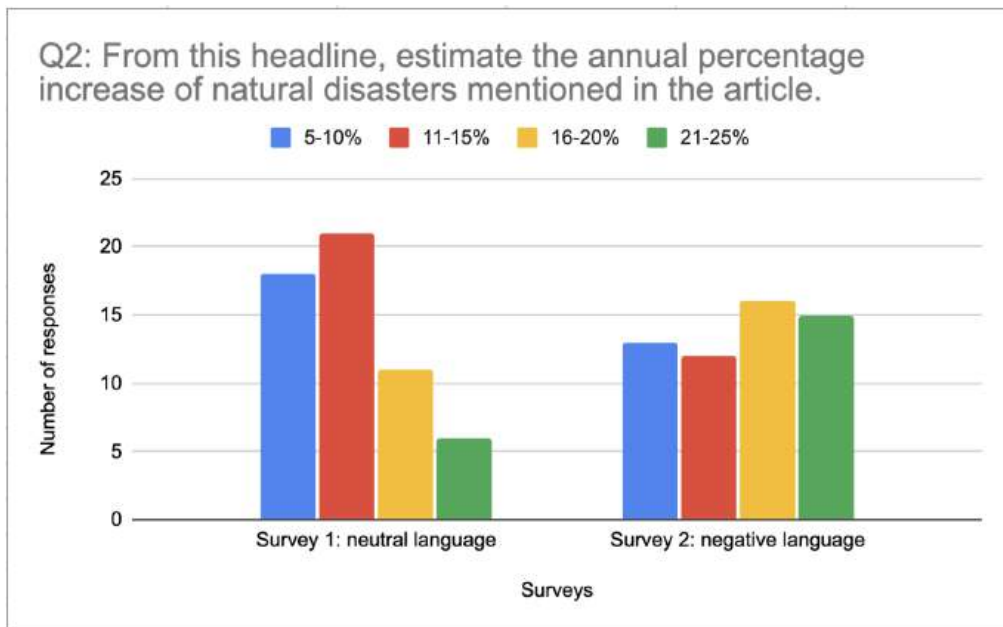


Figure 4: Graph comparing responses to Q2 from Survey 1 and Survey 2 respondents

In Q2, Survey 1’s answers were skewed towards lower percentage ranges, whilst Survey 2’s answers were more evenly distributed. Survey 1’s most popular choice was the lower-leaning range of 11-15%, whilst Survey 2’s most popular choice was the higher-leaning range of 16-20%. The average of Survey 1 responses was 13.3% and the average of Survey 2 responses was 15.8% (each average is derived from averaging middle values of each percentage range), which has a percentage difference of 17.5%.

Q3	10,000-30,000	30,001-50,000	50,001-70,000	70,001-90,000	Total
Survey 1: neutral language	19	23	7	7	56
Survey 2: negative language	21	13	7	15	56

Figure 5: Table displaying responses to Q3 from Survey 1 and Survey 2 respondents

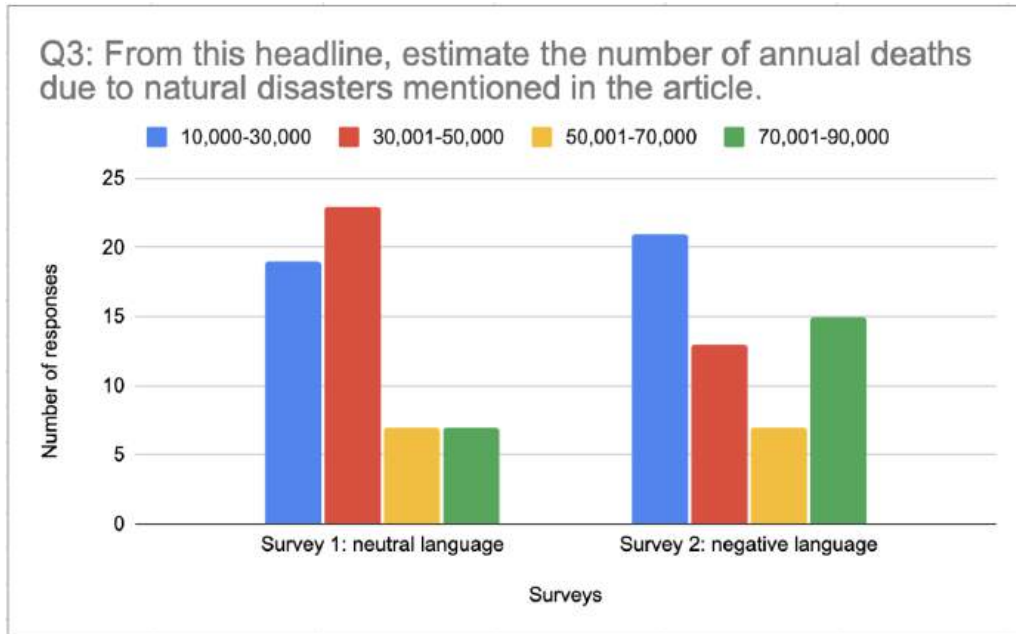


Figure 6: Graph comparing responses to Q3 from Survey 1 and Survey 2 respondents

Interestingly, in Q3, slightly more Survey 2 respondents predicted the lowest range of the number of annual deaths due to natural disasters compared to Survey 1 respondents. Whilst 33.9% of Survey 1 respondents chose the lowest range, 10,000-30,000 deaths, 37.5% of Survey 2 respondents did. In other words, unlike many may assume, the respondents who read negative language were more likely to predict the lowest range of deaths than the respondents who read neutral language in this particular question of this survey.

Resultantly, Survey 2 responses were extreme on both sides whilst Survey 1 responses were skewed towards lower ranges. Notably, 42 respondents of Survey 1 predicted lower-leaning ranges whilst only 34 respondents of Survey 2 did. This could suggest that most respondents of both surveys predicted lower-leaning annual deaths, but negative language also triggered some respondents of Survey 2 to choose the highest range, making the data polarized. Overall, the average of Survey 1 responses was 40,714 and the average of Survey 2 responses was 45,714 (each average is derived from averaging middle values of each death range), which has a percentage difference of 11.6%.

Analysis of Findings

All in all, negative language in headlines evokes substantially more negative perceptions of news articles in comparison to neutral language. In all cases, negative words in headlines triggered more people to make high predictions of the severity of an event compared to neutral words.

As the results of Q1 and Q3 from both surveys display similar trends, it can be presumed that a person's answer is predominantly dependent on their prior knowledge or

opinion of the topic. Perhaps most people heard climate change was bringing extreme weather and assumed the value of the increase would be very high, without considering that small values may be significant in the atmospheric scale. Nonetheless, an independent samples t-test revealed that the responses from Survey 1 significantly differed from those of Survey 2, $t(110) = 2.360$, $p = 0.020$, $d = 0.370$. Also, a chi-square analysis of Q2 responses found a statistically significant difference, $\chi^2(3, N = 112) = 8.044$, $p = 0.045$, $V = 0.268$. Hence, regardless of whether their fundamental beliefs were changed, their immediate decision-making was affected by language.

Even though Q3 did not show statistical significance according to the chi-square analysis, $\chi^2(3, N = 112) = 5.787$, $p = 0.122$, $V = 0.227$, it did show a nuanced insight. While negative language typically leads people to make more negative predictions, in this case, it also unexpectedly leads some people to make more positive predictions. This suggests that negative language can sometimes prompt a counter-reaction, causing people to consider less dire outcomes. This could be due to the backfire effect, which is when people become more entrenched in their beliefs after they have seen new information that firmly contradicts them (Swire-Thompson et al., 2020). Two types of backfire effects could potentially explain the phenomenon: the familiarity backfire effect and the worldview backfire effect. The worldview backfire effect happens when someone feels compelled to defend their worldview after receiving information that contradicts their beliefs. On the other hand, the familiarity backfire effect happens when false information is reiterated within the retraction, strengthening the belief. However, it is to be noted that the backfire effect has been difficult for researchers to find and replicate. Perhaps the results of this experiment could hint at the existence of the backfire effect in certain conditions. In summary, neutral language led to more moderate estimates, whereas negative language caused polarization, with respondents divided between very low and very high predictions. This highlights the complexity of human behavior and the importance of understanding how different framing effects can influence perceptions in varied ways.

Another interesting angle to observe is the survey results compared to real-world data. In general, respondents who read neutral language predicted values closer to real-world data. For Q1, the real-world answer is approximately 0.08°C (calculated from the average global temperature in 2020, $1.20 \pm 0.1^\circ\text{C}$ above the pre-industrial level (WMO, 2021), and the average global temperature in 2023, $1.45 \pm 0.12^\circ\text{C}$ above the pre-industrial level (WMO, 2024)). The average of Survey 1 responses was closer to 0.08°C at 0.14°C compared to the average of Survey 2 responses at 0.16°C . Furthermore, 0.08°C is closest to 0.10°C , which was chosen by three more respondents who read neutral language. Slightly better results are revealed in Q2. For Q2, the answer was 9.1% (calculated from the increase to “28 weather and climate disasters in 2023” from “the previous record of 22 in 2020” (Smith, 2024)). The average of Survey 1 responses was 13.3%, which is closer to 9.1% than the average of Survey 2 responses at 15.8%. Moreover, 9.09% was within the range of 5-10%, which was chosen by

five more respondents who read neutral language. Additionally, the most popular choice in Survey 1 is 11-15%, closer to 5-10% than the most popular choice in Survey 2, 16-20%. The clearest results lay in Q3. For Q3, the answer was 40,000-50,000 per year (Ritchie and Rosado, 2024). The average of Survey 2 responses was 45,714, right around the middle between the real-world range whilst the average of Survey 1 responses was 40,714. Here, the most popular answer for respondents who read neutral language, 30,001-50,000, matched with real-world data, but the most popular answer for respondents who read negative language did not. It could be interpreted that the similarities between survey values and real-world values correlated with the question's relevance to the headline. Question 1 was about temperature, which is most directly linked to the headline. Question 3, however, was about deaths, which requires double-layer thinking when related to the headline. From this research, it is clear that neutral language encourages more accurate perceptions of news stories.

Limitations

This study has several limitations that must be acknowledged. One limitation concerns the interpretation of responses gathered through the survey. The use of multiple-choice questions, even with the option for “other, please specify,” may not fully capture the depth of participants' perceptions. Incorporating more detailed open-ended questions in future studies could capture the nuances of their interpretation of the headlines and offer richer insights into how language framing affects perceptions.

Another significant limitation is the potential influence of participants' prior knowledge and opinions about the environment. This could confound the effects of word choice, making it challenging to isolate the impact of neutral versus negative language on their perceptions. To mitigate this limitation, future research could assess participants' baseline knowledge and attitudes, allowing for a more controlled analysis. Additionally, expanding the scope to include a broader range of topics could help isolate the effects of language framing more effectively.

Conclusions and Recommendations

This study reveals that word choice significantly influences public perception of climate-related news and likely other types of news. Generally, negative headlines led to higher predictions of event severity, whereas neutral headlines resulted in more moderate estimates. Occasionally, though, negative language results in polarization, with most people choosing extremely negative or extremely positive answers, indicating a complex interaction between language and perception. In addition, comparisons with real-world data showed that predictions based on neutral language were closer to actual values, underscoring the effectiveness of neutral language in promoting realistic perceptions.

These findings suggest that media outlets should adopt guidelines for balanced headline writing to avoid sensationalism and encourage accurate public understanding.

Journalism training programs could include modules on the impact of language framing to sensitize future journalists to these effects. This approach can help mitigate the polarization and fear induced by negatively framed headlines. Furthermore, educating the public on language framing and promoting media literacy can empower individuals to critically evaluate news stories.

Future research should explore diverse contexts to generalize these findings. For example, studies could use a wider range of headlines to test the effect of word choice on the perception of various topics of news stories. Cross-cultural studies could provide insights into how people from different countries interpret and respond to language framing in the news. Additionally, examining the impact of positive headlines compared to neutral and negative ones could offer a more comprehensive understanding of language effects. Sharing these findings can foster a more informed and less polarized public, ultimately enhancing the quality of news consumption and discourse.

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Alzheimer's Disease By Madhav Donthula By Madhav Donthula

Abstract

Alzheimer's has seen a 145% increase in death rates over the past two decades. Early diagnosis is critical, but symptoms may be subtle, avoiding the eyes of many neurologists. As a result, the Food and Drug Administration (FDA) has been experimenting with the use of machine learning in the field of neurology. Throughout this project, machine learning models were developed for assessing disease severity by training on over 3000 brain scans categorized as moderate, mild, very mild, or non-demented provided by Kaggle. Due to class imbalance, severities had to be consolidated to avoid bias. The multi-layer perceptron was able to achieve an accuracy score of 52%. In comparison, the random forest model achieved 91% in identifying the stage of Alzheimer's progression based on brain imaging patterns invisible to the human eye. Looking into the confusion matrix reveals that the random forest's inaccuracy tends to misdiagnose. False positives are preferred to false negatives because valid Alzheimer's detection outweighs the potential harm of false positives. Artificial intelligence (AI) systems could be a powerful second opinion to improve diagnosis accuracy and provide evidence-based guidance on the most effective treatment options before making life-altering prescriptions.

Introduction

Machine learning gained its attention in 1959 by Arthur Samuel through the use of pattern recognition. As a result, it has been used to replace several time-consuming activities in many fields including medical care¹. Later on, in 2015, Japanese pharmaceutical company Eisai used wearable data from 122 individuals with varying dementia to run an ensemble machine learning model². Ensemble machine learning combines multiple models to predict outcomes more accurately than one³. This development has elevated the interest in machine learning's viability in diagnosing neurodegenerative diseases like Alzheimer's

Alzheimer's disease affects millions worldwide, incrementally robbing them of crucial memories and cognitive function. Compared to HIV and stroke, which had their death rates decrease in the past two decades, Alzheimer's saw an increase⁴. There is no clear cure for Alzheimer's. Still, medications have been proven to slow the effects to a tolerable state.

Early diagnosis is essential to provide the best care, yet subtle symptoms often evade detection even by the most qualified neurologists⁵. After studying over 1,000 patients, the National Alzheimer's Coordinating Center database found that 1 in 5 Alzheimer's cases tend to be misdiagnosed⁶. An increased probability of misdiagnosis can give patients false hope and delay or prevent further care altogether. Alzheimer's shares similar effects as other medical conditions including vascular dementia and Parkinson's disease making it challenging to differentiate without a second opinion⁷. With the rise in Alzheimer's cases, neurologists have been investigating various diagnostic tools that could diagnose at onset or before onset. Consequently, the FDA has granted limited approval for companies to explore the potential of

machine learning in healthcare and medical diagnosis in the past few decades. According to a statement in 2024, the FDA approved approximately 97% of AI/ML-enabled devices⁸.

Machine learning relies on several biomarkers to identify the prevalence of Alzheimer's. These biomarkers include amyloid plaques and tau tangles⁹. As these plaques add up, they disrupt communication between the neurons and lead to neuronal damage. MRI, CSF, and PET scans are one of the many tools used to diagnose Alzheimer's, but using an MRI is one of the best options available¹⁰. MRIs withhold the ability to detect shrinkage which is a result of Alzheimer's. However, machine learning models offer new hope by spotting brain patterns that are hard to see for the human eye, helping neuroscientists make life-altering prescriptions.

This research aims to create a machine-learning model that can effectively read and identify the presence of Alzheimer's when given an MRI scan. The objective is to teach the machine to recognize the biomarkers that indicate Alzheimer's and make educated predictions on future scans. Data was gathered from the internet but came with a class imbalance that had to be resolved. Next, two machine learning models were trained and compared based on accuracy.

Methodology:

The research was based on an experimental research design from the Kaggle dataset provided by Sachin Kumar. The data consisted of 6400 individual MRI scans that indicate dementia varying from non-demented, mild, very mild, and moderate. To ensure reliability, the scans were compared to ensure equal representation was given to each level of dementia. It was determined there was an imbalance that could have led to undercoverage. The “Non Demented” images far outnumbered examples labeled “Moderate Demented,” “Mild Demented,” and “Very Mild Demented.”

Class - 1: Mild Demented (896 images)

Class - 2: Moderate Demented (64 images)

Class - 3: Non Demented (3200 images)

Class - 4: Very Mild Demented (2240 images)

To resolve this class imbalance, strategic consolidation of the three demented categories was completed during preprocessing. Combining these related classes balanced the data, giving the models equal exposure to demented and non-demented scans. Unifying the labels enabled the algorithm to learn a robust dementia detection system not skewed by lopsided classes. This data-wrangling step was key for fair model training. Lopsided classes would have potentially produced a biased model and reduced the accuracy score resulting in a flawed model.

Next, the split of the train and test sets was made by using `train_test_split` and setting the fraction of data used for testing as 0.2, this ensured each model had sufficient data to learn

and test its accuracy in detecting Alzheimer's. Random Forest and Multi-Layer Perceptron Classifiers were then created and compared based on the results.

MODELS:

Random Forest: The random forest classifier is a commonly used ensemble machine learning algorithm that combines multiple decision trees rather than one. We used 100 decision trees for this model. This makes it a valuable choice for the dataset because of the decision trees' image recognition capabilities. An accuracy score of 0.914 was returned.

Figure 1 shows the confusion matrix that is generated from the Random Forest model:

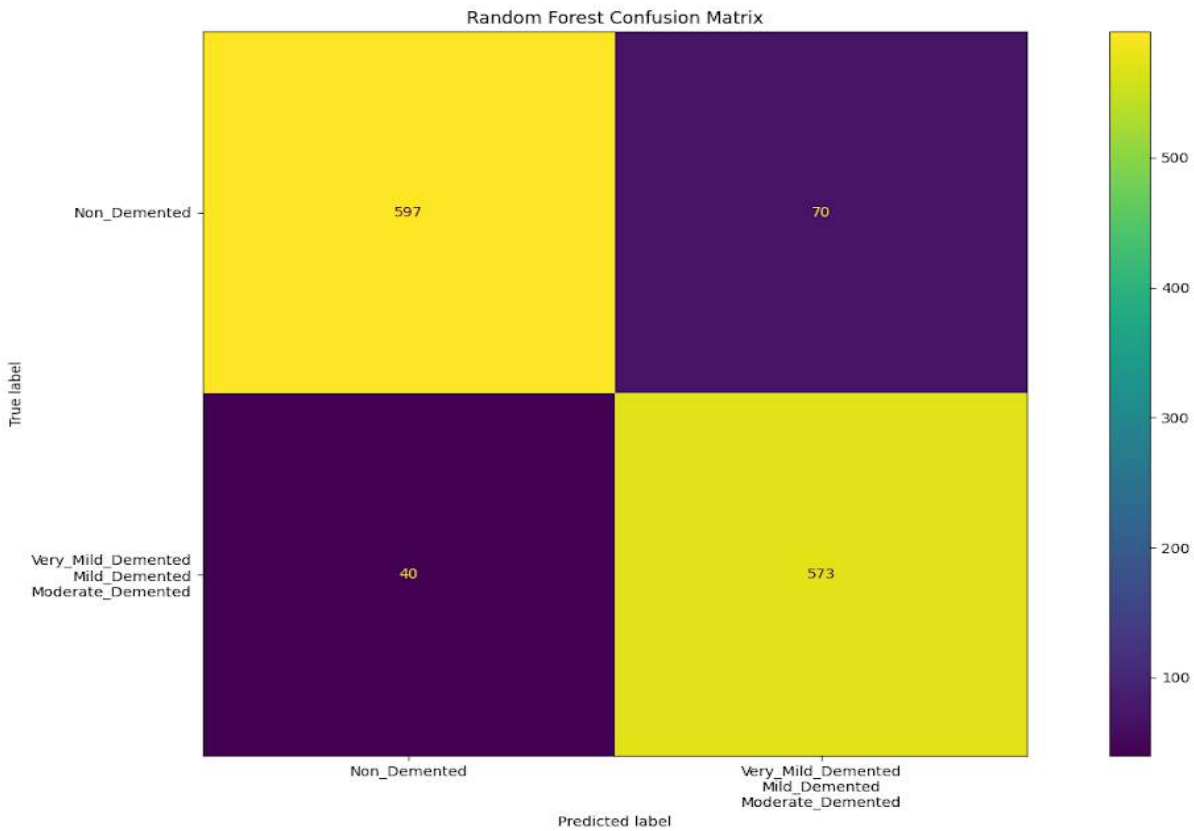


Figure 1: Confusion matrix of the model.

Multi-layer Perceptron Classifier (MLP): We also used the MLP Classifier, which is another commonly used machine learning model that can capture patterns in data. However, our dataset yielded an accuracy rate of only 51%. As observed in Figure 2, the training loss (blue line) and validation loss (orange line) are constant throughout the epochs. This means that the model is not learning over time.

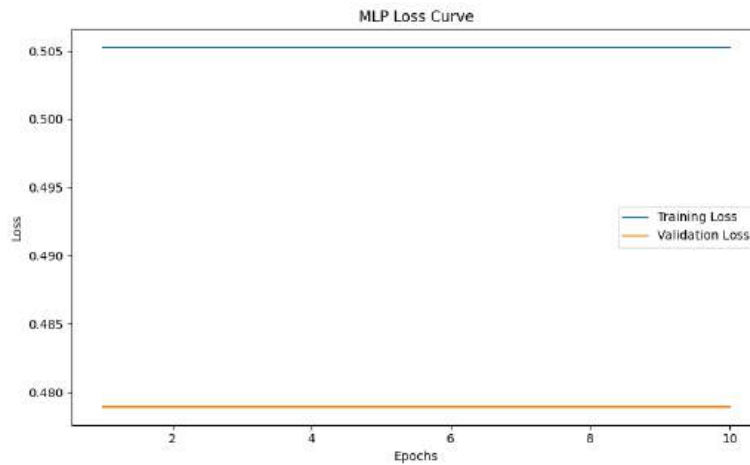


Figure 2 shows the loss over epochs for the MLP classifier:

MODEL NAME	ACCURACY
Random Forest	0.914
Multi-layer Perceptron Classifier (MLP)	0.521

A determination was made that Random Forest performed at a higher percentage in detecting Alzheimer's. Due to the large performance gap, additional exploration was conducted to compare findings in previous research.

Discussion

This project demonstrates the potential of machine learning to aid in the precise diagnosis of Alzheimer's disease. Two classification models were trained on the same dataset and accuracy ratios were outputted. The random forest model achieved a promising accuracy of 91% in categorizing brain scans. On the other hand, the multi-layer perceptron only achieved an accuracy score of 51%. A contributing factor was the lack of the Multi-layer Perceptron Classifier's ability to pick up high dimensionality and the complexity of image features in our dataset. According to Medium.com, "spatial information is lost when the image is flattened (matrix to vector) into an MLP"¹¹. The research indicated MLP would be sufficient for simpler tasks but is not an ideal model to depend solely on for accurate diagnosis information. Another factor that may have contributed to the poor accuracy score is class consolidation. Combining multiple severities of dementia, even if it is necessary to compensate for the class imbalance, poses a challenge for the machine to identify the key biomarkers.

Contrastingly, the Random Forest model's 91.4 % accuracy surpasses unaided human diagnosis, which is prone to oversight and misdiagnosis. Using a combination of the machine

learning Random Forest model and human interpretation, the potential to achieve a nearly perfect diagnosis significantly improves. Further, this model can bolster doctors' confidence in their diagnoses and validate their identification of early signs of cognitive decline.

Analyzing the confusion matrix illustrates why the Random Forest's accuracy score is capped at 91%. Explanations for this phenomenon are indicated by the multitude of aspects including the inability to account for every variable found on each scan. Some scans may indicate other disorders not related to Alzheimer's such as Parkinson's and these biomarkers may muddle the machine's ability to read the scan accurately. This is shown in the "false positives" and "false negatives" readings. Fortunately, the model's inaccuracy comes from more false positives than false negatives. A false positive reading is denoted when the model incorrectly concludes the presence of Alzheimer's. A false negative is recorded when an individual does have Alzheimer's but the machine does not detect the disease. Research indicates that misdiagnosis can lead to a patient being given inappropriate medications or therapies. Both of these can harm the patient and the healthcare field. Still, false positives are generally considered better for most professionals because of the health implications that can occur with a false negative¹². Hence, this prevents machine learning's viability as a single diagnostic tool.

Although 91% is satisfactory, machine learning is limited in its capabilities and are not advanced enough to be used without human contact. In addition, past applications of Machine Learning generated several problems in the healthcare field. The biggest one was the underrepresentation of minorities¹³. This is when the machine learning model is trained on an undiversified dataset causing it to produce biased answers. This limits the model's ability to assess individuals of differing racial identities and may lead to more errors over time. Another issue is AI's hallucination rates which lead to false outputs that may harm patients. Hallucinations are when AI models tend to generate inaccurate conclusions and validate them. This can be severely harmful to those who solely rely on AI for their medical determination causing them to worsen their condition or allow it to go untreated. Consequently, most healthcare AI chatbots have to be closely monitored by their agent companies to avoid harm to the patients.

This research indicates that further testing and training in machine learning can yield higher results. Specifically testing and training on larger and diverse populations can widen the accuracy of the model when applied to real-world situations. Additionally, accompanying the various brain scans, such as PET and CT scans, can strengthen AI's inference and provide doctors with more precise results. In conclusion, it is the determination of this research that AI can reach its designated state of autonomy with further exploration into the components that impair accurate readings.

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Martian Pavements: Adapting Earth-Based Engineering for Extraterrestrial Habitation

By Anika Deshpande

Abstract

In today's age of technological advancement, space travel has become a more prevalent topic than ever before. Extraterrestrial human habitation, specifically on Mars, is no longer a distant fantasy, but a tangible and imminent possibility for upcoming generations. Effective transportation is essential for the distribution of critical resources, the facilitation of economic activities such as the production, transport, and exchange of goods, and the enhancement of social connectivity within the settlement. Roadway pavements could become a critical component of the surface transportation system of a Martian settlement. This paper explores the key considerations for building Martian pavements, detailing an effective comparison with pavements on Earth. The required elements for a successful pavement network were identified by studying the history and development of pavements on Earth. The most recent and current pavement engineering practices and processes associated with traffic, materials, climate, construction, and maintenance are summarized. Although martian pavements are essentially an extension of those on Earth, Martians-to-be will still need to envision the type of vehicles that will be used, collect data on materials and climate, implement autonomous machinery that will suit any harsh conditions of Mars, and create a plan for the maintenance in order to effectively implement an essential pavement infrastructure.

Introduction – Humans on Mars!

As you cast your gaze into the night sky above, the crimson-colored planet is seen. Once a distant figment of the imagination, the prospect of human habitation on Mars is now within the realm of possibility. Mars is the fourth and next planet from the sun after Earth. It shares similar days and seasons to those of Earth due to the similarity in the rotational period of Mars and the tilt of the rotational axis relative to the ecliptic plane in comparison to Earth (Liu et al, 2022). Its similarity to Earth has spurred great interest in the further exploration of Mars and the possibility of human settlement. This is evident in that over 30 probes have explored Mars and acquired data and images from exploring its surface (Liu et al, 2022).

Some recent efforts have been made by both National Aeronautics and Space Administration (NASA) and Indian Space Research Organization (ISRO). NASA's Curiosity Rover has traversed the Martian terrain since its arrival in 2012. Through its sophisticated instrumentation and exploration, Curiosity has uncovered valuable insights into Mars' geology, climate, and potential for habitability, such as the presence of key elements to support microbial life: nitrogen, oxygen, sulfur, phosphorous, and carbon (*Mars Science Laboratory: Curiosity Rover*, 2024). ISRO's Mission Mangal (*Mangalyaan*) primarily focused on studying the Martian atmosphere and surface, but also contributed to data on Mars' atmospheric composition, dust storms, and seasonal variations. With its five sophisticated

payloads including the Lyman Alpha Photometer (LAP), the Mars Exospheric Neutral Composition Analyzer (MENCA), the Methane Sensor for Mars (MSM), the Mars Color Camera (MCC), and the Thermal Infrared Imaging Spectrometer (TIS), *Mangalyaan* comprehensively studied the surface composition of Mars and contributed to a well-rounded comprehension of the planet and its intricacies (George, 2024). The United Arab Emirates (UAE) and China have also operated craft on the Martian surface with Hope and Tianwen-1 respectively.

Looking to the future, NASA has set the 2030s as a reasonable timeframe for the first human exploration mission on Mars. Additionally, private space exploration companies like SpaceX are looking into possibilities. SpaceX CEO Elon Musk has stated that humanity must become “a multiplanetary species” if it is going to survive and is working on a plan that could send a million people to live on Mars in the 21st century (Drake, n.d.-b). As aspirations like these for settlements on Mars intensify, the establishment of durable and functional infrastructure becomes imperative. Infrastructure such as payload transport systems, entry, descent, and landing (EDL) mechanisms, and surface technologies have been considered critical to the success of both crewed and uncrewed missions. A study by Kolemman Lutz, Bruce Mackenzie, and colleagues identified that payloads and systems can be efficiently deployed from reusable launch vehicles (RLVs) using payload transport elevators, vertical elevator shaft towers, cable car systems, and separate EDL procedures. Innovative surface deployment mechanisms such as cable-zip line systems and tethered spacebot deployment rovers were regarded to be crucial in establishing transportation networks and supporting a variety of mission objectives. These systems were concluded to both enable the deployment of essential infrastructure and offer versatility in building, maintaining, and deploying vital components such as water production plants, power reactors, and life support systems (Lutz et al, 2021).

Among this multitude of infrastructure, pavements may also be considered indispensable. Pavements can also provide stable surfaces for movement, equipment deployment, and infrastructure installation in addition to connectivity for Martians. In the harsh Martian environment, where the regolith may pose challenges for stability and traction, well-designed pavements can ensure safe and efficient transportation of payloads, equipment, and personnel. Moreover, pavements will also facilitate the establishment of foundational structures, such as habitats, research facilities, and resource extraction sites, by providing stable platforms for construction and assembly activities. As settlements expand and evolve, pavements can serve as essential connectors, enabling seamless movement between various facilities and enhancing operational efficiency. Therefore, the development of robust pavement technologies tailored to Martian conditions could be considered paramount for the success and sustainability of future human settlements on the Red Planet.

As human presence on Mars evolves, the need for robust infrastructure will become increasingly pronounced. Pavements may be needed to not only support transportation needs but also provide foundations for expanding habitats, scientific research facilities, and resource

extraction sites. Collaboration between space agencies, private enterprises, and international partners will be crucial in realizing these ambitious goals and laying the groundwork for sustainable Martian colonies. Furthermore, the development of Martian pavements may garner attention not only from space agencies but also from the broader scientific community and the public, as it represents a tangible step towards realizing humanity's interplanetary ambitions.

Motivation

When considering transportation methods on Mars, both ground and air transport may be viable options. However, air transport, such as a spaceplane, can require significantly more energy due to the challenges of overcoming gravitational pull and atmospheric drag. Simulations performed by De Curtò & De Zarzà indicate this by showing a spaceplane could consume an average of 4.37 GJ per round trip, including ascent and descent, whereas a magnetic levitation (maglev) train—representing ground transport—used only 3.47 MJ per 100 km on flat terrain (De Curtò & De Zarzà, 2024). Financially, the maglev train system presented high average costs (USD 6014.32 million, with a standard deviation of USD 1008.46 million) due to the extensive infrastructure required, compared to the spaceplane's lower and more predictable costs (USD 1497.91 million, with a standard deviation of USD 251.09 million) (De Curtò & De Zarzà, 2024).

Despite such studies providing useful information on potential transportation alternatives, there is a gap in current considerations of roadways as a means of transportation. Additionally, pavement roadways are advantageous for the following reasons: 1. Roadways can allow for more efficient transport of heavy cargo compared to air transport by consuming less energy. 2. Pavement roadways may be considered more accessible compared to railroads and can be constructed quickly and easily to connect businesses and homes. 3. The resources required to construct pavements may be less than those for railroads, especially in manufacturing.

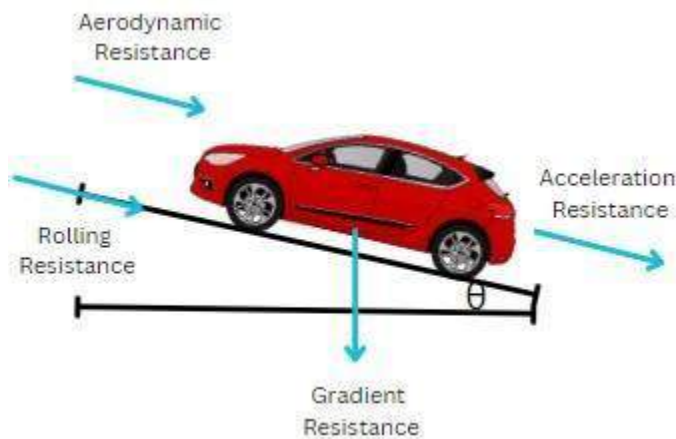


Figure 1: This figure depicts the various resistive forces acting on a vehicle as it ascends an incline. The gradient resistance (F_{grad}) is the force needed to counteract gravity, which

increases with the slope angle and the vehicle's weight. Rolling resistance (F_{roll}) is the force required to overcome energy losses between the vehicle/tires and the road surface. Aerodynamic resistance (F_{aero}) is the drag force that opposes the vehicle's motion through the air, depending on the vehicle's speed, shape, and frontal area. Lastly, acceleration resistance (F_{acc}) refers to the force required to accelerate the vehicle. In terms of energy consumption, the use of roadway transportation could potentially demand less energy as suggested by calculating the sum of the forces on a moving vehicle in Equations I-II based on the forces illustrated in **Figure 1**. Due to the force of gravity being roughly 40% of that on Earth (Williams, 2015) and the air density being 0.02 kg/m^3 (considered negligible as per Equation II.) (*Mars Fact Sheet*, 2024), these forces on a moving vehicle on Mars can be represented by Equation II.

I. $F_{trac(Earth)} = F_{aero} + F_{grad} + F_{acc} + F_{roll}$

Here, $F_{aero} = 0.5 \times \rho \times v^2 \times A \times Cd$ where ρ is air density in kg/m^3 , v is velocity of the vehicle in m/s , A is frontal area of the vehicle m^2 , and Cd is air drag coefficient of the vehicle.

$F_{grad} = m \times g \times \sin\phi$ where m is mass of the vehicle + payload in kg , g is gravitational constant (9.8 m/s^2), and ϕ is up gradient angle in radians

$F_{acc} = m \times a$ where m is mass of the vehicle + payload in kg , and a is vehicle acceleration in m/s^2 .

$F_{roll} = Crr \times m \times g$ where Crr is coefficient of rolling resistance, m is mass of the vehicle + payload in kg , and g is gravitational constant (9.8 m/s^2).

The same definitions apply to the following.

II. $F_{trac(Mars)} = 0 + 0.4 \times F_{grad} + F_{acc} + 0.4 \times F_{roll}$

III. $F_{trac(Earth)} > F_{trac(Mars)}$

Therefore, the energy required to drive on Mars is less (Equation III), making it a potentially viable option for transportation.

Given that air transport can pose demands such as advancements in propulsion technology due to Mars' low air density, maglev trains may require overcoming a significant upfront investment, pavement roadways may offer a more cost-effective and energy-efficient solution with other added benefits such as increased payload and better connectivity, which is why this paper focuses on them as the preferred transport method.

History of pavements on Earth

Here on Earth, pavements and roads have evolved from those of early civilizations over several millennia to the modern road networks we are so familiar with now. In *Paving our ways: A History of the World's Roads and Pavements* (Lay et al., 2020), the authors provide a wealth of information regarding the characteristics and principles of pavements from different eras throughout time. The following section contains multiple summarized excerpts of the above-mentioned book.

The earliest existing pavement dates from approximately 2500 BC Egypt, when the Old Kingdom built pyramids and temples. Because the majority of the construction took place near Cairo and the stone for construction had to be obtained from the Gebel Quatrani Quarries which were nearly 70 km southwest, the Lake Moeris Quarry Road had to be constructed in order to bring the basaltic stone to Giza. Considering the Egyptians did not have vehicles that were drawn by animals, the stones were transported on sleds or logs which rolled on the road surface. In terms of “urban” pavements, Mesopotamian civilizations are credited with building some of the most well-known processional paths between the Tigris and Euphrates Rivers dating from about 700 BC. These ceremonial pavements were meant to showcase opulence and great power. Due to the scarcity of suitable building stone in Mesopotamia, early constructed pavements predominantly utilized locally made bricks crafted from sun-dried mud, as well as fired bricks formed by compressing clay and then baking them in kilns fueled by oil. Additionally, imported slabs derived from layered limestone were incorporated. These elements were occasionally bonded together using naturally occurring bitumen, which was prevalent in the region and commonly employed for waterproofing and sealing tanks. Although Mesopotamian empires constructed extensive road networks to assert control over their vast territories, the roads were frequently constructed using rudimentary materials like rammed earth, which offered limited quality compared to more sophisticated modern day paving techniques. There was a low demand for manufactured pavements outside urban centers, especially as the influence of Mesopotamian empires waned over time. (Lay et al., 2020)

Beyond the prestige of Mesopotamian roads, Roman roads were exceptional in their time. With an estimated total length of well over 100,000 km, likely influenced by Carthaginian pavements and Minoan construction, the Romans similarly used cement. They further improved their design by adding volcanic tuff which forms a mortar when reacted with water. The building of arterial roads was fueled by the manpower of young men who were required to devote around 2 years to municipal projects. The construction of extensive road networks (for example, the Via Appia) allowed the Roman army to seize and control its growing territories. Additionally, they allowed for the consolidated Roman government to exert control and tax the extent of its vast empire, as well as facilitating the safe return of newly acquired treasures.

The Roman road-making process involved constructing longitudinal drains along the road edges, excavating the road alignment to solid ground, filling it with local stony material, compacting it with hand-held timber rams, and leveling the surface with sand. This constructed layer, called the agger, served as the subgrade for the pavement and could elevate the road by up to a meter above the surrounding land. This process was advantageous in that the deep longitudinal drains efficiently managed water, preventing damage and aiding drainage. Secondly, constructing these drains often provided material for the agger. This dual purpose ensured proper drainage while contributing to road construction. Additionally, the elevated agger raised the pavement level, reducing flood risks. Lastly, the raised pavement

facilitated defense against roadside attacks, enhancing traveler safety. Despite the pains taken to construct and operate these pavements, Roman roads disappeared because of insufficient maintenance as the empire diminished around 300 AD. These Roman records document that roadways required significant labor, which depended on the type of equipment used in construction.

The city of Phillipopolis in the 4th and 5th centuries Common Era (CE) saw the rise of decorative mosaic pavements. This architectural transformation can be attributed to the persecution of Jews in the beginning of the 5th century CE (Topalilov, 2020). The new Christian buildings that emerged during that time were decorated with these mosaic pavements. Often made from sandstone and terracotta, these mosaics had a variety of colors ranging from white, yellow green, brown, red, and black tesserae. In the subject of partial reparations, damaged or warped motifs were restored by marble and brick slabs (Topalilov, 2020)

In the 13th century, construction and repair phases of pavements were applied, particularly the Primary Mix and the Scott Repair. The Westminster Abbey Sanctuary pavement stands out as a unique example, potentially one of a kind north of the Alps. The Primary Mix, laid around 1268, utilized *cocciopesto* mortars with crushed ceramics as a pozzolanic additive. Mortars were integrated into the design, filling millimeter-wide gaps between stones. Microscopic examination revealed a well-carbonated paste with hydraulic properties, unreacted lime, and particles of partially or unburned limestone. (Hughes et al, 2012)

In Medieval England, pavements were essential to the success of the commercialized economy due to its role in attracting trade and commerce. Dating back to the early 14th century, these pavements were meticulously crafted with various materials to withstand the rigors of daily traffic. Near the market in New Winchelsea, Sussex, parts of Quarters 18 and 19 were paved with flat-laid Tilgate sandstone for a firm surface, larger slabs, edge-set smaller stones, and a stone kerb to redirect surface water, demonstrating thorough construction. Furthermore, sections within Pipewell Gate and the area descending from St. Leonard's Church into the marshes showcased another prevalent paving method: Tilgate stone pitched on edge, creating a tightly packed, resilient surface. These designs were made to endure heavy foot traffic and the passage of carts and animals. (Harvey, 2010)

The construction process was intricate and intensive. Historical records detail the procurement of materials, such as stone and gravel, from nearby quarries. Laborers were hired to extract and transport these materials to the construction site, where skilled paviors laid them down. Excavations at various sites revealed layers of sandstone cobbles, clay, gravel, and crushed tile. (Harvey, 2010)

Beyond mere convenience, these roads served as vital arteries for commerce, facilitating the flow of goods and people to and from the bustling market. The constant wear and tear of daily traffic necessitated regular maintenance and repair to the cobbled streets, as

evidenced by historical accounts and archaeological findings of patched stones and repair materials like flint and chalk. (Harvey, 2010)

The civil engineer Pierre-Marie Jerome Trésaguet of France proposed a pavement structure that relied on the subgrade for structural strength and stiffness, unlike the Roman method which he deemed unnecessarily massive. His approach involved emphasizing longitudinal drains and kerb stones to aid water removal from the pavement. Trésaguet's method utilized a layer of vertical quarried stones without the awkward bottom layer of horizontal stones used by Gautier. He advocated for two upper layers of broken stone, well-compacted and hammered into gaps between larger stones, providing a more cost effective and operationally efficient pavement compared to Gautier. His method gained traction in 18th-century continental Europe and Russia.

In contrast, Thomas Telford introduced a pavement construction method that prioritized stability and durability when he was tasked with improving Highland roads in Scotland. His approach involved laying a solid foundation of hard stones, followed by layers of broken stones strategically placed to withstand the rigors of traffic. Telford's meticulous attention to stone size and the elimination of clay and soft rock fillers set new standards for pavement longevity and performance.

Around 1802, property manager John McAdam began road maintenance and construction in the Bristol area of southwestern England. Observing various pavement performances, he concluded that a compacted layer of broken stones directly on a solid subgrade could be effective. Following Trésaguet's approach, McAdam sloped his subgrades across the roadway towards roadside drains for water drainage. McAdam favored interlocked stones over binding agents, believing broken stones would naturally form a smooth, weather-resistant surface when compacted. He insisted on a pavement surface impermeable to water to protect the subgrade. By utilizing smaller stone pieces from the breaking process, hand-rammed into surface voids, McAdam achieved the desired solid surface. In McAdam's method, the compacted layer of broken stones behaves as a single cohesive unit, distributing surface loads effectively before reaching the subgrade, obviating the need for costly layers of large stones used by previous methods. McAdam's revolutionary broken-stone principle remains integral to modern highway construction, with the term "macadam" coined since 1820 to describe courses of compacted broken stones working together through natural interlock (Hughes et al, 2012).

In the early 20th century, asphalt technology saw notable advancements. John McAdam's observation that applying bitumen onto broken stone surfaces could create effective rural pavements was critical in the advancements to come. Additionally, asphalt became recognized as a viable option for urban pavements when Trinidad bitumen was incorporated. As experience grew, asphalt's utility expanded beyond surface courses to include structural base courses, with various mix proportions explored. Notably, "gap-graded mixes," characterized by larger stones and increased bitumen content, gained popularity, known as stone-filled sheet asphalt in the USA and hot rolled asphalt (HRA) in Britain. While HRA

exhibited excellent tension and fatigue resistance due to its high bitumen content, it faced compression limitations, particularly in hot climates.

The mechanization of road-making processes throughout the 20th century was perhaps the most significant change. Mechanistic design methods refer to procedures capable of connecting the analytical calculation of pavement response, derived from the fundamental engineering properties of materials established in the laboratory, to the real-world performance of operational pavements (Fwa, 1999). Over the years, researchers have made substantial strides in developing theoretical models and test procedures that would improve the accuracy and efficiency of pavement analysis. For instance, the development of multi-layered elastic theories (theories that take into account the elastic properties of the asphalt, base, and subbase and the interactions between them to predict how the pavement will respond to loads, temperature changes, etc.) has facilitated the analysis of deflections, stresses, and strains in flexible pavements under traffic loads (Fwa, 1999). Advancements in paving machines, compaction techniques, and asphalt spreading, automatic control systems and GPS technology further enhanced precision and efficiency in asphalt placement. Additionally, the production and refinement of bitumen saw advancements, including enhancements in ductility through the addition of materials like natural rubber. Despite empirical approaches in pavement material selection, the 20th century marked a period of significant progress in asphalt technology, laying the groundwork for modern pavement construction practices.

Contrastingly, concrete still technically had no experience being used as an independent pavement at the beginning of the 20th century. Up until this point, concrete had been widely used as a base course rather than a full pavement. This can be attributed to the fact that the machinery did not then exist that would allow the production of concrete and for it to be placed in the amount and to the finish required. However, in 1899, Francois Hennebique's triple arch concrete bridge over la Vienne River in France improved concrete in the minds of others. His reinforcing of the concrete with steel bars to resist cracking was revolutionary.

In 1913, the construction of about 40 kilometers of pavement near Pine Bluff, Arkansas, marked the first major concrete highway. Improved methods for mixing, transporting, compacting, and finishing concrete led to positive experiences with concrete pavements in the U.S., gaining global attention. However, cracking remained a significant issue, often caused by tension from traffic loads or restricted shrinkage. Initially, pavements featured a full-thickness longitudinal joint along the centerline, with transverse joints added later at intervals of around 15 meters, eventually reduced to 5 meters to better manage thermal and shrinkage cracks. Reinforcing steel, either as rods or mesh, could be used to minimize cracking, with transverse joints spaced about every 25 meters if reinforcement was intermittent. The introduction of vibrators for concrete placement and compaction greatly improved its quality. Another key innovation was slipform paving, where a self-powered machine shapes wet concrete into a finished pavement without side formwork. Guided by

string-lines or lasers for accuracy, the machine uses a vibrating screen for surface finishing and places steel reinforcement as needed. (Lay et al., 2020)

From ancient civilizations to modern metropolises, pavements have been integral to human progress as their quintessential characteristics evolved over time (**Figure 2**), shaping economies and connecting societies across the globe.



Figure 2: This figure gives a general timeline depicting the key characteristics of pavements throughout different eras. *This image has been generated using AI technology. Please note that any discrepancies or inaccuracies present in the image should be disregarded, as it is intended solely to provide a visual representation of the content discussed in the above section.*

As of 2018, global road networks span vast distances, with approximately 64,285,009 km (39,944,852.7 miles) of roads worldwide (De Loisy, 2019), underscoring the critical role of pavements in modern infrastructure development. Historic developments in pavement design and construction indicate that traffic type, climate, materials including foundation, equipment, labor, and capital costs are significant factors to be considered, and the following sections take a deeper dive into some of these factors.

Traffic and loads

Understanding vehicle and pavement loading dynamics is essential for designing durable and sustainable road infrastructure, both on Earth and potentially on Mars. The interaction between vehicles and pavements imposes various stresses and strains on the road surface, which can lead to deterioration and structural damage over time. On Mars, the lower gravitational force (approximately 40% of Earth's gravity) (Williams, 2015) alters the way these loads are applied to the pavement structure. Dynamic traffic loads, characterized by the repetitive application of forces from moving vehicles, pose challenges for pavement

performance. For example, heavy truck traffic can result in significant damage to an asphalt surface, creating a chicken wire cracking pattern (Machemehl & Lee, 1970/1974).

Traffic analysis is an integral part of pavement design, requiring accurate data collection and growth factor estimation for the design period. Actual traffic data, if available, provides the most reliable information. However, if unavailable, average values for different highway classes can serve as a guide.

One method of analyzing pavement deterioration has been the AASHO Road Test. The AASHO Road Test, a seminal study conducted in the midwest, laid the groundwork for understanding the impact of dynamic traffic loads on pavement performance, as shown in **Figure 3**. Central to this understanding was the development of axle load equivalency factors, essential for expressing varying axle loads in terms of a single design parameter. These factors, derived from extensive testing and analysis, quantify the equivalent effect of different axle loads on pavement structures, facilitating standardized pavement design and maintenance practices.

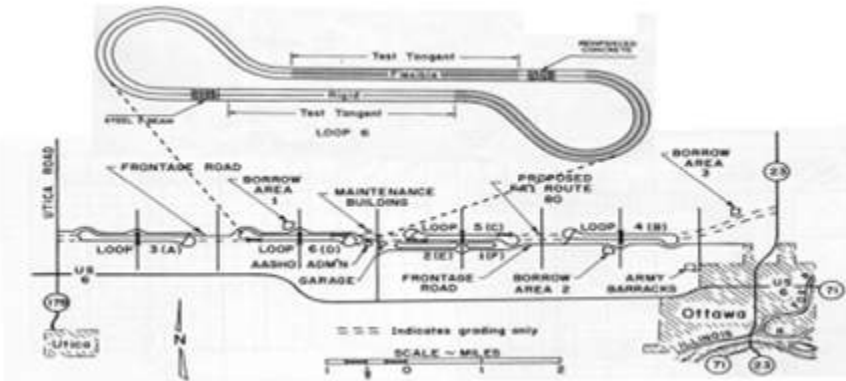


Figure 3: This figure shows the 5 loops that were exposed to traffic loadings, where each lane in the traffic loops was subjected to specific truck axle type and axle load (Dean, 2020).

Utilizing data from past traffic studies, which typically include axle weight distributions and equivalent single axle load (ESAL) values, engineers can predict future traffic patterns and estimate design ESALs. The structural number (SN) is a dimensionless value used in the design of flexible pavements, representing the overall strength of the pavement structure. It combines the thickness, strength, and quality of the pavement layers into a single value, which is used to estimate the pavement's ability to withstand traffic loads over time. For rigid pavements, slab thickness (D) is the primary design factor, reflecting the depth of the concrete slab, which directly impacts the pavement's ability to distribute loads and resist cracking. By assuming a structural number (SN) for flexible pavements or slab thickness (D) for rigid pavements, and selecting appropriate equivalence factors from tables provided, engineers can calculate the anticipated ESAL applications for a given project. This process allows for the estimation of pavement deterioration over time, guiding decisions regarding pavement design, materials selection, and maintenance strategies. However, the

accuracy of ESAL predictions relies heavily on the quality of traffic data and the validity of assumptions made during the design process (Huang, 1993).

Climate

Climate factors also significantly influence pavement performance, particularly through their effects on soil moisture content. On Earth, key climate factors that affect roads include rainfall, and temperature. Rainfall contributes to soil moisture, impacting soil's properties such as modulus and stiffness, which can lead to increased permanent deformation and reduced structural integrity of pavements. Excessive moisture can decrease effective stress due to high pore pressure, resulting in more fatigue cracking and distress in asphalt mixtures. In fact, moisture-related damage occurring on flexible, spray-sealed pavement surfaces results in a 30% performance reduction and an increased frequency of defects such as potholes or rutting. Therefore, minimizing moisture damage through pavement design improvement is an endeavor that could result in millions of dollars of savings (Kodippily et al, 2018). However, because Mars is not subjected to notable rainfall events, the impacts of rainfall may not need to be considered.

Temperature changes will be especially influential, with higher temperatures and greater temperature variations leading to increased thermal tensile stress and more longitudinal and transverse cracking. Additionally, these factors affect pavement rutting (**Figure 4**), with high temperatures causing significant deformation in asphalt layers (Qiao et al, 2013). Therefore, additional research needs to be conducted on the extreme and variable conditions of Mars, as they will require strategy to maintain pavement performance and longevity. Because the temperatures on Mars are maintained below 273 K (31.73 degrees Fahrenheit) (Fairén, 2010), strategies will have to be constructed in which pavement longevity is apparent despite the cold weather. In extreme weather regions on Earth such as wet-freeze climates, design and materials considerations are selected to withstand and provide longevity against distresses like cracking, frost heaving, material degradation, and thaw weakening (Chen et al, 2019). To achieve increased durability, asphalt binder types like anti-stripping agents are currently used in wet-freeze climate zones (Minnesota, Michigan, Wisconsin, Illinois) to prevent asphalt binder from stripping off aggregate particles in hot- mix asphalt pavement production and HMA moisture damage (Chen et al, 2019). Such considerations may be useful and will need to be evaluated to prevent similar distresses from occurring in Martian pavements.

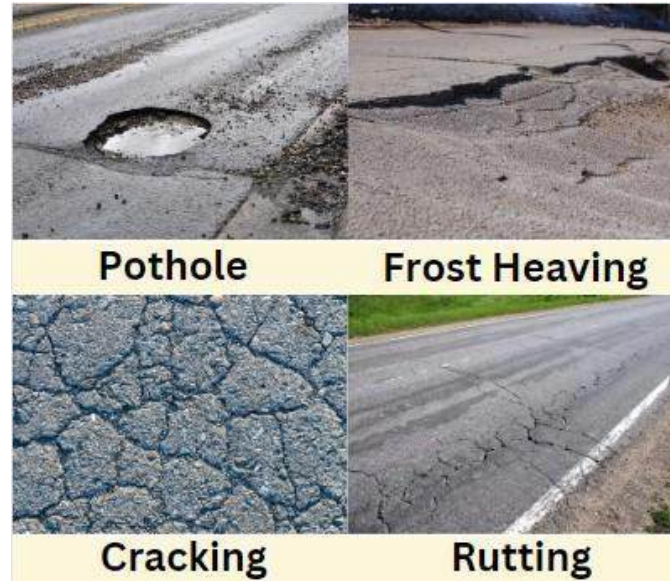


Figure 4: This figure shows common pavement deformations such as potholes, cracking, frost heaving, and rutting (*How to Fix Alligator Cracking in Asphalt Pavement*, 2021); (*Understanding Asphalt Potholes*, n.d.); (“You Asked, We Answered: What Are Frost Heaves? Are They Unique to N.H.?” 2018); (Webmaster, 2021):

Pavement materials

Pavement materials are selected to withstand loads from both traffic and climate on Earth, and determining what we can anticipate on Mars is a challenge to be considered. Construction and engineered materials need sources for raw materials, which may need further processing and manufacturing prior to use.

Here on Earth, pavement performance is significantly impacted by the uniformity of the subgrade, the layer of soil beneath the pavement that provides foundational support and stability. High-quality subgrades require an understanding of soil properties, proper grading practices, and quality control testing. Natural subgrades vary widely and are classified based on their load support and drainage characteristics, as seen in **Table 5 below**.

Subgrade Soils for Design	Unified Soil Classifications	Load Support and Drainage Characteristics	Modulus of Subgrade Reaction (k), psi/inch	Resilient Modulus (M _R), psi	CBR Range
Crushed Stone	GW, GP, and GU	Excellent support and drainage characteristics with no frost potential	220 to 250	Greater than 5,700	30 to 80
Gravel	GW, GP, and GU	Excellent support and drainage characteristics with very slight frost potential	200 to 220	4,500 to 5,700	30 to 80
Silty gravel	GW-GM, GP-GM, and GM	Good support and fair drainage, characteristics with moderate frost potential	150 to 200	4,000 to 5,700	20 to 60
Sand	SW, SP, GP-GM, and GM	Good support and excellent drainage characteristics with very slight frost potential	150 to 200	4,000 to 5,700	10 to 40
Silty sand	SM, non-plastic (NP), and >35% silt (minus #200)	Poor support and poor drainage with very high frost potential	100 to 150	2,700 to 4,000	5 to 30
Silty sand	SM, Plasticity Index (PI) <10, and <35 % silt	Poor support and fair to poor drainage with moderate to high frost potential	100 to 150	2,700 to 4,000	5 to 20
Silt	ML, >50% silt, liquid limit <40, and PI <10	Poor support and impervious drainage with very high frost value	50 to 100	1,000 to 2,700	1 to 15
Clay	CL, liquid limit >40 and PI >10	Very poor support and impervious drainage with high frost potential	50 to 100	1,000 to 2,700	1 to 15

Table 5: This table reveals the suitability of different soils for subgrade applications (American Concrete Pavement Association et al., 2013)

Achieving a uniform and supportive subgrade requires effective compaction to enhance strength and minimize long-term settlement. Compaction involves mechanically increasing soil density and is measured by the Standard Proctor Density, which determines the optimal moisture content at which a soil type achieves its maximum dry density. Cohesive soils should meet 95% of Standard Proctor density, while cohesionless soils need a minimum relative density of 65%. Proper moisture control, appropriate lift thickness, and thorough quality control are essential to avoid issues such as localized settlement and pavement cracking, particularly in areas like utility trenches and bridge abutments. Over excavation and proper filling techniques, avoiding materials like large stones and organic matter, are crucial to maintaining pavement integrity and preventing subsidence (ACPA et al., 2013).

The production of asphalt and cement for pavement surface layers involves substantial effort and sophisticated industrial processes. Asphalt is produced by heating aggregate materials and mixing them with a thinned asphalt cement (BridgeHouse Marketing, 2018). Cement manufacturing is an energy-intensive process that involves extracting raw materials like limestone and clay, heating them in a kiln to form a clinker, and grinding the clinker into a fine powder (Testbook, 2023). Other base materials such as crushed stone, gravel, and sand are extracted from quarries and processed through crushing, screening, and washing to achieve the desired size and quality.

On Mars, manufacturing base materials would likely require the utilization of in-situ resources. Examples of this could include basalt, which is a kind of igneous rock that forms as a result of the cooling of magnesium and iron-rich lava. Additionally, volcanic ash can supplement Martian pavement construction because it can react with lime and water to form hydrates that have the capability to hydraulically gel. Hence, volcanic ash can be used as a mixing material in cement and an addition to concrete after grinding has occurred (Liu et al, 2022). For binding materials, a combination of local minerals and possibly imported elements from Earth could be used to create a Martian equivalent of cement or asphalt.

Effective compaction methods, including the use of various rollers and ensuring correct lift thickness, are vital for creating a solid foundation. Regular inspection and quality control measures, such as using a Dynamic Cone Penetrometer (DCP) to estimate in-place soil stability, help ensure that the subgrade meets the necessary standards for long-term pavement performance (ACPA et al., 2013).

Materials extraction and production

Each life cycle stage of the pavement construction process has its own contribution to the overall environmental impact of the process. These contributions can be broken down as follows: Asphalt production makes up 45-95%, transport to the site accounts for 1-8%, laying and compaction is 1-12%, in situ 0-1%, and end-of-life is 0-40% (Anthonissen et al, 2016). According to Giani et al. (2015), Gschosser and Wallbaum (2013, and Gschosser et al. (2014), extraction and material production have the greatest environmental impact in the life cycle. Therefore, it is important to consider the environmental impact of materials extraction as a significant factor when constructing pavements on Mars. The harsh Martian environment—with its thin atmosphere, extreme temperatures, and pervasive dust—presents unique challenges for traditional extraction and production methods. The energy-intensive nature of materials extraction, which relies on solar or nuclear power, adds another layer of complexity. Efficient use of energy and resources will be crucial to minimize the environmental footprint of these activities.

Furthermore, unforeseen challenges will inevitably arise as Martian pavement construction moves from theoretical plans to actual implementation. These might include the difficulty of operating machinery remotely or autonomously in a harsh and variable environment, unexpected interactions between Martian dust and construction materials, and the potential for equipment malfunctions due to the extreme conditions. Developing adaptable technologies and strategies to address these challenges will be essential for successful and sustainable pavement construction on Mars. The innovative solutions created to overcome these challenges could also provide valuable advancements for construction practices on Earth.

Pavement construction

The challenges that will come with the labor and construction of pavements on Mars require special consideration. The environment on Mars presents numerous hazards, including extreme temperatures, high levels of radiation, and a thin atmosphere, making it crucial to consider the challenges of labor and construction for pavements on the planet. Current pavement construction machinery is insufficient for such hazardous environments due to their labor-intensive nature. Therefore, the development of autonomous machinery is essential for safe and efficient construction on Mars. To address this need, research and development efforts are underway to create autonomous robots. For instance, researchers at Ohio University are working on an intelligent concrete construction system that enables real-time remote control of paving operations (Maynard et al, 2012). The recent innovations in the use of drones in the construction process, which has increased efficiency and accuracy in construction (Mahajan, 2021), demonstrate a promising avenue for creating efficient pavements on Mars.

Associated construction costs for building roads should also be noted. These costs tend to vary to a great extent based on location, specifications, and additional infrastructure requirements. For a major road with 2 lanes, each 12 feet wide, and two 3-foot shoulders, costs range from \$5.34 million per mile (\$3.34 million per km) in the Northeast USA to \$6.04 million per mile (\$4.04 million per km) in the Southeast USA. For major freeways or interstates with four lanes and shoulders, costs can rise to \$19.22 million per mile (\$12.01 million per km) in the Northwest USA, and up to \$20.31 million per mile (\$12.69 million per km) in the Southeast USA. Elevated freeways in urban areas have the greatest costs, reaching \$68.45 million per mile (\$42.78 million per km) in Central USA and \$71.33 million per mile (\$44.58 million per km) in the Southeast USA (*Order of Magnitude Road and Highway Costs* | *Compass International*, 2023).

These costs include clearing, excavation, stone, paving, concrete, reinforcement, structural steel, safety barriers, labor, equipment, and design and construction management, with an accuracy of +/-20%. In fact, the total highway and street construction spending in the US has increased by nearly \$82 billion over the past 2 decades (**Figure 6**). When considering pavement construction on Mars, these costs are expected to increase drastically because of required specialized autonomous machinery, materials transport from Earth, and unique Martian environmental challenges.

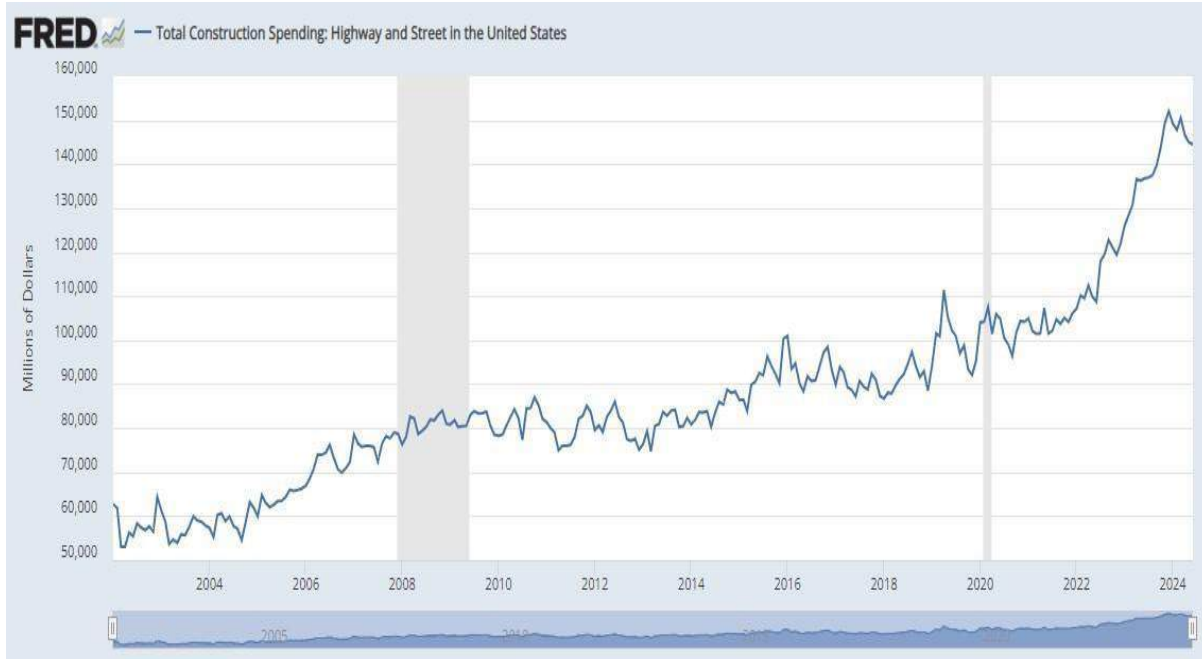


Figure 6: This figure shows the total cost of highway and street construction in the US as it has increased over the last 20 years, highlighting that these already high costs on Earth will only increase further due to factors outlined above (*Total Construction Spending: Highway and Street in the United States, 2024*).

A note on pavement maintenance

Maintaining pavements on Mars poses unique challenges, especially for pavement asset management systems (PAMS). Over the past two decades, advancements in computer vision and AI have enhanced engineering applications that manage large datasets, crucial for PAMS in providing tailored solutions for roadway maintenance. However, adapting these technologies to Mars's extreme conditions and lack of infrastructure makes integrating AI and automation vital.

PAMS decisions are data-intensive, with concerns about the quality and cost of automated data collection. On Mars, where human presence is minimal, reliance on automated systems is essential. Current cost-effective methods like smartphones and UAVs, limited to specific distresses such as cracks and potholes, must be standardized for broader applications, given the impracticality of manual inspections (Peraka & Biligiri, 2020). However, on Mars, where human presence is minimal, reliance on automated systems is essential.

Data analysis also presents challenges due to high costs and complexity. While image processing and machine learning techniques have been applied to detect pavement distresses, their potential for broader applications, such as performance prediction and maintenance prioritization, remains largely unexplored (Peraka & Biligiri, 2020). These capabilities will be crucial for maintaining pavement infrastructure on Mars.

Establishing a standardized pavement condition index is another critical issue, as varying scales and variables currently cause inconsistencies in maintenance decision-making.

On Mars, a unified index incorporating the planet's unique conditions will be essential for effective maintenance planning.

Finally, a significant gap exists between research and practical application. Testing proposed cost-effective studies and DL technologies on a larger scale could shift PAMS from data-driven to technology-driven, aligning with sustainability goals. On Mars, this shift will be crucial, as autonomous systems must manage pavement maintenance with minimal human intervention, reducing risks in the hazardous environment.

Contrast, challenges, and conclusions

Anticipated roadway pavements on Mars may follow similar principles to those on Earth, however there are several caveats due to the extraterrestrial idiosyncrasies of Mars. This paper examines important aspects of the pavement process, highlighting the comparison of future Martian pavements to those on Earth. The findings derived from this exploration are as follows:

1. The evolution of pavement construction across time on Earth offers invaluable insights for developing road infrastructure on Mars. From the durable stone pavements of ancient Egypt and Mesopotamia to the sophisticated road networks of the Romans, the fundamental principles of road building—utilizing local materials, ensuring structural integrity, and facilitating drainage—remain consistent throughout civilizations. These principles can be directly applied to Martian pavement construction, however adapted to the unique conditions of the Martian surface.

As history has shown, the materials and methods used in roadway construction evolved to meet the specific needs and conditions of their times. In the same way, Martian pavements will need to be designed with the planet's extreme conditions in mind, such as its low gravity, harsh climate, and resource limitations.

2. The study of vehicle and pavement loading dynamics, though applicable both on Earth and potentially on Mars through understanding core concepts like vehicle-pavement interactions, load equivalency, and traffic analysis, requires specific adaptations and considerations on Mars.

One notable difference will be the characteristics of vehicles operating on Mars. Unlike Earth, where roadway pavements are designed to accommodate a wide range of vehicle types, from passenger cars to heavy trucks, Martian explorers will need to consider potential vehicles and their characteristics. These vehicles may be specially engineered to navigate the Martian conditions, handle unique environmental challenges, and utilize different propulsion systems. As a result, the loads they impose on pavements will vary, influencing the stress and strain dynamics differently than on Earth.

3. A critical aspect of pavement construction is the uniformity and quality of the subgrade. On Earth, achieving a stable subgrade involves assessing soil properties,

proper grading, and quality control. Similar principles are expected to apply on Mars, where the use of local materials like basalt and volcanic ash could provide the necessary support and stability.

The production and use of materials for pavements on Mars will also draw parallels to Earth's practices, involving processes like the extraction and processing of aggregates, and the manufacturing of binding agents similar to cement and asphalt. The utilization of Martian regolith, combined with the development of new materials suited to the Martian environment, will be essential. Moreover, effective compaction and quality assurance practices must be adapted to the harsh conditions on Mars, which may affect material properties, to prevent pavement deformation, cracking and settlement.

4. On Earth, rainfall and temperature influence the structural integrity of pavements. These factors contribute to challenges such as increased permanent deformation, cracking, and moisture-related damage like rutting and potholes.

Therefore, the extreme conditions on Mars, such as mostly low temperatures further challenge pavement design and longevity. As Mars experiences temperatures well below freezing, strategies must be developed to ensure pavement durability and performance under such cold conditions. However, Mars' temperatures also experience significant fluctuations, making Earth materials possibly unavailable or a poor fit to the temperature variation on the red planet. Therefore, in situ materials and design considerations should be engineered to suit the climatic conditions of the planet.

5. Each life cycle stage of pavement—from materials production to end-of-life—consumes energy and contributes to the overall environmental impact, with extraction and material production being the most significant contributors. Being on Mars complicates traditional extraction methods and energy use, necessitating innovative and efficient approaches.

Effective energy use on Mars will be crucial for minimizing the environmental footprint of pavement construction. Moreover, the development of in situ energy sources is necessary to yield the required energy provision in an uninterrupted and efficient manner. To this end, a critical future research direction could be the further exploration of technologies aiming to convert radiant energy from the lunar surface into usable electricity. Additionally, Recycling Martian regolith for construction materials could reduce the need for energy-intensive material transport from Earth.

6. Current pavement machinery and construction process, which is labor-intensive, is inadequate for Mars' hazardous conditions. Thus, the development of autonomous machinery is critical, with ongoing research, such as the intelligent concrete construction systems at Ohio University, aiming to address this need.

Cost considerations further complicate Martian pavement construction. On Earth, construction costs vary widely depending on location and project specifications,

ranging from \$5.34 to \$71.33 million per mile. When applying these costs to Mars, the expenses will be significantly higher due to the need for specialized machinery, the transport of materials from Earth, and the challenges posed by the Martian environment. Therefore, While the basic principles of pavement construction provide a foundation, the successful implementation of Martian pavements will require overcoming substantial technical and financial hurdles.

7. One of the main challenges on Earth is related to the timely maintenance of pavements and can pose unique challenges on Mars. the reliance on advanced Pavement Asset Management Systems (PAMS) which are integral to managing pavement condition and performance. On Earth, these systems benefit from advancements in computer vision and artificial intelligence (AI) that improve data handling and analysis. However, on Mars, the lack of human presence and existing infrastructure amplifies the need for highly automated systems for maintenance of pavement infrastructure and assets. The cost and quality of automated data collection remain of substantial concern. Current cost-effective methods, such as smartphones and unmanned aerial vehicles (UAVs), are primarily used for detecting specific distresses and need adaptation for network-level applications on Mars, where manual inspections are not feasible.

Additionally, the complexity and cost of data analysis present obstacles. While deep learning (DL) techniques show promise for broader applications, including performance prediction and maintenance prioritization, their use on Mars must address the planet's harsh conditions and limited resources. A unified pavement condition index tailored to Martian specifics is crucial for effective maintenance planning. This standardized index will support accurate and reliable maintenance decisions, essential for enduring the planet's severe weather and terrain.

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Harmonizing the Cosmos: An Algorithmic Approach to Converting Stellar Data into Music By Jarmin Weng

Abstract

At night, stars fill the sky, and kids, adults, and seniors alike unite to stare at a seemingly random configuration of bright specks of light. But these stars have a story to tell: all through the constellations formed in the night sky. Unfortunately, millions of people in the world who are visually impaired do not have the luxury of enjoying the intricacies of the night sky and constellations to their fullest. Music lovers like me would also appreciate a sound representation of constellations. This project aims to design an algorithm that translates the physical properties of a constellation's stars into musical sounds and plays them together as a chord. Integrating astronomical data with music theory can aid in exploring new auditory experiences of celestial objects and the beauty of the night sky. The methodology involves a JSON library to store the necessary star data from the website "TheSkyLive.com", Wien's displacement formula to convert the temperatures of stars to usable sound wavelengths, and the Javax sound package to generate the sound. The musical chords produced encapsulate the stories behind the constellations, and the corresponding melodies have the potential to serve as leitmotifs in film music and music education. Sonifications as an auditory method of expression have the potential to revolutionize data interpretation by making complex information more accessible and engaging and fostering new interdisciplinary collaborations between scientists and artists like me.

Introduction

The origin of constellations is not certain, but the Egyptians and ancient Chinese were known to apply patterns to help discern stars in the night sky. The constellations we know today follow those of Ancient Greece in Ptolemy's *Almagest* (Royal Observatory Greenwich). Dependence on the sky became essential for many early cultures and also many agricultural cultures today. Farmers today need a great deal of knowledge of the night sky, as understanding celestial movement is essential for predicting precipitation and weather. Additionally, constructing sites that track celestial movement like Stonehenge in England and the pyramids in Chichen Itza requires immense precision, and constellations would aid in this detailed work.

Interdisciplinary strategies combining technology and the arts, specifically music, have an extensive history. One of the famous works of the past few centuries involving music and science is Gustav Holst's "The Planets Suite," written in 1918 (BBC Music Magazine). Holst, as he was entering his forties, became fascinated with theosophy and astrology, which resulted in the creation of an orchestral suite in seven movements. It quickly became one of the most well-known orchestral pieces in British classical music history. Every movement describes a planet in the solar system, including the vicious 'Mars, the Bringer of War' and the triumphant 'Jupiter, the bringer of Jollity'. Creative American composer John Cage wrote a solo piano

work titled “Etudes Australes” in 1975, which directly translates from French to “Southern Studies” (Liebner). That is exactly what the work was: a collection of 32 virtuoso studies based on maps of the southern night sky. From these maps, Cage randomly selected the locations of planets and translated them into pitches. This work thus shows the beauty of the seemingly random nature of the night sky.

Today, the digital era has elevated the uses of this interdisciplinary approach. Connecting the night sky with music is not new. Connecting planetary stories with music has already been done. Connecting constellations, the ultimate stories of the sky, however, has not been explored thoroughly before. This project aims to create a unique set of notes for each night sky constellation while also investigating the potential uses of these sonifications. The stellar data for each constellation were extracted from an online source, and are converted in Java to musical elements through direct relationships involving Wien’s law.

Literature Review

Since 2020, the Chandra X-ray Center, in collaboration with NASA, has produced numerous sonifications for 2D images of stellar objects (Chandra X-ray Center). They typically produced a sound from each column of pixels, where every pixel of the image was assigned pitches and volumes. NASA writes that: “No sound can travel in space, but sonifications provide a new way of experiencing and conceptualizing data” (NASA). From the videos posted on their websites, it appears that the majority of the notes created are autotuned to a specific scale: typically a major pentatonic scale, making the result more pleasing to the ear. However, the sounds produced by NASA are not representative of the stellar objects, but rather the 2D images of the objects, as different images of the same object would produce drastically different sonifications. This project aims to use the data of stars in the constellation itself to create a unique sonification that is representative of the constellation.

Relationships between color/wavelength and pitch have been investigated extensively. Flute and sound encyclopedia Flutopedia outlines the relationship between color and sound on its page titled “The Color of Sound.” It introduces the many approaches to the sound-colour relationship over the centuries: either indirect or direct (Goss). The former describes the assigning of a discrete sound to a discrete color, while the latter describes a formula or mapping between a pitch and a color that often forms a continuous spectrum of sounds mapping to a spectrum of colors. An example of an indirect sound-colour relationship is the Scriabin correspondence. Generational composer Alexander Scriabin (1871–1915), based on his experience with synesthesia, a condition that causes one sense to be perceived as a different sense, mapped distinct keyboard pitches to specific colors. He thus created the Clavier à Lumières system as in Figure 1. Regarding a direct relationship, the website suggests converting the frequency of sound to a frequency of light by doubling the sound frequency until it reaches a frequency in the range of 400–800 THz. This method essentially increases the sound by a musical octave until the note’s frequency reaches the range of visible light. However, this direct relationship in reverse (mapping light to sound) would often

produce unpleasant sounds. This project will utilize this mapping method in reverse but will incorporate an autotuning method.

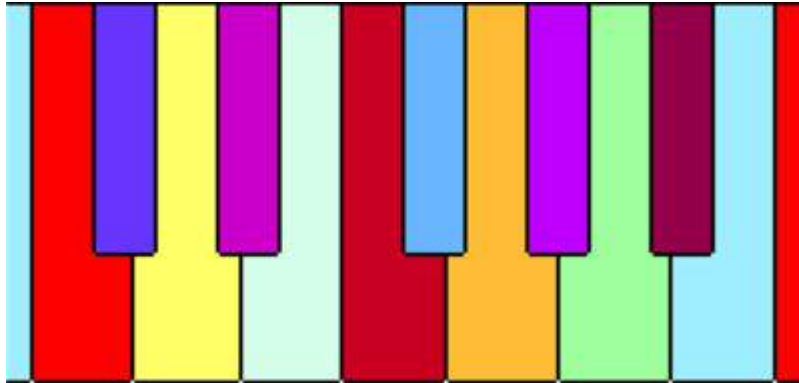


Fig 1: The Clavier à Lumières system⁶

Methods

The algorithm comprises an original dataset and multiple programs in the programming language Java. The temperature and visual magnitude of the individual stars of the constellations, collected from the website “TheSkyLive.com,” are inserted into a JSON database.

The ‘StarDataExtractor’ class reads the JSON file containing the stellar data with the use of a FileReader. The JSON data is parsed to extract information for specific constellations and is stored in a HashMap where the key is the constellation name and the value is another HashMap containing the individual star data, in the form of object ‘StarData’. The ‘StarData’ class is a data structure that stores information about each star, including the star’s temperature, magnitude, and wavelength, and also provides methods to set and retrieve these properties.

The ‘SoundGenerator’ class converts the stellar data into sound and, in the process, maps star properties to sound parameters (frequencies and amplitudes) and generates sound with a chord to represent constellations. First, the temperature of the star, which is representative of the color of the star, is converted into a corresponding wavelength using Wien’s law:

$$\lambda_{max} = b/T$$

, where λ_{max} is the peak wavelength in nanometers (representative of the most dominant color), $b \approx 2.89777 \times 10^6$ is a proportionality constant, and T is the absolute temperature in Kelvins. This wavelength is then converted to a corresponding frequency by dividing the speed of light in a vacuum by itself. This is then mapped to a corresponding base sound frequency by repeatedly dividing this light wave frequency by 2 until it reaches the range of the first audible octave: under $(30.5+32.7)/2 = 31.6$ Hz, halfway between the frequencies of notes B0 and C1. The resulting base frequencies are then mapped to the frequencies of the nearest keyboard tone. The star's visual magnitude is inversely mapped to the relative

amplitude of the sound wave, as the smaller the visual magnitude, the brighter the star. Although the visual magnitude is exponentially related to a star's luminosity, it is not mapped to the sound amplitude using an exponential function, as it would result in too much variation in the resulting sound amplitudes, where the constellation would be mapped to a set of notes that were either virtually inaudible or extremely loud.

To make the notes audible, the resulting discreet note frequencies are all translated up 5 octaves by multiplying the note base frequencies by 2⁵ times. These star properties individually form waves that combine with the other waves to create a chord. The generated samples are normalized to prevent clipping and distortion, where the relative sound amplitudes are adjusted so that their absolute values do not exceed 1. This is done by finding the maximum absolute sample amplitude and, if it is greater than 1, scaling all samples by this maximum value. To play the sound, an 'AudioFormat' object is created with the following parameters: a CD quality sample rate of 44100, 16 bits sample size, 1 channel, true for signed, and false for big-endian. A 'SourceDataLine' is then obtained from the 'AudioSystem' and is opened and started for playback. The audio samples are then converted from double precision to 16-bit signed integers, packed into a byte array, and written to the 'SourceDataLine'. After writing and playing all the samples, it is drained, stopped, and finally closed.

The frequencies and amplitudes could also be interpreted in different ways. For example, the stars used to form the shape/pattern typically drawn could be used to create a melody. This method is explored in the results section.

Results

Results from a few constellations from the 88 analyzed are shown below:

Table 1. Musical Data for the Stars in Constellation Lyra

Star name	Temperature (K)	Frequency (Hz)	Nearest musical note	Visual magnitude	Relative amplitude
Vega	10125	1048220	C	0.03	33.3
13 Lyrae	3663	379223	F#	4.04	0.248
Sheliak	10125	1048220	C	3.45	0.290
Sulafat	10795	1117584	C#	3.24	0.309
δ2 Lyrae	3541	366592	F	4.3	0.233
κ Lyrae	4368	452210	A	4.33	0.231

The constellation Lyra is visually dominated by one of the brightest stars in the night sky, Vega. The chord it produced is similarly dominated by Vega's musical note, the middle C. The chord produced, C-C#-F-F#-A, is a very mysterious-sounding chord that would be typically.

Table 2. Musical Data for the Stars in Constellation Andromeda

Star name	Temperature (K)	Frequency (Hz)	Nearest musical note	Visual magnitude	Relative amplitude
δ Andromedae	4158	430469	G#	3.27	0.306
Alpheratz	11744	1215831	D	2.06	0.485
Mirach	3677	380672	F#	2.06	0.485
Nembus	4158	430469	G#	3.57	0.280
\omicron Andromedae	11407	1180943	D	3.62	0.276
Almach	4000	414112	G#	2.26	0.442

The constellation Andromeda describes the daughter of Queen Cassiopeia and King Cepheus of Greek mythology, who has the nickname of the Chained Lady as her parents put her in chains and offered her as a sacrifice to the sea monster Cetus. The chained lady's cries are well heard within this chord. The three brightest stars, Almach, Alpheratz, and Mirach form the chord G#-D-F#, a dissonant chord that would suit to represent a loud dissonant screech if played at a high register.

Table 3. Musical Data for the Stars in Constellation Crux

Star name	Temperature (K)	Frequency (Hz)	Nearest musical note	Visual magnitude	Relative amplitude
Ginan	3918	405622	G	3.59	0.279
Acrux	14633	1514924	F#	1.33	0.752
Gacrux	3663	379223	F#	1.63	0.613
Mimosa	14354	1486039	F#	1.25	0.800
α 2 Crucis	15229	1576626	G	1.73	0.578
Imai	14354	1486039	F#	2.8	0.357

The constellation Crux, also known as the “Southern Cross,” forms a cross between four stars in the night sky. The “Jewel Box” cluster, or the κ Crucis Cluster, resides in the constellation, and the constellation’s musical chord is exactly that: a simple cluster of two notes, G and F#. A melody that alternates between these two notes, G and F#, would resemble the tolling of a bell.

Table 4. Musical Data for the Stars in Constellation Cassiopeia

Star name	Temperature (K)	Frequency (Hz)	Nearest musical note	Visual magnitude	Relative amplitude
γ Cassiopeiae	12489	1292960	D#	2.47	0.405
Shedar	4368	452210	A	2.23	0.448
Ruchbah	8748	905662	A	2.68	0.373
Caph	7217	747161	F#	2.27	0.441
Segin	12489	1292960	D#	3.38	0.296
Achird	6088	630278	D#	3.44	0.291

Queen Cassiopeia's constellation produces a chord quite similar to that of her daughter Andromeda. The chord of Cassiopeia, A-D#-F#, forms the notes of a diminished triad, with a very sad tone. This sad, defeated, and almost regretful tone of this diminished triad shows Cassiopeia’s emotions after she was forced to sacrifice her daughter to soothe the god Poseidon’s wrath.

The frequencies and amplitudes could also be interpreted as a melody. The stars that make up the “W”-like shape in the constellation Cassiopeia follow the order: Segin, Ruchbah, Gamma Cassiopeiae, Shedar, and Caph (Editors of Earthsky). The 5 stars’ nearest musical notes could be used in that order to form a representative melody, and as the stars have relatively similar visual magnitudes/brightness, each note is assigned the same duration.



Fig 2: Cassiopeia's melody

Discussion

It is amazing how in music, one note can completely change the tone of a chord. From a musical interval perspective, the difference between Cassiopeia and Andromeda's chord is just a half-tone. If the nearest note of Caph, the second brightest star of Cassiopeia, were a half-tone higher (G), it would have the same tone as Andromeda's chord. It is also fascinating how the constellation's meaning and the musical chords its stars create complement each other. The musical chords well represent the meaning of the constellations, and every musical chord is unique.

The melody of Cassiopeia is short but effectively captures the sad and regretful story of the Queen of Aethiopia. Such a short melody in music could be used as a leitmotif (Sones). A leitmotif, German for "leading motif," marks a musical phrase representing a certain character, place, or idea. German composer Richard Wagner's development of this technique, most famously in the music drama cycle "The Ring," was a catalyst for its use in film music. Famous film themes today, such as John William's "The Imperial March" and multiple themes in Howard Shore's *The Lord Of The Rings*, all employ leitmotifs in film. The potential of these short constellation themes as leitmotifs is huge. For example, if the story of Andromeda and Queen Cassiopeia were to be a film, the melody of the two respective constellations could be used as leitmotifs, effectively representing the atmosphere and characters of the film.

There are a lot of other possibilities for using this research too. In astronomy education, these audio tools can bring stars and constellations to life through exciting lessons. Entertainment - Immersive experiences in planetariums and museums that take you on space adventures with out-of-this-world sounds that actually represent the night sky. Converting astronomical data into sound can help bridge the gap between astronomers and the public, allowing complicated scientific ideas to become viable for a wider audience while attempting greater outreach in science communication.

Conclusion

This project demonstrates the possibility of converting star data into chords of music by visualizing and using technical methods to create a unique auditory experience for constellations. Wien's displacement was applied to convert the star temperatures into sound wavelengths, and the sound was generated with the Javax sound package. The algorithm

transforms the temperature of stars and visual magnitude to sound pitch parameters. The generated chords, which echo the historical and mythological connections of star patterns in our skies inspire an alternative way to communicate science. This approach has the potential to elevate research in astronomy education by adding a layer of soundscape immersion that enables people who are visually impaired or disadvantaged. Additionally, the generation of melodies from the constellations could be used as leitmotifs, which could be employed in film music. It also presents new avenues for engaging experiences at planetariums and museums. The algorithm could be further refined with a full background database of the stars in each constellation, allowing the night sky to sound complete.

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Labor and Awakening: An Oral History of a Female Migrant from Shanxi to Inner Mongolia during the Great Leap Forward By Rongyu Zhou

Abstract

This study investigates internal migration experiences during the Chinese Great Leap Forward Period (1958-1961). Through an interview with Fanghua Li, a Chinese woman who began migrating in 1960, an understanding of how this transformative era influenced her ideals and awakened her gender consciousness is gained. While prior studies on the Great Leap Forward have primarily relied on large-scale data and analyzed the era from a top-down, macro perspective, this research emphasized the importance of individual life history, values, and attitudes towards gender and work, this study aims to examine how she compared herself to men and to other women before and after the internal migration, therefore to demonstrate the emergence of her gender consciousness and the societal realities that prevented her from becoming fully aware of gender equality phenomenon happening in her life.

The Chinese Great Leap Forward Period seems like a transformative era that propelled China towards modernity (MacFarquhar and Fairbank). In order to explain an individual's experience during this period, this article's theoretical framework is built on an analysis of feminism, labor, and modernity. Using those theories, this study illustrates how Fanghua moved from the private to the public domain for perceived liberation and came to understand the significance of attaching value to domestic work by examining modernism and industrialization beliefs. Furthermore, to better understand the experiences of Fanghua during the Great Leap Forward Period, this study delves into theories of women's labor valorization. This concept refers to the recognition and appreciation of the economic and social value of women's work, both paid and unpaid. By valuing women's work, labor valorization aims to challenge the gendered division of labor that often undervalues and marginalizes women's contributions to society and the economy. This, in turn, helps to lessen the perceived distinction between men and women in the contemporary world of economic output valuation, which is grounded in theories of work and gender. By examining this concept, the study aims to provide insights into how women's labor was perceived and valued, and how this influenced women's different experiences and perspectives.

The academic consensus generally recognizes three major waves of feminism to date (Rensen & Guo & Paludi). In feminist studies, the relationship between gender and power has consistently been a crucial research theme. The dominant tendency within various Western feminist schools is to interpret "gender equality" as the "equality of rights" on the basis of "sameness" between the sexes. This approach seeks to secure equal rights and opportunities for women by minimizing gender differences, with its fundamental principles largely rooted in the ideas of modern liberal feminism (Guo). In China, scholars have also observed the relationship between feminism and power. The concept of "women's rights" in modern China developed in tandem with contemporary notions of "civil rights," intertwining the capabilities, rights, and status of Chinese women with the national imagination. This relationship reflects

the belief among intellectuals of that era in a prosperous future for China. Consequently, in the context of modern Chinese thought, the idealized image of women transcended specific binary gender structures and became a mirror for envisioning the future nation (Zhai). In this paper, salary is examined as a significant manifestation of power. Through a feminist lens, the analysis explores how salary confers greater status and power upon the subject of this study, and how this, in turn, influences her gender consciousness.

On the other hand, some scholars have interpreted the impact of feminism from the perspective of compliance. For instance, Judith Butler, building upon Foucault's critique and deconstruction of the "subject," posits that the "intrinsic essence" of gender is, in fact, a product of repeated behaviors that conform to gender norms. Gender identity, according to Butler, is constituted through the continuous performance of these behaviors. (McIntosh and Butler). This paper discusses a case study of a woman whose gender consciousness awakens amidst social transformation but who continues to comply with gender roles. The analysis focuses on how her actions, taken in conformity with familial and socially constructed gender roles, reflect the limitations of her gender consciousness within the context of her time.

Theoretically, by delving into the life history of Fanghua during the Great Leap Forward Period, this research aims to contribute to the existing literature on this transformative era through an individual's perspective. Drawing on the theories of modernity, labor, and gender, this study argues that macro-level policies implemented during the Great Leap Forward radically restructured the trajectory of individual life. By examining the life history of Fanghua, the study highlights how personal growths, especially the gender awareness were embedded in the realities of the economy and politics of the time. Furthermore, the development of society's economics and politics resulted in the emergence of gender consciousness, although full awareness of gender equality was constrained by societal norms and structures.

Methodology

A qualitative approach is employed that mainly includes gathering oral histories through conducting interviews. The interviewee, Fanghua Li (alias), was born in 1943 in Shanxi province and migrated to Inner Mongolia at the beginning of 1960 during her 17th. She worked as an apprentice and laboratory technician at Baotou Aluminum Manufacturing and went to work at Baotou Electric manufacture in 1974. She met her husband in the Inner Mongolia factory, married, and had 3 children. Later on, she moved to Beijing in 57 years old to take care of her grandchildren and stayed there for 24 years.

Notably, Fanghua Li is also the grandmother of the researcher. Therefore, the interview for the oral history collection lasted 3 hours and 25 minutes in-person at Fanghua's home, but given Fanghua's relationship with the researcher, the paper also relies on ordinary, everyday contact between them. To validate the information gathered from the interview, the researcher uses secondary resources.

Additionally, the analysis also relies on old photographs to get visual representation of Fanghua's life in the past. This methodology allows for a more personalized and nuanced understanding of the experiences of the a Chinese woman during the Great Leap Forward Period, and provides a unique perspective on how macro-level policies affected individual lives.

As for the structure, this paper will first give a brief introduction of the background of Great Leap Forward and the condition that brought Fanghua to Inner Mongolia. Then the paper will display the story of the individual through sections of her comparing herself before and after migration, comparing herself with other women in rural areas, and comparing herself with men to show historical era' influence and policy on individuals. Following that, analysis of theories of labor valuation and gender helps understand individuals' experience and change of ideology. Lastly, through the examination of Fanghua's experiences, this research provides valuable insights into how macro-level policies shaped individual lives and how individuals navigate societal norms and structures. This research contributes to the existing literature on the Chinese Great Leap Forward Period by examining an individual's life history, by highlighting the significance of women's labor valorization and gender consciousness, this study offers a unique perspective on the transformative era of the Great Leap Forward and its impact on individuals.

Historical Background of the Great Leap Forward

The Great Leap Forward was a period of rapid industrialization and collectivization in China, initiated by the Communist Party of China under the leadership of Mao Zedong in 1958. It was a response to problems caused by the first Five-Year Plan and as a means of transforming the nation from an agrarian economy into an industrialized society comparable and superior to Western developed countries. Prior to the Great Leap Forward, China was utilizing a Stalinist model of economic development, which concentrated on developing heavy industry and generating resources for industrialization from an impoverished pre-industrial sector of the economy. This model had led to existing economic controversies, such as unbalanced development between heavy and light industries and agricultural stagnation (Duara).

By initiating the Great Leap Forward, Mao and the CCP aimed to address agricultural stagnation by developing small-scale industries that would absorb the surplus and convert labor into a "kind of investment," While avoiding diverting any resource from the main economic investment-heavy industry to agriculture (Team).

This new economic strategy adopted by China involved mass mobilization of unemployed or under-employed labor, including women in rural areas like the individual in the case study, to participate in labor-intensive investment projects, as the Chinese economy prioritized the growth of both heavy and light industries. The approach involved restructuring the countryside and the city and demographic reorganization as a large proportion of

agriculturalists in the countryside moved to industries in cities. This provided individuals with new opportunities and identity.

Specifically, during the 1960s to 1980s in China, the urgent need for labor working in labor-intensive industries led to a shift in working patterns and more women attending the industry. Thus, in order to make women stay motivated, the CCP utilized political propaganda such as saying women are as strong as men and offering them names such as “iron women” (Zhou & Guo). These popular sayings made women believe that they were contributing to the economy and national development, and that they had the virtue of actively participating in societal transformation. Moreover, policies like “Tong gong tong chou”, which means equal pay for the same job no matter gender difference, further encouraged the sharing of workloads and benefits among male and female workers in a workplace or community, with the aim of promoting equality and fairness.

However, this propaganda of “de-gendering description”, which describes women and men as having the same strength and ability, was perceived by women as “equality”, which resulted in erasing gender differences and forcing them to perform the same amount of work as men in productive labor, regardless of their biological characteristics. This also led many women in that era to work regardless of physical and physiological differences, resulting in health issues. (Zhou and Guo).

Despite the unrealistic goals set by Mao during the Great Leap Forward, such as China becoming the world’s leading steel manufacturer with 100 million tons by the end of 1962, an increase of 2,000 percent in just five years, the initiative has had a significant impact on China’s modernity and labor practices (Brown). However, the unrealistic goals also led to widespread famine and starvation, with peasants resorting to extreme measures to survive, including cannibalism (Brown). This research analyzes the gender and labor issues that arose under this background. In the following part, I will further consider the individual experience and gender awareness of Fanghua within this context.

The Start Point: Fanghua’s Migration to Inner Mongolian

This section explains the starting point and reasons for Fanghua’s migration in order to show the beginning of the process of awakening gender consciousness and reveal that the era and the social realities influence individual experiences and individual values. Ironically, Fanghua’s migration from Shanxi province to Inner Mongolia to escape mandatory labor to build the local reservoir became the beginning of her becoming a more important member in the construction of socialist modernity:

“Later, I thought in a selfish way. If I can’t find a job [outside] the village cadre will force me to work in the reservoir. It was just after the Chinese New Year, in the cold winter. Later, my father told me, if you don’t leave, you will have to go work in the reservoir! It will freeze you to death!”

The start point clearly shows how individual experiences and decisions are influenced by broader social and economic realities. Fanghua’s family saw the opportunity for her to

move to Inner Mongolia as a way to escape mandatory labor for the reservoir construction and potentially find better work opportunities. Building a reservoir in 1960s is in line with the background of Shanxi during the era, because from 1959 to 1961, there were serious natural disasters in China, which were “unprecedented in the 1950s” and affected 44.63 million hectares of land ¹. And in 1960, Hebei, Shandong and Shanxi (Fanghua’s home) were the most severely affected, accounting for more than 60% of the cultivated land area being affected by 3 years natural disaster. Thus, building a reservoir to store water to cultivate land was a good solution to ensure food supply. This marked the beginning of her later work in the factory, and it was also the beginning of the gap between her as a working woman and ordinary rural women, which will be deeply analyzed later.

After this work experience, Fanghua was able to find work in Inner Mongolia with the help of her cousin Tianxiang, thanks to the policies of the Great Leap Forward. As mentioned, Great Leap Forward policy aimed to develop Chinese agriculture and economy, resulting in high ^[1]demand for workers in government-run companies. Fanghua’s migration to Inner Mongolia allowed her to take advantage of these policies and seek better work opportunities, which was a way of escaping the harsh labor that she faced in her village. This shows how government policies can have both positive and negative impacts on individuals and how individuals can navigate these policies to improve their own circumstances.

Recalling the process of her entering Inner Mongolia, Fanghua mentioned it was largely the needs of the times and policies that forced her to build the reservoir. She vividly described the process of her family members telling her to leave Shanxi to escape the forced labor.

My cousin Tianxiang suggested that I move to Inner Mongolia for work. He told me this one day before I left the village. At that time, I had another cousin who was three years older than me. She was 21 years old and was sick. Her mother had passed away, and she was left alone at home. I went to visit her, and I found her in a dire condition. She couldn’t sit up on her own and needed me to hold her. She was dying, and her father was also present. Initially, he refused to let me go because he wanted me to take care of his daughter.

Then my father came looking for me, and he called my name. He was worried to death and said, “If you don’t go, you have to work in the reservoir tomorrow! It’s tiring!” He was so anxious that he scolded me and asked if I was dead. I quickly said, “My father scolded me, I have to go.” My uncle eventually allowed me to leave my second cousin, but unfortunately, when I returned home, I found out that she had died. I didn’t know what happened to her after I left.

In this story, Fanghua’s personal feelings and the needs of the times collided violently. Fanghua describes her experience of hearing the “news” from her cousin Tianxiang emotionally with huge sadness and a sense of urgency. When she was informed of the option of moving and being informed by her male cousin Tianxiang to move to Inner Mongolia for work, she went to her female cousin Yajing’s house to visit her. For whatever reasons for the visit, Fanghua’s immediate action to call the meeting must signal Yajing’s importance to her.

As Yajing died, the story is overlaid with a layer of sorrow, which implicitly reflects Fanghua's grief caused by the departure from her hometown as she brought up the death of Yajing when being asked about the feelings of migration. As a young woman, leaving the family must have been a tough and frightening thing to Fanghua thus she brought up the sad story of Yajing's death to reveal her sorrow for leaving and represent a separation from her own village as her close relative that played with her during all her childhood died. As it indicated here, Fanghua's decision to work on the reservoir was influenced by a combination of factors.

Through her later experiences, Fanghua's sense of self is reshaped by her migration. She begins to see herself as more capable and independent than before, which is reflected in her comparisons with other women in rural areas who did not migrate. Fanghua recognizes that her migration has allowed her to gain new skills, broaden her horizons, and become more self-reliant. This newfound confidence also leads her to challenge gender norms and expectations. By examining how Fanghua compares herself before and after her internal migration, how she compares herself with women in rural areas who didn't migrate, and how she compares herself with male workers, this paper aims to demonstrate the transformation of self and changing perspectives that arise from migration and contribute to the rise of gender awareness.

Comparing Herself: Before and After Migration

This section presents excerpts from Fanghua's perspective on how she compares her own situation before and after her migration to Inner Mongolia. Prior to her migration, Fanghua had to do many chores in her family's home. She felt that her family did not take her labor seriously and took it for granted:

My family doesn't take me seriously. I just worked with them. I had to feed the pigs, the donkeys, the sheep, the chickens, everything. Other students took a nap for a while, so they did not fall asleep in the afternoon, I worked at noon, and then I dozed off at school in the afternoon.

Being the eldest child in the family but lacking male siblings, Fanghua had to take on the role of supporting the family and performing labor-intensive work, as soon as she finished schooling in the morning, she would have to work for the family during the noon. The tiring process affected Fanghua's education experiences as this work made Fanghua exhausted. She believes that this is because her family does not take her education, or even her life, seriously.

However, her migration to Inner Mongolia brought significant changes to her life. Fanghua felt a sense of empowerment that she had not experienced before. By showing her father's attitude when picking her up at the train station (described in the following excerpt), as well as her discomfort and impressive feelings about how she was treated, she recognized that the work she was doing in Inner Mongolia was valued and appreciated, and this gave her a sense of pride and accomplishment:

My dad picked me up at the train station early. A few decades ago, there were no cars, so my father drove a donkey. He let me sit on the donkey. But I'm afraid of donkeys, so I didn't go on. My father didn't ride the donkey until I stopped sitting on it.

Before having a job, they didn't take Fanghua "seriously"- Fanghua repeatedly emphasized it in her interview, but after that, when Fanghua came back to the village, her father went to the train station very early to welcome her. Considering Fanghua's family was poor and they had very few donkeys that are used for major farm works, under that condition, donkey was the best transportation tool in her family. Fanghua's experience of being welcomed by a donkey and having her father lead her to sit on it, rather than himself, the highest-status man in the patriarchal family, demonstrates the significant change in her family's attitude towards her after she obtained a job in Inner Mongolia, and this transformation in family members' attitudes also reflects the materialized mindset of people during an era that valued economic output. Having a job with a salary was also highly valued, as it allowed individuals to contribute to their family's economic well-being and gain a sense of independence and self-worth.

Due to the social and family norm that valued her job, Fanghua found that her work in Inner Mongolia was highly valued and appreciated, which in turn boosted her own self-esteem and sense of self-worth. When asked which place she is more familiar with, Shanxi or Inner Mongolia, Fanghua's response was paradoxical. She said that she was more familiar with Inner Mongolia than her hometown of Shanxi. This seemingly contradictory response provides valuable insight into Fanghua's evolving identity:

Researcher: Which place do you think you are more familiar with, Baotou (the province in Inner Mongolia) or your hometown?

Fanghua: Baotou, I got used to the place, I am not familiar with my hometown (Shanxi). Because in my hometown, I was too young to understand anything. In final years in Inner Mongolia, the children went to school and I retired. I know everywhere and I don't sound awkward to others. Now I came here (Beijing), I speak but some people do not understand, I felt awkward. I don't speak Mandarin either. But I chose not to say, I go back to our hometown (Shanxi), the village praise me. Unlike some people who go out and speak Mandarin when they come back, the villagers will say that they have forgotten their roots. After that, I don't speak Mandarin. In fact, if I want to speak Mandarin, how can I not learn Mandarin as a teenager (that learn languages so fast) !

Researcher: So now, is your accent more inclined toward an inner Mongolian or Shanxi accent?

Fanghua: More Inner Mongolian. I stayed in Inner Mongolia for a long time, you see, when I was 17 years old, I went to Baotou. I came to Beijing again when I was in my 50s, and now I have stayed here for so long.

This shows that Fanghua developed a personal identification with the place and the work she was doing there, even her dialect is more like that of the Inner Mongolian's. Studies have shown that the preference of dialect is correlated with identity. The use of dialect is not just a product of social and demographic factors but can also be an intentional performance of identity (Reem Bassiouney). So, a use of certain dialect over the other might signal one's inclination of the region with the preferred dialect and the identity there. In the case study, Fanghua regards her dialect more inclined to the one of Inner Mongolia, thus showing Inner Mongolia life is a key part of her life and identity.

Through Fanghua's comparison of her experience before and after obtaining a job, we see how her family's attitudes towards her changed significantly, highlighting how work increased a female's status in a patriarchal family. Prior to her migration to Inner Mongolia, Fanghua's family did not take her labor seriously, and she was expected to perform labor-intensive work without receiving recognition or appreciation. However, her migration to Inner Mongolia and subsequent employment at the factory allowed her to break away from traditional gender roles and expectations.

Comparing Herself with Women Left in the Village

In this section, the analysis on interview materials illustrates how Fanghua compares herself and other women to demonstrate her implicit wish for the valorization of domestic labor. At the same time, this further underscores her preference for her Inner Mongolia identity, as well as her evolving perspective on labor and valorization, indicating an increased gender awareness.

By describing the societal reality and environment in which she lives, Fanghua highlights the limitations and challenges faced by most people, particularly rural women, in her community:

It just seemed like the societal reality and environment: Most people just study till junior high school, know how to read a few words, are able to call on people's names, then begin to work. I'm one of the best people at my age, going out (in big cities) with my cousin to find a job. Otherwise, if I were a rural old lady, I would be dead! Because, as old granny in village, I have no place to see the doctors ! You see, I am now, seeing doctors for those disease(in cities Inner Mongolia).

Fanghua is very grateful for her fate. She compares her own fate with that of rural women's she perceived. She claimed that she would have "died" if not going into the village because she currently has diseases like hypertension and hyperlipidemia and felt really glad that she met some doctors who could give her proper treatment. One conclusion being reached is that her identity confirmation of city workers and preference over Inner Mongolia partly depends on the level of health insurance.

They(other women) all went to the reservoir, when they could (only) eat some white corn, steamed corn flour into a cake, they were not happy. I did go to Baotou(City in Inner Mongolia), you see how good (it is)!

The preference also depends on the food quality, which is a huge contrast with what she eats in Shanxi town. She mentioned that women who stayed in the rural areas could only eat different forms of cereals, but she eats far better than that. Notably, from her description, it seems that she distances herself from the women in the village, as she uses "they" and "I" to refer to herself and the other women respectively.

Still, the comparison does not only limit to a form of feeling being superior, it also includes sympathy for those women and the realization of gender inequality:

Earning a salary is a big deal in the countryside, unlike in the city. My sister who is four years younger than me was very unfortunate. After I went out to find a job, she had to support the family at home. She had to do the housework and assist with the field work. The guys of the family went to the field, she needed to cook at home! She looked after the children and her younger brother. She had to cook before the workers came back. She had no chance to go to school. She was in first grade when she was a teenager, and she mainly worked. Don't look at the few children who died later. They didn't die at birth. They lived for three or four years and died of illness. My sister had to take care of the baby, feed them and hold them.

The above demonstrates that Fanghua realized the unequal and burden young women faced in the countryside. Not earning money like Fanghua, her sister had to finish all kinds of family work ranging from taking care of kids to cooking. But those do not have value attached and respect. In fact, the unpaid domestic labor faced by women suggested by Fanghua is widely discussed by scholars. Unpaid labor refers to women working with household duties without being paid. It is indeed an important aspect of productive activities and an indispensable factor that contributes to the well-being of household and economy.

However, this also means that women are limited in the private domain and are facing unequal treatments since domestic labor is put under the shadow of invisibility and outside the production boundaries (Pushpendra Singh). Domestic labor is often regarded as women's responsibility and not as a contribution since it does not bring direct economic value. Fanghua's family's mindset directly reflects this ideology, as they express no gratitude when she works in the fields, which is considered common in rural areas. However, when Fanghua started working in the factory and sends salaries back, she is respected more than before. This suggests that Fanghua's higher status compared to other women in the village is mainly due to her work and salaries in the factory.

Additionally, Fanghua's description reveals the double burden faced by rural women, which means not only doing housework but also assisting with farm work and participating in agricultural production (Hershatter). The double burden that women experienced was also due to policies during the era. Chinese communist party, inspired by Marxist theory during the first half of the twentieth century recognized the liberation of women as a key component of the transition to socialism, thus they made women work in the public sector (Tsai, K. S.).

Political propaganda further ensured women's willingness to the action. It is claimed that women are as strong as men in order to encourage women to work in the public sector

and contribute to the development of the Chinese economy (Zhou & Guo). Because those sayings give them a sense of equality and respect, women working in the public sector devote themselves unconditionally and work hard regardless of their physical restrictions while feeling proud of their sacrifice (Zhou & Guo). While most or the majority of women in the rural areas regard working in farmland or factories as an honor, Fanghua cast doubt on the extra suffering women had and the extra labor they paid, showing her awareness of gender inequality. Fanghua's increased gender awareness regarding the double burden will be further discussed in the next section.

Comparing Herself with Male Workers

The section explores Fanghua's comparison of male and female workers in terms of salary spending and promotion opportunities. Through her descriptions and lived experiences, an increase in gender awareness is evident, albeit limited due to societal constraints. Many women have internalized their unpaid role in working for their families and accepting a lower status due to adherence to filial piety and societal norms. However, Fanghua's attention to the burdens faced by women serves as evidence of her growing awareness regarding gender inequality in her own life.

Her heightened gender awareness becomes evident when she compares her own situation to that of her husband, leading her to realize the double burden she carries. This symbolizes her new perspective toward labor valorization, extending even to domestic labor. This paper signals her realization as a significant shift in her gender awareness as she begins to reassess the value of her contributions to the family and acknowledges the inequitable distribution of labor between women and men.

For example, she talked about her work experience at Inner Mongolia:

"I was working on a sewing machine in Inner Mongolia, making clothes, knitting sweaters, and making shoes. I did all of them. I don't go to the street to wander around, your grandfather just drew the pattern and I cut it down and put the cloth on the picture to sew."

This excerpt demonstrates her specific feelings about work in the family. Fanghua and her husband had the same job, but her comparison showed that she had to do more intense domestic labor, such as sewing and caring for the family. The word "only" signals Fanghua's feeling of unfairness because she stressed the very little work her husband did in the household. Her awareness of the unequal amount of work and double burden represents her perspective and challenge of the existing social norm of exerting extra pressure on women, which was caused by her migration.

It is after wages mark the exact value of a person's worth that Fanghua rethinks the extra work that is not assigned salaries. If Fanghua had stayed in the countryside and not been exposed to wages as a way of measuring labor, she probably would not have been aware of this inequality. This is because (1) rural social norms regard it as normal for women to take care of their families; (2) rural areas are self-sufficient with the family as the production unit,

and everyone works together and eats the harvest together. There is no objective and comparable labor valuation standard similar to wages, so there is no inter-comparison within a family. When Fanghua entered the city and had a salary, she started to have an objective standard and horizontal comparison with men. Working outside the home and working at home are all hard physical work, but domestic labor was not paid. And compared to her husband, Fanghua did more labor work but only received the same amount of money from the factory compared to her husband, this phenomenon made her aware of the inequality. Fanghua also mentioned that women and men spent their labor income in different ways:

“It seems like the food stamps for men are not enough, but the food stamps for women are enough. Because women are reluctant to spend, and even reluctant to eat when they are hungry.”

This part describes her opinion that women spend money sparingly and save it for family needs, while men spend it for their own sake, often using up the allocated amount. It seems that women had more bonds with the family than men do, and they care and support their extended family in rural areas more than men do (according to Fanghua). However, the way of using money also demonstrated Fanghua’s limited gender awareness due to the societal environment, which will be discussed in the next part.

In Fanghua’s village, it was the norm for women to give back the bride prices to their families after marriage, especially for the use of helping their brothers with giving dowries. However, Fanghua’s leave made it impossible for her to achieve this responsibility, thus, she sent back her salary to “compensate” for not fulfilling the social role. Her obedience to the norm and internalization of responsibility made her sacrifice her own interest for her brother. The way she spent her salary reveals that she still had beliefs and followed the patriarchal mindset of supporting men in the families:

At that time, the fire fee and the installation fee were issued, and I sent money to my home. Indeed, in the end, I saved up money to marry him. In our place, if there is a daughter and a son, The daughter is married off. This is the custom.

Though she achieves her value in labor, she sends nearly all the money back to the family. That is because of the perceived responsibility individuals, especially the biggest kids in the family, had and her observance of filial piety.

I always give money to the family. I’ve never saved any in my life. I’m just tired. I made all my girls’ shoes. I make the soles of shoes, I pull the thread with my teeth, and I bite the thread. That’s pathetic (that I was always working).

However, from the above excerpt, we can see Fanghua’s regret for devoting herself to her family. Despite she did not explicitly mention it, it is clear that she felt empathetic for her exhaustion and that is a form of where she realizes some wrongness for her behavior of sending all her money back to her family. If she had not regretted it, she would not have displayed this pathetic tone, showing dissatisfaction with her work and life.

Her gender awareness has a strong relationship with her job, which reflects the background that Great Leap Forward increased the labor demand in industries. However,

through this excerpt, we see that her awareness needs to be further raised. She still believed that she had the responsibility to give the money back to her family because she was not like other women in rural areas who married themselves to earn bride prices and give those to their brothers to get wives. The limitation of her gender awareness is also due to the social environment, where everyone believes women should care for the family, and support male members. Therefore, women's social status and gender awareness is highly related to social policy and economic development and societal norms.

Fanghua's limitation of gender awareness can also be demonstrated by her perspective on the issue of equal work but different pay. She states that she had the same job as the male workers and earned roughly the same amount of money. However, she also declares that in her factory, workers within the same position can earn different amount of salary if some of them are believed to perform the task better. There are no standers of the "better" but only depending on the employers' subjective views. When being asked whether women or men had higher chances of being perceived as "better", she calmly says it's the men workers and explains using the physical superiority of men as they are stronger and don't have regular absenteeism due to periods.

This article will not debate the effective of "tong gong tong chou (equal work equal pay)" policy, but seeks to analyze Fanghua's attitude towards same work and different salary. It's true that men are, on average, stronger than women on manual labor due to biological difference. However, when Fanghua use this biological difference to explain her job specifically, it's not in the context because her job is chemical testing reagent modulator, which requires not manual labor but the precision of blending the potion that meets the required proportion. Her sayings of explaining the difference of salary are not based on her first- hand experience but likely sayings that emphasize biological differences. The limitation of her gender awareness comes from her internalizing this patriarchal perception and superiority of men without taken into consideration of her working reality- that in non-manual intense jobs, men workers were still more likely to earn more.

On the other hand, her acceptance of the exception of different salary of same job also reflects her internalization of social norms. According to Chenglong Yu, women's acceptance happens due to 1) Allowing women to work in public domain and work in previously man-dominated careers was already satisfying for them. 2) There are men and women in every family, so that overall, there's no so much difference between families- the unit of production in Chinese context. So, for women, they only compare themselves with other women and as long as families' overall difference in their mind are not huge, it's overall a just way to allocate salary. 3) They thought that male preference had always been true and accept the reality(Yu).

For Fanghua, the first reason probably aligns closely with her beliefs since she expressed and admit all the benefits she gained through working in big factories and already felt very satisfied that she was one of the few from her village that came out successfully for work and experienced increased familial status. It's likely that she was already really satisfied

and enjoyed those benefits. And the third reason might partly align with Fanghua as she mentions,

- 1) *The village people long for boys. Because girls cannot go to the fields.*
- 2) *The custom passed down by our ancestors is that it is better to have boys.*

She did realize the corruption of patriarchal mindset as she criticizes the society, but after criticizing, she partly admits the “unavoidable” superiority of man due to fate (the social norm and old customs). It’s likely that she still partly internalized the norms in her village and the old customs as she uses those to help her explain things. It’s understandable that, under a patriarchal society, her mindset was affected.

Again, her explanation of unequal pay also demonstrated how her gender awareness was limited by the society and affected by political propaganda, showing how gender awareness was highly dependent of the societal, economic and political reality.

Further Analysis: Fanghua’s Change and Empathy Toward Women in Rural Areas

This section of the narrative explores Fanghua’s growing empathy for her mother’s experiences after marriage and work. Through her experiences, Fanghua began to rethink her mother’s life in the family and showed sympathy, further displaying her feeling of injustice and increasing gender awareness.

“She had to farm at home. There was a whole family waiting for her to cook and take care of them! ...She had so many children. She would get up very early in the morning and cook. After I go to school, my mother work at home and cook lunch for the family. After lunch, when everyone else had gone to sleep, she would go to the mill with a winnowing basket. She was so busy all day that she had no time to rest. How pitiful! Your mother once said the other day: “I think the most pitiful and hard living one is my grandma - that is my mom, your mom’s grandma! With so many children who died at the age of 3 or 4. You can see, raising so many children, how hard and pitiful of my mom is, she is so thin, only 60 or 70 kilograms.”¹²¹

Fanghua’s family worked as agriculturalists, with both her father and mother earning a living through farming. However, Fanghua notes that the work was not evenly distributed, with her mother having to farm and take care of the family. Fanghua emphasizes that her mother worked exhaustively, from farmwork to domestic labor, and that she was pitiful. While Fanghua does not explicitly mention how she came to feel this way, she expresses that her mother worked tirelessly and that her workload was significantly greater than that of the other family members.

Fanghua exhibits an understanding of the gendered division of labor, acknowledging that women undertake a significantly larger amount of work than men, and recognizing the value of domestic labor. Since in the previous excerpts, she mentioned that she had to work all day without sleep. Her empathy for her mother and emphasis on the amount of work she does demonstrates this awareness. In a patriarchal village where individuals adhere to social norms, such as her father and brothers, may not place importance on Fanghua’s mother’s contributions to the family through domestic labor, viewing it as a given. However, Fanghua

highlights this common occurrence in her village, indicating her recognition of the prevalent inequality.

Also, Fanghua's growing awareness of gender inequality is linked to her own experiences after marriage and work. Her exposure to wages as a way of measuring labor made her realize the unequal distribution of labor between men and women, including domestic labor. As she became aware of her own double burden, she also began to rethink her mother's experiences and recognize the injustice of the situation. Fanghua's empathy for her mother's experiences reflects her increasing gender awareness. It highlights the importance of challenging societal norms that perpetuate gender inequality and recognizing the value of domestic labor.

Conclusion and Discussion

In conclusion, this paper discusses the process of Fanghua increasing gender awareness. The Great Leap Forward led to an increase in job opportunities, resulting in Fanghua's migration to Inner Mongolia. This migration allowed her to gain an elevated status, prompting her to reflect on her past life. By comparing her family members' change in attitudes, she reveals her self-identification as a migrant worker. Furthermore, Fanghua compares herself to women remained in the rural village, strengthening her identity construction as an Inner Mongolian working women and developing a sense of sympathy toward rural women facing double burden. Her growing awareness demonstrates her challenge to existing norm and her advocacy for rural women.

During the process of discussing the unequal distribution of work in rural areas, she connects her own experiences after marriage, where she still faced a double burden. After earning salaries as direct comparison indicators between Fanghua and her husband, her sense of unfairness and the need for domestic work valorization grew stronger.

However, Fanghua's gender awareness was limited by societal norms and propaganda. Still, she adhered to the social role of supporting male family members while sacrificing herself and took the unequal pay for the same work for granted. Thus, this paper concludes that an individual's life trajectory and ideals are greatly shaped by political and economic development, as well as societal environment.

This study elaborates with details of the individual in the case study. The researcher chose her grandmother, because of this close relationship, she obtained qualitative data that are detailed and well-explained. Such rich data could not be gained by someone without the relationship, contributing to the oral history studying community with a clear record. The research examines society through the lens of a single individual, explaining the relationship between gender and social development from the perspective of individual life history.

However, the depth and diversity of interviews in this study are insufficient. Further research could involve interviewing more people connected to Fanghua, allowing for a more comprehensive understanding of her life history from multiple angles, rather than relying solely on her account.

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The Impact of Hay Fever on Public Health in Japan By Sara Tiwari

ABSTRACT

Hay fever, or allergic rhinitis, significantly impacts public health in Tokyo, Japan due to high levels of airborne pollen from cedar and cypress trees. This study examines the perceptions of 169 individuals with hay fever living in Tokyo regarding the health complications and psychological effects of hay fever. Results indicate that most respondents have experienced symptoms for over five years. At least one in five participants reported a negative impact on their daily lives, and nearly one in three experienced reduced productivity. Despite the widespread use of over-the-counter medications, several respondents found them ineffective or problematic due to side effects. The study highlights the need for personalised treatment strategies and improved public health measures. Integrating awareness campaigns and health technology, such as mobile applications, could enhance symptom management and patient outcomes. These findings emphasise the importance of developing targeted interventions and further research to address the broad impact of hay fever in urban settings like Tokyo.

INTRODUCTION

Hay fever, also known as allergic rhinitis, is a common allergic condition characterised by symptoms such as sneezing, runny nose, and itchy eyes, among others that usually affect the eyes, nose, and throat. In Japan, the prevalence of hay fever has been steadily increasing, with seasonal outbreaks that affect a substantial portion of the population—over 40% (Klein and Nippon Research Center). Tokyo, as a densely populated urban province, experiences high levels of airborne pollen from various sources. These sources particularly include cedar and cypress trees, planted in abundance after World War II to reforest the country rapidly and boost timber production, thus making the country, and especially the Kanto region, a prime location for studying the impact of hay fever on public health (Otake).

The importance of understanding the implications of hay fever extends beyond the physical discomfort it causes. There is reason to believe this condition has broader public health implications, including increased healthcare utilisation, decreased productivity, and adverse effects on mental health. Despite the availability of various treatment and prevention strategies in Japan, hay fever remains a significant burden for many individuals, particularly during peak pollen seasons from February to April, which is one-fourth of a year, every year.

Inomata et al. performed a cross-sectional study using a smartphone application called AllerSearch to understand the part that nasal and ocular symptoms play in hay fever. The study found a clear relationship between the severity of hay fever symptoms and reduced quality of life. Symptoms like nasal congestion, facial itchiness, redness of eyes, and tearing were strongly associated with decreased quality of life, with the research demonstrating that there is a wide range of variability in how hay fever affects an individual's quality of life. Patients experienced different combinations of symptoms, and these variations had a

substantial impact on their overall well-being, indicating that hay fever should not be treated as a one-size-fits-all condition; personalised intervention strategies are needed to address these differences. This can lead to more efficient use of healthcare resources and better health outcomes on a national level. The research highlights the importance of multidisciplinary efforts in managing hay fever, as it affects a significant portion of the Japanese population and can lead to substantial medical costs and economic losses. The Inomata et al. study concludes that health apps like AllerSearch can facilitate comparably better clinical care by providing personalised data and customised regimens for patients with hay fever.

Another cross-sectional study, by Muzalyova and Brunner, used a self-reported survey to analyse the choices made by those suffering from hay fever concerning their condition. From the survey, individuals could be grouped into two sets, with one being those who actively seek medical support and the other being those who self-manage symptoms. From their surveyed sample, over 70% of individuals prefer to manage the allergy themselves with only 40% opting for being under medical supervision and 30% utilising immunotherapy. The two aforementioned groups are not mutually exclusive, though, with four in five individuals with hay fever using anti-allergic medicines, regardless of whether or not they self-manage or have consulted a medical professional. A majority of survey participants believed that the drawbacks associated with managing their hay fever were minimal in comparison to the benefits of doing so; however, those with more severe symptoms, as perceived by themselves, were more likely to believe that the effort required to manage their symptoms was greater than if their symptoms were milder. Overall, the perceived severity of one's own symptoms, the perceived seriousness of hay fever as a whole, and the effectiveness of perceived self-efficacy in managing symptoms had great impacts on whether or not an individual takes any kind of action to manage their hay fever.

The paper's concluding note was that despite a significant number of allergic individuals opting for self-reliant allergy management rather than seeking medical supervision, given the widespread use of over-the-counter anti-allergy medication, seeking advice from medical professionals, even at the pharmacy level, is a valuable alternative. Educational interventions to improve allergy management by increasing awareness of risks, especially for individuals with inadequate allergy management, can potentially improve one's quality of life.

Australia, another country in the Asia Pacific region, faces seasonal hay fever at levels comparable to Japan, with allergic rhinitis affecting 17% of the population there (Medek et al.). The Medek et al. study investigated the impact of climate change on human health, specifically focusing on hay fever sufferers in Canberra, Australia. The authors found a significant relationship between the severity of nasal and itch symptoms reported by hay fever sufferers and the levels of pollen in the environment in Canberra. After a certain threshold of pollen concentration was reached, symptom scores remained high, indicating a strong association between pollen load and nasal symptoms. This study suggests, similarly to the Inomata et al. study, that online methods could be beneficial for managing allergic rhinitis and could be adapted to monitor other health conditions affected by environmental variables,

advocating for the potential of online tools to provide patient-specific information and clinical advice. The authors recommend this strategy as being particularly useful in Australia since many hay fever sufferers do not consult healthcare professionals there, which is similar to the circumstances in Japan.

An older study, carried out in Nottingham, England by Richards et al., observed that over-the-counter treatments were used by 54% of sufferers, with drowsiness being a common side effect reported by one-third of the users. More people perceive themselves to be hay fever sufferers than those who have been diagnosed, suggesting that many sufferers self-treat without consulting a doctor, and, thus, it is harder to obtain accurate statistics.

It is evident that conducting novel research in the field of hay fever is of utmost importance, especially research focusing on Japan and the work productivity of the urban population. This research aims to investigate the impact of hay fever on public health in Tokyo, focusing on its prevalence, associated health complications, and psychological implications.

Understanding the full scope of hay fever's impact can prove to be useful when developing public health strategies and policies. By identifying the specific challenges faced by individuals, this study aims to gain deeper insight into how the burden of hay fever can be alleviated and how quality of life can be improved for affected individuals. Furthermore, the findings of this study have the potential to contribute to the global body of knowledge on allergic conditions and their management, providing insights that may be applicable to other regions with similar environmental and demographic characteristics.

METHODOLOGY

Research Design

This study utilised a mixed-methods approach, combining quantitative research techniques with some qualitative features to gain a comprehensive understanding of the impact of hay fever on public health in Tokyo. The primary research involved administering a semi-structured, self-reported online survey, and the responses were analysed to understand how people in Japan with hay fever perceive the disorder.

Data Collection Methods

Survey Design

The primary data collection tool was a semi-structured self-reported survey, designed to capture both quantitative and qualitative data. The survey included multiple-choice questions to gather statistical information on the prevalence of hay fever, the severity of symptoms, the side effects experienced, medication usage, mitigation strategies, and the psychological impact of hay fever. The survey also featured short-answer questions and one optional long-answer open-ended question, all of which to collect qualitative data on individual experiences with hay fever. Thus, this design aimed to comprehensively provide

statistical information while capturing the unique, personal stories of those affected by hay fever, reflecting the varied intensity and nature of the condition.

Survey Distribution

To ensure a broad and diverse sample, the survey was distributed via various social media platforms, informing all viewers of the posts or broadcast messages that this research has a predominant focus on Tokyo and hay fever. The survey was open to anyone, whether or not they had been diagnosed with hay fever, to capture a wide range of experiences and perceptions. The results are to only be used for research purposes, and all participants were made aware of this before they could fill out the survey.

RESULTS

The survey was open between 24 April 2024 and 26 May 2024, and 357 responses were received during this time. A total of 190 individuals (53.2% of total respondents) reported having hay fever; this included those who have either been diagnosed or suspect that they have it. Of the 190 people, 169 currently reside in Japan. This dataset of 169 was used to conduct analysis as this group solely consists of Japanese residents who have hay fever, with the term “have” being used loosely as described above. The demographic details of the individuals who reported having hay fever symptoms can be found in Table 1.

Table 1: Demographics (n=169)

Age

Under 18	8
18-27	33
28-37	73
38-47	47
48-57	8
Over 57	0

Highest level of education completed

Less than high school diploma	4
High school diploma	19
Associate's degree	6
Bachelor's degree	91
Master's degree	38
Professional / Doctoral degree	11

Employment status

Full-time worker	112
Part-time worker	21
Student	28
Not working	8

Category of residential area

Rural	7
Suburban	40
Urban	122

Table 2: Presentation of hay fever (n=169)

Diagnosis of hay fever

Formally diagnosed by a doctor	94 (55.6%)
Suspected, but not diagnosed	75 (44.4%)

Time elapsed since first onset of symptoms

Less than 1 year	22 (13.0%)
1 to 5 years	37 (21.9%)
More than 5 years	110 (65.1%)

Common symptoms by severity (1= low; 5 = high)

	1	2	3	4	5
Sneezing	13 (7.7%)	25 (14.8%)	55 (32.5%)	39 (23.0%)	37 (21.9%)
Runny nose	9 (5.3%)	33 (19.5%)	35 (20.7%)	41 (24.2%)	51 (30.2%)
Itchy eyes	20 (11.8%)	23 (13.6%)	35 (20.7%)	46 (27.2%)	45 (26.6%)
Sore throat	109 (64.5%)	13 (7.7%)	29 (17.1%)	9 (5.3%)	9 (5.3%)

Other symptoms reported by at least 5% of respondents

Itching / Rashes	22 (13%)
Difficulty in breathing	17 (10%)
Cough	15 (8.8%)
Headache	15 (8.8%)
Fatigue	11 (6.5%)

As Table 2 shows, the majority of respondents have been experiencing symptoms of hay fever for over 5 years. Symptoms with the greatest incidence of high severity are runny nose and itchy eyes, with over half the respondents reporting a severity of 4 or 5. Sore throat

does not appear with severe symptoms in this group, with over 70% of the respondents reporting a severity of 1 or 2.

Some other commonly reported symptoms include itchiness or rashes, difficulty in breathing, cough, headache, and fatigue. Some uncommon but concerning symptoms experienced by respondents were nausea, dizziness, and fever.

Table 3: Perceived impact of hay fever symptoms on the quality of life (n=169)

Impact of hay fever symptoms on overall quality of life

Negative	138	(81.6%)
No impact	30	(17.7%)
Positive	1	(0.7%)

Affects ability to perform daily activities

Always	16	(9.5%)
Frequently	58	(34.3%)
Occasionally	60	(35.5%)
Rarely	35	(20.7%)

Effect on productivity at work or school

Reduced	112	(66.3%)
Increased	5	(3.0%)
No impact	52	(30.8%)

Days of work/school missed in a year due to hay fever symptoms

0	129	(76.3%)
1-5	35	(20.7%)
Over 5	5	(3.0%)

Increase in stress or anxiety due to hay fever symptoms

Yes	88	(52.1%)
No	81	(47.9%)

When asked how they perceive the impact of hay fever symptoms on their overall quality of life, the overwhelming majority of respondents (81.6%) reported a negative impact. 66.3% reported lower productivity in school or work, and 43.8% reported that their hay fever symptoms always or frequently interfered with their ability to do their daily activities. In addition, 52% of respondents reported an increase in stress or anxiety due to their hay fever

symptoms. While most respondents did not report missing any days of work due to their symptoms, one person did report losing as many as 25 working days in a year due to the severity of their symptoms.

Table 4: Steps to mitigate hay fever symptoms

Measures to reduce exposure to pollen (n = 169)

	Always	Sometimes	Never
Wearing a mask	37 (21.9%)	90 (53.3%)	42 (24.8%)
Wearing protective glasses	17 (10.1%)	21 (12.4%)	131 (77.1%)
Avoiding outdoor activities	37 (21.9%)	83 (49.1%)	49 (29.0%)
Using air purifiers	13 (7.7%)	7 (4.1%)	149 (88.2%)

Usage of over-the-counter (OTC) medication (n = 169)

Never tried	16	9.5%
Did not have a significant effect	11	6.5%
Could not manage the side effects	8	4.7%
Symptoms reduced slightly	67	39.6%
Symptoms reduced by a lot	67	39.6%

Types of OTC medication tried (n = 153)

Antihistamine tablets	118	77.1%
Nasal sprays	62	40.5%
Eye drops	36	23.5%

Reasons for not trying/discontinuing OTC medication (n (Never tried) = 16; n (Discontinued) = 17)

	Never tried	Discontinued
Worried about side effects	1	3
Symptoms not severe enough	5	2
On prescription medication	3	2
Financial constraints	2	0
OTCs not effective	1	7
Prefer non-medical methods	5	4

Using masks and avoiding outdoor activities during periods of high pollen in the environment were the most common measures used by respondents to minimise their exposure to pollen. Nearly 90% did not use an air purifier to filter out pollen in their homes or vehicles.

Some other methods to minimise pollen exposure that respondents mentioned were frequent washing, especially after returning home (9), using a face or nasal spray that traps pollen (6), drying laundry indoors (5), and removing outer garments like overcoats, before entering the house (5).

Over 90% of respondents report having tried over-the-counter (OTC) medication, however only 39.6% report seeing a substantial reduction in symptoms from them. While 16 respondents reported not using OTCs, from the responses it would appear that an additional 17 respondents have discontinued OTCs. The most commonly reported reason for not trying OTCs was not having sufficiently severe symptoms and preferring non-medical methods of controlling symptoms. The most commonly reported reason for discontinuing OTCs was that they were not effective enough.

Among the OTCs used, antihistamine tablets were the most common (77.1%), followed by nasal sprays (40.5%).

Finally, only seven respondents reported undergoing immunotherapy to manage their hay fever symptoms. Five received sublingual treatments and two received antigen injections.

DISCUSSION

The findings of this study show that hay fever has fairly severe effects, with over half of the respondents reporting severe runny noses and itchy eyes, and small but significant numbers reporting other more impactful symptoms like itchiness, difficulty in breathing, and headaches. Unsurprisingly, hay fever also has a negative impact on people's stress levels, productivity and quality of life. Oddly, five respondents reported increased productivity due to hay fever symptoms; this is likely an error.

The fact that only 94 people (55.6%) had a formal diagnosis seems to be in line with the findings by Muzalyova and Brunner. One surprising finding was that although 94 of the 169 had received a formal diagnosis from a medical professional, as many as 110 respondents reported having hay fever symptoms for over five years. A closer examination of the data revealed that of the 110, only 75 (68.1%) had a formal diagnosis. While this is significantly higher than the overall surveyed population, about one-third of those with long-term symptoms were trying to manage them on their own. This indicates the need to build awareness among the public to seek a diagnosis and find appropriate measures to manage symptoms. Awareness programs targeting general physicians to encourage their patients to seek appropriate treatments may also be useful.

One unfortunate finding is that at least two respondents reported not taking OTC medications due to financial constraints. This observation suggests that the economic burden of hay fever is an overlooked issue. Given the chronic nature of the condition, recurring costs for medications may create financial strain, particularly for those without access to insurance or subsidies. This finding aligns with broader discussions in public health regarding equitable access to healthcare resources, highlighting the need for effective education and improving access to the tools needed to manage hay fever.

A significant finding in the study was the direct relationship between hay fever severity and psychological impacts. A substantial majority (81.6%) reported hay fever having a negative effect on overall quality of life. This aligns with the research conducted by Inomata et al., which noted that symptoms of hay fever exceeding a personal threshold of tolerance were strongly associated with decreased quality of life. The Inomata et al. study also reached the conclusion that there is a wide range of variability in how hay fever affects an individual and their quality of life. This is reinforced by the results of this study as well. Although only 20.7% of respondents claimed that hay fever symptoms rarely affect their ability to perform daily activities, among the other 79.3%, there was great variability in how often their symptoms affect their ability to perform daily activities, with an insubstantial 9.5% claiming that hay fever “always” affects their ability to perform daily activities. Ultimately, this aspect of the study reinforces the hypothesis that hay fever’s impacts extend beyond physical discomfort and have tangible effects on mental health.

One of the most significant findings of this study is that 66.3% of respondents reported reduced productivity due to hay fever symptoms. This result is particularly concerning when considering Japan’s strong work culture, where long working hours and expectations of overtime are extremely common due to this being regarded as the hallmark of a hardworking employee in Japanese culture. In the 2015-2016 fiscal year, over one thousand cases of *karōshi* were registered by the Japanese government (Duarte). This phenomenon of *karōshi* (過労死), translated to “death from overwork” in English, and the normalisation of long hours may mean that workers feel pressured to maintain high levels of productivity, even while suffering from conditions like hay fever. This is further emphasised by the fact that, despite experiencing significant symptoms, 76.3% of respondents do not miss any days of work or school during hay fever season each year. This suggests that many may feel compelled to continue attending work or school, even when unwell, to meet societal and workplace expectations.

Hay fever's debilitating symptoms as well as the side effects of OTC medication being used to minimise the impact of a particular symptom but consequently worsening the effect of others such as fatigue and headaches, as observed by the survey responses, most likely compounds the issue, especially during the spring pollen season. Reduced productivity during regular working hours may lead employees to compensate by working longer hours, thereby creating a vicious cycle where physical well-being is sacrificed to meet job demands. This is supported by the observation that 66.3% of respondents reported lower productivity, yet overtime is a common solution to make up for such shortfalls. In turn, longer work hours may also exacerbate the stress and fatigue caused by hay fever, worsening both mental and physical health outcomes. Moreover, the concept of presenteeism, when employees are physically at work but not fully functioning due to illness, could also be prevalent. This stresses a key point for public health intervention: improving awareness of hay fever's impact on productivity, especially by encouraging businesses to adopt more flexible or accommodating measures during peak hay fever seasons, such as offering remote work

options or allowing flexible work hours. As of the Japanese spring season in 2024, the Japanese government has already begun urging companies to offer remote work during hay fever season, which is a positive step that should be expanded nationwide (Okada).

At the end of the survey, respondents were asked to add any other observations or perceptions they had about their hay fever symptoms. The responses in this section had some noteworthy points for further exploration:

- Many respondents who had moved to Japan from elsewhere reported that their symptoms were mild or non-existent in their home countries. However, three respondents reported the reverse: their symptoms were milder in Japan than in their home countries.
- One person reported fewer symptoms since they adopted a pet dog and were forced to walk outdoors more, thus increasing their exposure to pollen. This is perhaps a case of accidental immunotherapy.
- While most respondents were allergic to pollen from Sugi trees (*Cryptomeria japonica*), some mentioned that they were more allergic to pollen from Sakura (*Prunus serrulata*), or other fruit trees.
- At least two respondents reported that their symptoms became milder after they began consuming probiotic foods regularly, indicating a possible immunomodulatory effect of probiotic bacteria on anti-pollen immunoglobulins.
- Drowsiness was reported as a common side effect among those who took antihistamines.

CONCLUSION

It must be noted that this survey was conducted entirely online and in English. Both of these factors limit its reach and applicability to the wider population of Japan or even the Tokyo region. Additionally, since this was a self-reported survey, the respondents' claims regarding the severity of symptoms or specific mitigation measures cannot be verified. However, the number of people reporting severe symptoms, reduced quality of life, and increased stress all strongly indicate that most people suffering from hay fever perceive significant negative impacts and would benefit from public health measures that help manage and mitigate the disorder. Awareness campaigns could help individuals recognise their symptoms earlier and seek medical treatment, with health technology in the form of mobile applications, a commodity available to a majority of the Japanese urban population. As Inomata et al. showed, smartphone applications have great potential to benefit people suffering from hay fever by providing personalised regimens to mitigate and manage their symptoms. Moreover, public health strategies should focus on expanding access to affordable OTC medications and promoting environmental reforms, such as reducing air pollution in urban areas, which may help alleviate symptoms for many sufferers. The above findings regarding the relationship between hay fever symptoms and productivity highlight the critical

situation of Japan in a broader cultural context. Without proper interventions, hay fever could continue to significantly affect work-life balance and overall public health.

This small-scale study ultimately emphasises the need for further research to understand the broader prevalence and perception of hay fever in Japan and find effective methods to mitigate its effect on the population. Future research should also look deeper into the impact of hay fever on productivity and the potential reasons behind it. Corporations, in particular, need to conduct internal analyses to determine why employees continue to work despite reduced productivity—whether due to societal pressure, fear of job loss, or lack of support systems. There could also be important differences in how people of different age groups, genders, or employment levels respond to these pressures. Understanding these dynamics could lead to more targeted and effective workplace policies and interventions that better support employees suffering from hay fever.

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Fiscal Fiasco: The Economic Crisis of Sri Lanka By Aarush Mishra

Introduction:

It is prudent to understand the importance of fiscal deficit and the measures to limit this. Sri Lanka, an island nation, is passing through the worst phase of economic crisis. Multiple factors are responsible for this. The nation has a long history of taking debts, loans, or financial assistance from international financial institutions. But for years it appeared that things might get normalized once the civil war was over. However, fiscal deficit worsens over a while. Few triggering events led to the present crisis. Was this supposed to happen sooner or later? This study aims to understand the causes that led to this economic crisis and its after effects.

Method:

This study is based on the analysis of available literature, studies and print material. A thorough internet search has been done. Relevant articles and books are read. Opinions of different authors and sources are taken into consideration to understand the factors that put the island nation in this precarious situation.

Results:

1. Before getting into the economic jargon, it is vital to understand a few basic definitions and concepts.
 - (i) **National Budget:** The government forecasts the earnings and expenditures for the year or specific period. The government generates revenue from various sources like taxes. It spends money on defense, infrastructure development, welfare projects etc.
 - (ii) **Budget surplus:** This happens when the government collects more revenue and spends less.
 - (iii) **Fiscal deficit or Budget deficit:** This happens when the government spends more than the revenue generated for the year. Most of the developing countries have fiscal deficits which need to be controlled. This deficit could be because of excessive expenditures, especially in welfare schemes or 'freebies' distribution as a popular measure to win elections. Reduced tax collection or revenues also contribute to fiscal deficit. This is an important concept to provide any economy with a stable status. The government tries to meet the deficits by borrowing from international institutions, central banks or issuing bonds.
 - (iv) **Fiscal debt:** This is the total debt accumulated over a period due to each year's fiscal deficit. It is the money borrowed by the government to balance the fiscal deficits. It is also the total public debt burden.
 - (v) **Gross Domestic Product (GDP):** GDP is the total monetary value of finished goods and services produced by a country in a year. It is an important criterion to assess the performance of any economy.
 - (vi) **Inflation:** It is the rate of increase in the price of a good or service over a given period of time. Higher inflation decreases the purchasing power.
 - (vii) **Currency printing:** Countries cannot print excessive or unlimited amounts of currency. To a layman, it may appear an easy fix solution to curb poverty or deficits. A country cannot print excessive amounts of currency because more of it

will affect the terms of trade and will lead to inflation. For example, in a community, there are one hundred people and each one earns one thousand rupees. The total goods and service requirements of the community is one lac rupees. In this demand and supply (total income of the community) are balanced. In this, if we add one lac rupees more by printing more currency, each individual will get two thousand rupees but the supply remains at one lac rupees. Hence, it will lead to higher inflation and now the same good will be available at higher prices. By this, it is clear that the government prints currency according to the growth in GDP or to replace the damaged currency. In 2008, Zimbabwe printed excessive amounts of currency that led to a very high rate of inflation. This went to the extent that the people stopped taking that currency and started trading in USD.

- (viii) **International Sovereign Bond (ISB):** It is a government-issued debt instrument in foreign or domestic currency. The government sells them to raise money to meet the demands. It offers a precise rate of interest for the specified period and a complete face value on maturation (when the validity of the bond is over). In simple terms, it is like taking a loan from the market. It is considered to be less risky than corporate bonds. However, the interest yield may fall due to a fall in the credibility of the government. Also, if issued in foreign currency, there can be depreciation in the yield and value of the bond if the exchange rate of the domestic currency falls.
- (ix) **International Monetary Fund (IMF):** The International Monetary Fund (IMF) was established in 1944 with 44 founding member countries and currently boasts a membership of 190 nations. Its primary objective is to foster sustainable growth and prosperity among all member countries. This is achieved by endorsing economic strategies that bolster financial stability and monetary cooperation, crucial elements for enhancing productivity, job creation, and overall economic well-being. The IMF's core missions encompass advancing international monetary cooperation, promoting trade expansion and economic growth, and discouraging policies detrimental to prosperity. Collaboration among IMF member countries and other international organizations is pivotal in fulfilling these missions. The IMF evaluates each country's economic landscape and proposes policies conducive to sustainable growth. Moreover, it extends concessional loans and financial aid to member nations grappling with balance of payment challenges. Additionally, the IMF offers support and training to government officials, assisting member countries in fortifying economic institutions, enhancing statistical capabilities, and bolstering expertise in various domains such as taxation, expenditure management, monetary policies, financial system oversight, and legislative frameworks.
- (x) **World Bank:** It was also established in 1944. The main difference between the International Monetary Fund (IMF) and the World Bank lies in their respective purposes and functions. The IMF oversees the stability of the world's monetary system, while the World Bank's goal is to reduce poverty by offering assistance to middle-income and low-income countries. The World Bank is an international financial institution that extends loans and financial assistance to governments. Comprising the International Bank for Reconstruction and Development (IBRD) and International Development Association (IDA), it is one of five organizations

under the World Bank Group umbrella. Over the past three decades, it has broadened its loan portfolio to include NGOs and environmental groups, guided by the United Nations' Sustainable Development Goals and environmental and social safeguards. The bank directs its loans toward developing nations to combat poverty and actively participates in global partnerships and initiatives, including efforts to address climate change. Offering various training programs, it collaborates with entities like the Clean Air Initiative and the UN Development Business.

The key distinction between the International Monetary Fund (IMF) and the World Bank lies in their respective aims and roles. The IMF focuses on maintaining global monetary system stability, while the World Bank's primary objective is poverty reduction through assistance to middle- and low-income countries.

(xi) **Multilateral foreign borrowing:** Multilateral foreign borrowing refers to a credit or term loan offered to a borrower by multiple lenders under uniform terms and conditions using standardized documentation. This type of financing is sought by large nonbank corporations, financial institutions, and government entities when the required funding exceeds the capacity of any single relationship bank. Multilateral lending, also known as syndicated lending, aims to distribute credit and distribution risk among participating lenders.

(xii) **Bilateral foreign borrowing:** Bilateral foreign borrowing involves credit or term loans provided by a single lender to a borrower, as opposed to a multilateral facility. Bilateral financing is commonly used in most bank lending scenarios and is suitable for funding requirements that can be met by a single lender. Bilateral loans are generally smaller, less complex, and more cost-effective for the borrower. Large bilateral loans are often funded through loan participation arrangements.

In international relations, states may opt for multilateral channels over bilateral ones to obscure potentially contentious arrangements, particularly in areas such as trade, finance, development, or security policy. Empirical evidence suggests that multilateral aid channels are less influenced by political considerations, more responsive to recipient country needs, and better equipped to support global public goods. Recipient countries often prefer multilateral aid due to its selective targeting of poorer nations and its ability to provide preferred options for development assistance.

2. **Beginning of the Crisis:** Sri Lanka gained independence in 1948 but the island nation was under civil unrest for a long time. It had witnessed a civil war from 1983 to 2009. It has the population of 2.2 million. The full-blown crisis was seen in 2022 but the seeds were sown much earlier as, discussed under.

(i) **Fiscal Deficits:** The country has faced financial problems many times. Sixteen times it has taken financial aid from the International Monetary Fund. The country had presented seventy-three budgets with fiscal deficits between 1950-2022. This clearly explains the financial hardships or constraints faced by the country. During the civil war, the deficit was 8.4% of GDP. From 2010, the country showed signs of revival of economy and the average deficit was

contained to 5.7%. 2018 proved to be the best year with the deficit coming down to 5%. As an example, sources of revenue and expenditure for the 2018 budget are shown in the tables:

Revenue and grants	Value in LKR(in Bn)	Percentage
Indirect Tax	1659	74
Direct Tax	375	17
Non Tax	184	8
Grants	10	0.4
Total	2282	

Expenditure	Value in LK R(in Bn)	Percentage
Interest payments	820	28
Capital Expenditure (net lending)	751	26
Salary & services	630	24
Subsidies & Transfers	507	17
Goods and services	195	7
Total	2903	

The budget deficit was Rs 675 Billion, about 5% of GDP (total GDP was 14200 Bn). In the following years, 2019-2022, due to multiple reasons, which are discussed later, there was a sharp increase in the fiscal deficit. There was a shrinkage of GDP along with a rising deficit which reached 11.7% of the GDP in 2022.

- (ii) **Measures taken by the government to manage the deficit:** The government started financing the budget deficits by borrowing from issuing International Sovereign Bonds in 2007. Before 2007 the foreign debt was USD 2.15Bn. The government issued these bonds regularly to meet the deficits. Over the decade, in 2019, this foreign borrowing reached a staggering USD 17.6 Bn. This international borrowing amounted to 42% of the total in contrast to just 17% in 2007. In 2022, total debt was USD 88 Bn, out of which USD 33 Bn was foreign debt.
 - (iii) **Public Debt:** It is clear from the above discussion that over the last decade public debt gradually increased to the level of 100 % in the year 2021. Shrinkage of GDP and rising debts make the debt unsustainable, finally due to this the country defaulted to pay the debt in 2022.
 - (iv) **Depleting Forex:** In 2022 the country had just USD 2 Bn as foreign reserve, which was hardly sufficient for 6 weeks imports.
 - (v) **Depreciation of LKR:** Due to excessive printing of money, drying foreign reserves, international debt and other reasons LKR depreciated from 167 to 370 for one dollar.
3. Sri Lanka got into this unprecedented crisis due to multiple factors, some of which are discussed as under:
- (i) **Loss of Tourism:** Tourism was an important source of revenue for the country. In 2018, it contributed to 13% of GDP. It was also an important

source of foreign currency. In 2018, 2.3 million tourists visited the country. In 2019, there were multiple bombings in the churches on Easter leading to panic in the tourists. It was a big blow for this industry because Lanka was no longer a safe destination. This led to a drastic drop in the number of tourists which fell to 0.17 million (93% fall) in 2019. The following year was the year of the COVID-19 pandemic. This was associated with various curbs including international travel. These two factors led to a huge loss of revenue and employment in the country.

- (ii) **Tax Cuts:** From 2015 to 2018, there was an attempt to control the deficits. The government was working closely with the IMF recommendations. The union budget was focused on increasing revenues and cutting down expenditures. But then, there was an election in 2018. Then, the Opposition party promised a lot of tax cuts, freebies and subsidies without considering the financial implications of these measures to the fiscal deficits. This was going to put an extra burden on the already strained economy of the country. The new government swept to the office with lucrative poll promises. The government announced the tax cuts in November 2018. Few of the tax cuts were applicable immediately, others were from 01 January 2019 or 2020. It not only led to decreased revenue but also depleted the base of tax providers. The following taxes were cut or abolished :

Tax	Revised taxes	Remarks
Income Tax	Upto 3 million- 6% 3-6 million- 12% More than 6 million- 18%	Earlier the rate was Upto 6,00,000- 4% Next 6,00,000- 8% Next 6,00,000- 12% Next 6,00,000- 16% Next 6,00,000- 20% Balance after- 24%
Value Added Tax(VAT)	8	Historically this was the biggest contributor of revenue. The tax was reduced from 15% to 8%.
VAT on Condominium property	Abolished	
Pay As You Earn(PAYE) tax	Abolished	Every employee who earned a monthly remuneration over Rs. 100,000 was liable to PAYE. It was deducted at the source.
Nation building tax (NBT)	Abolished	2% tax (on turnover) intended to finance infrastructure construction after the end of the civil war in 2009. It was chargeable on imports,

		manufacturers, and service providers.
Capital gain tax on the share market	Abolished	
Corporate tax	24%	Reduced from 28% to 24%.
Economic service charge	Abolished	It served as an early payment of income tax
Withholding tax (WTH)	Abolished	This was a tax on dividends, interest, discounts, charges, natural resource payments, rents, royalties, premiums and retirement payments.
Debit tax on banks	Abolished	
Income tax on the construction industry	14%	Reduced by 50%
The tax-free threshold for turnover for VAT raised to	300 million Rs per annum	Earlier the threshold was 12 million Rs per annum
All Taxes on Places of worship	Exempted	
Debt Repayment Levy	Abolished	The levy was charged on financial institutions at 7% of Value Addition from Financial Services.

There was a sudden decrease in the taxpayer base. Approximately one million taxpayers were out of the base due to tax cuts. The tax revenue was drastically reduced from LKR 1025 Bn, in 2019 to LKR 523 Bn in 2020. Other factors are also responsible, like the Covid pandemic, for this 48% sharp fall in tax revenue.

Tax revenue collection is shown in the table

Year	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021	2022
Revenue in million USD	7654	7131	7794	8046	9978	10043	10958	10536	9715	6565	6562	5458

It is evident from the table that 2017 was the best year in respect of tax collection. 2019 onwards there is a sharp decline in the revenue collection.

iii. To cover the government spending and falling revenue, the Central Bank started printing local currency at a record level. The government was warned by the IMF against printing the currency at an unprecedented rate which would lead to an economic implosion.

iv. **External debts:** Earlier Srilanka had multilateral debts which over a period changed to bilateral. In 2005 Srilanka had a loan of USD 11 Bn which rose to USD 54 Bn in 2020, which was more than 100% of its GDP.

v. **Non-profitable Mega infrastructure projects:** Srilanka spent a massive amount of money on many infra projects. It took loans from China to build the Hambantota

International Port and the Mattala Rajapaksa International Airport. These projects turned out to be white elephants. Srilanka was not able to pay the loan amount for the Hambantota International Port and it is said that due to this it has to grant a lease of 99 years to China. Recently it has handed over the maintenance of Mattala Rajapaksa International Airport to an Indian and Russian company for the next 40 years.

vi. **Organic Farming:** The government suddenly banned chemical fertilizers and switched completely to organic fertilizers. The country was self-reliant in the production of rice and other crops. However, this sudden shift led to a decline in the production of rice. This method was more expensive and the yield was less. This led to an overall decrease in agriculture production forcing the island nation to import even rice also.

vii. **Russian invasion of Ukraine:** Russia was a good market for Srilanka. It was exporting tea and other spices to Russia. Russian- Ukraine war led to a decline in exports to these countries. These two countries were also big contributors to the tourism industry. This war caused a marked decrease in number of tourists from these countries.

viii. **Black marketing of USD:** The exchange value of Srilankan rupee started falling. In 2020 average exchange rate was 1 USD 185 Rs. which fell flat to 1 USD = 367 Rs in 2022. Despite all the pressure on the local currency the government tried to tightly control the exchange rate at about Rs 200 in Feb 2022. The credibility of LKR was falling hence the rate offered outside the banks was much better. Srilankans who were working outside the country started using other illegal means to send money at a higher exchange rate. This led to a fall in Foreign remittance to banks and black marketing of USD.

ix. **Losses from State-Owned Enterprises :** State-owned Enterprises especially Sri Lankan Airlines, Ceylon Electricity Board, Sri Lankan Transport Board, Ceylon Petroleum Corporation, and Hotel Developers Company Limited etc were putting a big hole in the revenue. These were among the 52 SOEs that were booking losses for a long time leading to extra pressure on the stressed economy of the country. In 2021 alone there was a loss of Rs 82 Bn from these SOEs. The cumulative figure is much higher.

x. **Depleting foreign reserves:** Srilanka was able to maintain USD 6-8 Bn foreign reserves from 2010 to 2021. As discussed earlier every year there was a constant fiscal deficit which was managed by borrowing through ISBs. However, after the Easter bombings, there was a negative impact on foreign investments and remittances which was worsened by Covid in 2020. Tourism and foreign remittances were also badly affected. By Mar 2022, reserves depleted to a critical level of USD 1.9 Bn. It depleted further by April 2022 and declared default on foreign debt repayments.

4. Effects of Economic Crisis: The crisis that started in 2019 and still going on has witnessed a very high inflation rate, a shortage of food, fuel, medicines and other essential commodities. The condition of the local population was too bad. There was not only a shortage but a lack of purchasing power to afford it. It led to civil unrest and the President fled the country and ministers had to resign. A new caretaker government was formed to address the worsening situation of the common people. Crippling shortages and long painful powercuts are also noted in a survey conducted by United Nations in 2022 and 2023 found that 55 % population of Sri Lanka is vulnerable due to the crisis and one-third of the population gets debt to meet essential needs like food, medicines etc. The inflation rate was 49.7 % in 2022 which was about 7% in the previous year. Following are the aftereffects seen following the economic crisis in March 2022:

(i) **Political turmoil:** In 2021, the government accepted that Sri Lanka is standing on the verge of the worst economic crisis. By the next year, there was a full-blown economic crisis. Foreign reserves reached an all-time low, there was civil unrest due to an acute shortage of essential commodities. It led to demonstrations and protests against the government. 41 lawmakers withdrew support hence the government lost the majority. The Prime Minister, Mahinda Rajapaksa had to resign in May 2022. Ranil Wickremesinghe was appointed the new prime minister of the interim government. The President, Gotabaya Rajapaksa, resigned in July 2022 due to increasing protests throughout the country. The Governor of the Central Bank of Sri Lanka was replaced in April 2022. In a nutshell, there was political instability and uncertainty.

(ii) **Civil unrest:** In March 2022 public protests started. These were both from opposition parties and non-political organizations. The general public was distressed because of rising prices and shortage of essential commodities. In March and April 2022, protests were very frequent. Curfews were imposed to control the protests. In May 2022 a peaceful protest turned violent and the protesters set on fire the Rajapaksa family house. On 09th July 2022, the protesters entered the President's Mansion. They created chaos inside the premises and even entered the swimming pool of the President's House. The President had fled the Mansion before the entry of the protesters.

(iii) **Food crisis:** In August 2021 the government declared a food crisis. There was a shortage of main food items like rice, vegetables etc. The prices were soaring high. There was no import of food items due to the limited availability of foreign reserves. In February 2022, local potatoes and rice prices increased by 74% and 64% respectively than one year ago.

(iv) **Fuel shortage:** There were only two oil companies in Sri Lanka. Ceylon Petroleum and IOC Sri Lanka. Sri Lanka is largely dependent on Oil imports. When foreign reserves started drying up, it had to decrease the oil imports to a great extent. It led to an acute shortage of fuel. There were long queues at filling stations. To control the mob help of the Sri Lankan Army was taken. They were deployed at the refilling stations to maintain law and order.

(v) **Electricity crisis:** The economic crisis led to a decline in the electricity supply. There was a severe shortage of electricity. Measures were imposed to curb the electricity consumption. Streetlights were switched off. Power cuts were imposed. In March 2022 it increased from 7 hours a day to 15 hours a day in April 2022.

(vi) **Cooking gas crisis:** There was a very scarce supply of cooking gas. There were long queues outside these gas depots and agencies. There was a long waiting list for getting gas cylinders. According to a report nearly 1000 bakeries closed down due to a shortage of cooking gas.

(vii) **Medicine and hospital crisis:** Medicines were in short supply. Even the essential medicines were not available. Routine and to some extent, emergency surgeries were getting cancelled. Life-saving stents and endotracheal tubes were not available easily. The hospitals and the whole medical system were collapsing.

(viii) **Unemployment:** Tourism contributed to a major source of employment but when the whole system was collapsing the tourism industry was worst affected. There was a very high rate of unemployment.

(ix) **Tourism:** As discussed earlier there was a sharp decline in the number of tourists due to the Easter Bombings, Covid, Russia- Ukraine war and civil unrest. Ultimately it led to a decrease in foreign remittances and revenues.

(x) **Education:** The government did not have money to import paper, They had to cancel the examinations. Many schools were shut due to the non-availability of school transport, books and papers.

(xi) **Entertainment:** The Entertainment industry completely collapsed. There was no money to pay for the foreign channels and broadcasters. Publication of daily newspapers was stopped due to paper shortage.

(xii) **Sports:** The government had no money to send the players to competition. They heavily cut down on the expenditures on sports.

(xiii) **Closure of embassies/ foreign missions:** Many embassies and missions were closed temporarily due to the financial crisis.

(xiv) **Foreign investments and orders:** In this situation, foreign investments dried up. The orders of foreign companies shifted to other countries. The demand orders for Sri Lankan tea were replaced with Indian tea.

(xv) **Inflation:** An inflation surge was noted in 2021. In Feb 2022 this reached the level of 17.5%. The year-on-year inflation for food items was about 24%. The prices of food items, cooking gas and fuel were skyrocketing.

(xvi) **Transport and international travel:** Due to the scarcity of fuel transportation was gravely affected. On 28th June 2022, the government stopped the sale of fuel to personal vehicles. Only public transport, trains, ambulances and emergency vehicles were allowed refueling. Aviation fuel was also in short supply which affected air travel. International flights were advised to come with extra fuel. Indian airports were used for refueling.

5. Remedial Measures: The country responded in many ways to come out of the crisis. They are as under:

- (i) **IMF Bailout:** IMF officials warned the Sri Lankan government about the unsustainability of debts and to refrain from printing more money in February 2022. The following measures were taken to get the IMF bailout.
 - a. Devaluation of Sri Lankan rupees: To attract more foreign revenues local currency was devaluated by the Central Bank of Sri Lanka.
 - b. A core committee was formed to guide the President about the policies and to discuss with IMF for a bailout.
 - c. The President expressed his desire to work with IMF.
 - d. In April 2022, IMF insisted that any loan to Sri Lanka should be sustainable. IMF considered Rapid Financing Instrument (RFI) loans for Sri Lanka.
 - e. Sri Lankan discussions with the creditors were lauded and IMF vowed to help Sri Lanka.
 - f. IMF stressed on credible and coherent strategy to restore macroeconomic stability.
 - g. IMF also suggested restructuring of the loans.
 - h. IMF approved USD 3Bn as an extended fund facility for Sri Lanka.
 - i. Other agencies like the World Bank and the Asian Development Bank came out to help the country.
- (ii) **Interim Government:** A unity government was formed and Ranil Wickremesinghe was appointed the new prime minister of the interim government. The government accepted that 8% of VAT is not sustainable. Taxes were revised and VAT was raised to 15%.

(iii) International help:

- a. India's Neighborhood First Policy: India stood with Sri Lanka in the crisis. India pledged to give USD 2.5 Bn aid to Sri Lanka. India granted a new line of credit worth USD 500 Mn for the purchase of petroleum products and USD 1Bn for food and medicines. India also sent 40,00 tonnes of rice and 270,000 tonnes of fuel to Sri Lanka.
- b. Other Countries like Singapore (raised USD 1,00,00 for the Red Cross Society of Sri Lanka) and China (USD 74 mn) also helped Sri Lanka.

(iv) Policy changes:

- a. Fiscal Policy and improvement of Tax collection: The government raised the taxes and realized that reducing the VAT to 8 % was a mistake.
- b. CBSI tried to contend the inflation by raising both the Standing Lending Facility Rate and Standing Deposit Facility Rate by 700 basis points.
- c. In June 2022, the government allowed oil-producing countries to sell oil in Sri Lanka to mitigate the energy and fuel crisis.

6. Impact of the remedial measures:

Realising the blunders committed and the will to overcome the crisis shall help the country draft policies to come out of this disaster in the long run. By November 2022, economic conditions improved. The inflation rate started coming down. Tourists began visiting the country. Supply of fuel, food and cooking gas improved. There are no long queues. Supply of essential medicines restored. Routine and emergency surgeries are being performed without many problems. There are no long power cuts. The government is working towards increasing the tax base and revising the tax slabs to increase the revenue. Most importantly, there are no protests and the general public is now aware of stringent measures to bring the country on the right track. They want the government to make the decisions that are best suited for the country. In February 2024, the country has projected a 1.8% growth rate. Middle and small manufacturing units are being promoted. Reforms are being implemented and the country is expecting recovery from the crisis in time ahead.

7. Conclusion:

The economic Crisis of Sri Lanka is due to a financial fiasco compounded by the Easter Bombings, Covid and Tax cuts. Popular measures and welfare schemes should be weighed against the sustainability of these efforts and the impact they would have on the economy of the country. Taxes are an important source of revenue that helps in the smooth functioning and growth of the country. Sri Lanka has gone through a lot in the last 5 years but now is on the path of recovery.

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CVCP-Fusion: On Implicit Depth Estimation for 3D Bounding Box Prediction

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Abstract

Combining LiDAR and Camera-view data has become a common approach for 3D Object Detection. However, previous approaches combine the two input streams at a point-level, throwing away semantic information derived from camera features. In this paper we propose Cross-View Center Point-Fusion, a state-of-the-art model to perform 3D object detection by combining camera and LiDAR-derived features in the BEV space to preserve semantic density from the camera stream while incorporating spacial data from the LiDAR stream. Our architecture utilizes aspects from previously established algorithms—Cross-View Transformers and CenterPoint—and runs their backbones in parallel, allowing efficient computation for real-time processing and application. In this paper we find that while an implicitly calculated depth-estimate may be sufficiently accurate in a 2D map-view representation, explicitly calculated geometric and spacial information is needed for precise bounding box prediction in the 3D world-view space. Code to reproduce our results is available at <https://github.com/safetylab24/FusionCVCP>.

1 Introduction

The 3D object-detection task has become commonplace when it comes to autonomous driving. As two fundamental modalities, LiDAR and camera sensors are capable of acquiring surroundings in different manners. LiDAR captures precise geometric and spatial information by providing an accurate 3D point cloud, while cameras contribute rich semantic details such as texture and color. However, previous approaches to multi-sensor fusion, partic-

ularly point-level fusion methods, fail to fully utilize the complementary strengths of these two modalities. Point-level fusion often leads to a loss of semantic information derived from camera data, resulting in sub-optimal performance in tasks that require precise 3D bounding box prediction and accurate object localization. Additionally, feature-level fusion may introduce geometric distortions when mapping LiDAR data onto a 2D image plane, which further limits robust detection, especially in tasks demanding high geometric accuracy.

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In this paper, we propose Cross-View Center Point-Fusion (CVCP-Fusion) [figure 1], a state-of-the-art model designed to address these limitations by combining camera-derived semantic density with LiDAR’s strong spatial data in the world-view (3D) representation. Unlike traditional anchor-based methods that struggle with rotational variance and require axis-aligned bounding boxes, our center-based approach simplifies detection by predicting object centers directly, making the system rotationally invariant and significantly reducing computational complexity.

Building on the established Cross-View Transformers(19) and CenterPoint(18), our architecture runs their backbones in parallel, enabling real-time performance. The CenterPoint backbone is optimized for detecting object centers using LiDAR data, which are then refined in the second stage using key-point detection to predict object properties such as size, orientation, and velocity. In contrast, the Cross-View Transformer enables the model to fuse multi-view camera features effectively into the BEV space, ensuring that the high-resolution semantic information is retained across modalities.

Multi-Sensor Fusion. One of the primary challenges in combining LiDAR and camera data is the discrepancy between perspective views from cameras and 3D views from LiDAR. Existing methods have typically focused on projecting LiDAR data into the camera view, introducing distortions that affect performance in geometric tasks such as 3D object detection and tracking. Our model addresses this by working in the shared BEV space, eliminating the need for perspective transformations that lead to information loss. By fusing multi-modal features within this unified space, CVCP-Fusion achieves higher detection accuracy while reducing computational overhead.

To efficiently utilize both input streams, the CVCP-Fusion architecture passes inputs through modal-specific encoders and combines extracted features in the 3D world-view space. CVCP-Fusion functions by extracting image features through an EfficientNet-B4 architecture and extracting LiDAR features through the Point Pillars Network(8) 3D Encoder. This

LiDAR encoding is simultaneously decoded into the 3D world-view space and up-sampled using an RPN, which is finally concatenated to the decoded image embeddings outputted from the cross-view transformer.

2 Related Works

Depth Estimation. Many current implementations (1; 9; 13; 14) utilize epipolar geometry and triangulation to explicitly compute camera extrinsics and depth. Despite being convenient, explicit depth calculations use multiple noisy inputs and require highly calibrated camera extrinsics data, often leading to inaccurate outputs for downstream calculations.

Monocular depth estimation aims to find depth information, rotation, and orientation to create a 3D representation of a view to allow for easier detection of features. Previously it has been shown that 2D estimations are significantly more robust than direct 3D estimates. Therefore, most algorithms (18), (10) first find features in 2D and regress to 3D using depth-estimation algorithms. Center Point (18) learns depth information directly while other algorithms such as ROI-10D (10) project to 3D to infer height.

While monocular object detection does not need to consider inputs from multiple sources, it does require accurate depth estimation. Depth information is often difficult to predict and requires a probabilistic approach. The Cross-View Transformers architecture addresses this by passing positional embeddings along with extracted features into transformer blocks, baking geometric reasoning into the weights of the transformer. This allows the transformer to convert a 2D multi-camera view input stream into a 3D world-view output while also incorporating information across the 6 different views.

Map-View Segmentation

One common approach to Map-View Segmentation is using a Geometric Transformation on a planar image to convert it to a BEV (2), (7), (5). However, more recent segmentation algorithms use a cross-view transformer architecture (11), (19), (12). This architecture achieves high accuracy with low computational

costs making its implementation more feasible in real time. One such algorithm is VPN (11), which passes depth information and RGB data into a view-transformer module to create a semantic map of the scene. One drawback to this approach is that it doesn't consider camera calibration matrices, and relies on the model to learn them. Thus, we incorporate CVT (19), a state of the art algorithm that uses cross-view-attention to create a BEV using implicit geometric reasoning. Its primary advantage is that it uses positional embeddings to reduce inference time compared to explicit geometric reasoning.

3D BEV Object Detection

3D Object Detection aims to create a BEV map-view segmentation of a scene which, in combination with height data, allows for 3D Object Detection. State of the art algorithms such as BEVDet (6) use an image encoder-decoder architecture with a 3D object detection head to create bounding boxes. Similarly, M²BEV (16) uses an image encoder to learn image features and then projects to the BEV space, allowing for joint 3D object detection.

This research paper aims to achieve such results by combining two state-of-the-art algorithms, particularly CVT and CenterPoint, to create a 3D BEV Map-View segmentation and perform 3D object detection utilizing the embedded height data.

3 Model Architecture

3.1 Cross-view Transformers

Cross-view transformers aim to efficiently generate a map-view semantic representation of a vehicle's surroundings using an attention-based model. The architecture implicitly learns a mapping from individual camera views into a canonical map-view representation using a camera-aware cross-view attention mechanism. Each camera uses positional embeddings that depend on its intrinsic and extrinsic calibration. These embeddings allow a transformer to learn the mapping across different views without ever explicitly modeling it geometrically. The architecture consists of a convolutional image encoder for each view and cross-view transformer layers to infer a map-view semantic segmentation.

The height of any voxel can be approximated using the 2D birds-eye-view (BEV) label. Therefore, while other architectures focus on learning the geometric depth of any pixel in the 3D voxel-space relative to the position of the car, CVT learns depth as a proxy to the known height of the voxel. The cross-view transformer allows the network to learn any geometric transformation implicitly and directly from data. It learns an implicit estimate of depth through the camera-dependent map-view positional embedding by performing the downstream task as accurately as possible.

3.2 Centerpoint

CenterPoint is a state-of-the-art framework that predicts center-based 3D Object Detection and Tracking. It creates 3D bounding boxes by using voxel-space coordinates to represent, detect, and track 3D objects over a continuous-temporal state space. This allows the model to determine positional data along with movement data such as velocity. Centerpoint argues the main issue in accurate 3D bounding box detection lies in the inherent information loss when unprojecting from a 2D input to a 3D voxel space. To address this, for each detected center, Centerpoint regresses to all other object properties such as 3D size, orientation, and velocity from a point-feature at the center location. Furthermore, it uses a second stage to refine the object locations and extract point-features at the 3D centers of each face of the estimated objects 3D bounding box. This allows the model to recover the lost local geometric information due to striding and a limited receptive field, and brings a decent performance boost with minor cost.

3.3 Cross-view Centerpoint

CVT was trained on the nuScenes camera-only data, and it achieved state-of-the-art results on speed and accuracy. Since CVT learns a proxy for depth, we can directly feed its output, a BEV with height data embedded, into Center-Point as a proxy for the learned 3D embeddings. The two-stage CenterPoint block then computes the final 3D bounding box and its velocity. We use the VoxelNet network because prior results show that it outperforms the PointPillars 3D detection encoder.

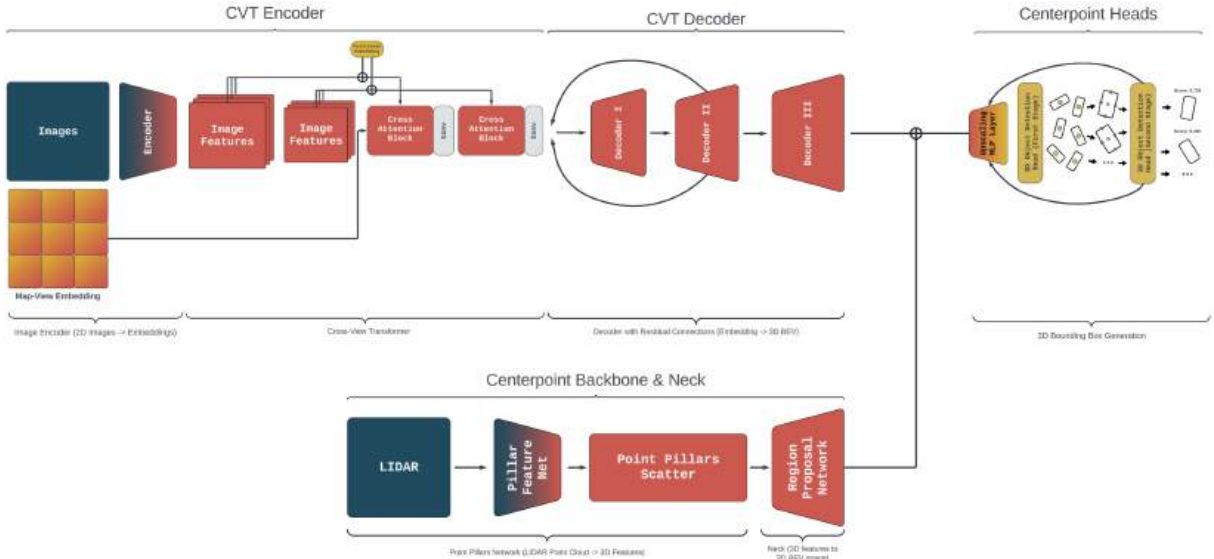


Figure 1: An overview of our proposed model architecture. For each image, we extract image features across multiple scales. Using known camera pose and intrinsics, we construct a camera-aware positional embedding. We learn a map-view positional embedding that aggregates information from all views through a series of cross attention layers. This is passed through a decoder which converts the learned embeddings into the 3D BEV space. In parallel, corresponding LiDAR data is passed through a Point-Pillars network and up-scaled using an MLP to the 3D BEV space. This is concatenated with the camera-derived BEV and convolved. Then, a 3D CNN-architecture detection-head finds object centers and regresses to full 3D bounding boxes using center features. This box prediction is used to extract point features at the 3D centers of each face of the estimated 3D bounding box, which are passed into an MLP to predict an IoU-guided confidence score and box regression refinement, allowing rotation of predicted 3D bounding boxes.

4 Results

We first present our 3D bounding box prediction results on the NuScenes test dataset for both the CVCP and LiDAR-Fusion models.

Dataset The nuScenes(3) dataset contains 1000 diverse scenes ranging over a wide variety of times in the day, weather environments, and traffic conditions. Each scene contains 40 frames and lasts 20 seconds, for a total of 40,0000 data points in the dataset. Data encompasses the full 360° scene around the ego-vehicle captured through 6 camera views. Each camera view has calibrated intrinsics K and extrinsics (R, t) at every time-step.

We find that CVCP-Fusion doesn’t perform well (mAP of 48.71 4) compared to other modern implementations of 3D bounding box predictors. One major difference between these models is that they forego the error prone implicit depth calculations of CVT and instead use explicit depth calculations.

Method	mAP
PointPillars (8)	68.4
CVCNet (4)	82.7
CBGS (20)	81.1
PointPainting (15)	77.9
PMPNet (17)	79.7
SSN (21)	80.7
CVCP	>10.0
CVCP-Fusion	48.71

Table 1: State-of-the-art comparisons for 3D detection on nuScenes test set. We show the mAP and mAP for the car class.

Implementation Details We tested two separate training methods when training the CVCP-Fusion model, one with a frozen CVT block and one training the entire model. For

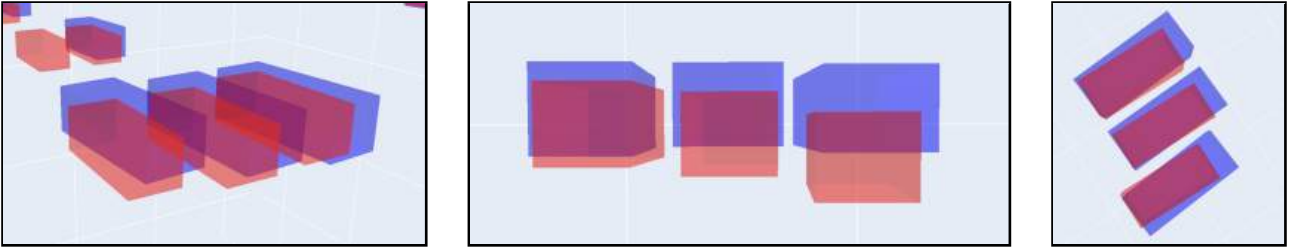


Figure 2: Perspective, front, and top-down views, respectively. These three cars and their corresponding predictions sampled from the nuScenes dataset (*ground truth in blue and predictions in red*) illustrate an example of CVCP-Fusion performing well at predicting rotation and bounding box coordinates in the x and y-dimensions, but failing to accurately predict in the z-dimension.

the former, we used two data-loaders to simultaneously pass in LiDAR and camera-view data. For the latter, we used two separate training sessions. The former to calculate decoder outputs from the CVT block [see top path in figure 1], and the latter to calculate RPN outputs from the Centerpoint block [see bottom path in figure 1], concatenate with the stored CVT tensors, and pass through the detection head. We saw similar performance in both training techniques and presented the latter approach’s results as it provided a slightly higher mAP.

5 Analysis

5.1 Findings during Testing

We find that bounding box predictions are inaccurate but consistently poor in a specific aspect: height. Going through predictions we found that CVCP-Fusion consistently accurately placed bounding boxes in the x and y directions but not on the z-axis. These three cars [figure 2] and their corresponding predictions sampled from the nuScenes dataset (*ground truth in blue and predictions in red*) illustrate an example of this situation. While other factors such as false positives and imprecise rotation predictions also somewhat contributed to our fusion model’s poor accuracy, height was consistently an issue the model struggled with the most. We found many more examples of this in almost every scene, with predictions being accurate in nearly every aspect except alignment in the z-axis.

We used a the ADAM optimizer, a base momentum of 0.85, a max momentum of 0.95, a learning rate of 0.001, and a batch size of 4 following Centerpoint’s implementation details for their most successful session. The model took about about 24 GPU-hours when training on 3 GPUs for 15 epochs. After testing many possible values, we finally used a confidence threshold of 0.35 when evaluating the model to achieve our best results. Our hyperparameter settings can also be found in our config presets on <https://github.com/safetylab24/CenterPoint-1>.

5.2 Speculation

Attempting to extract 3-dimensional features from the output of the CVT decoder may be inherently flawed. The current transformer implementation accepts positional embeddings that were learned to map from a 2D projected view (i.e., the multiple camera-view inputs) to a 2D top-down view (i.e., the original BEV map-segmentation CVT was designed for). The current architecture may not allow accurate 3D features to be extracted from the transformer’s output.

Furthermore, since depth is never explicitly calculated but is instead learned by the transformer, it isn’t highly precise. The model’s positional embeddings focus on ground-level relationships as they were designed to guide the model to output geometrically-informed outputs in the BEV space. This means that while height information *can* be inferred through depth estimations, it will likely be highly inaccurate. I.e., while depth estimations are suf-

ficient in a 2D output space, this indirect depth calculation may not be adequate to generating precise 3D predictions.

Future models attempting to utilize cross-view transformers to generate 3D bounding boxes will likely need to encode vertical information into the positional embeddings along with X-Y information to guide the model to predict more accurately in the third dimension. Additionally, models could simply bypass this issue by using explicitly calculated depth through triangulation and stereo matching; CVT successfully attempts to implicitly learn depth, but this estimation isn't sufficiently precise in a 3D space. Finally, future models could also use explicit depth calculations and implicitly predict "adjustments" that are added onto the explicit calculation. This could combat compounding errors that often arise when using imprecise camera calibrations and noisy estimates while still maintaining a reliable *general* depth estimate.

5.3 Ablation Studies

We tested our hypothesis that CVT couldn't perform accurately in the third dimension by removing the LiDAR block (i.e., moving to uni-modal input data, removing the concatenation, and adjusting layer sizes accordingly) of the CVCP-Fusion model to create CVCP. Training with both frozen and unfrozen weights yielded poor results as shown in [figure 4]. CVCP showed similar weaknesses in height predictions, but also failed to accurately predict the lateral (i.e., X-Y) position of objects. This suggests implicit depth calculations may be inherently unstable and may require large parameter sizes to perform well when being used in a larger dimension, which we leave to future work to test. This study further supports our speculation that CVT cannot accurately extract 3-dimensional features and struggles to provide precise depth calculations on larger scales without sufficient parameter count or embedding size.

6 Conclusion

CVCP-Fusion suggests that explicit depth calculations may be required for three-dimensional predictions. Implicit depth esti-

mation using transformers is likely too inaccurate to be used for 3D bounding box prediction. Multi-modal fusion attempts and larger model sizes may remedy the inherent instabilities of relying on a model to bake accurate depth estimation into its weights.

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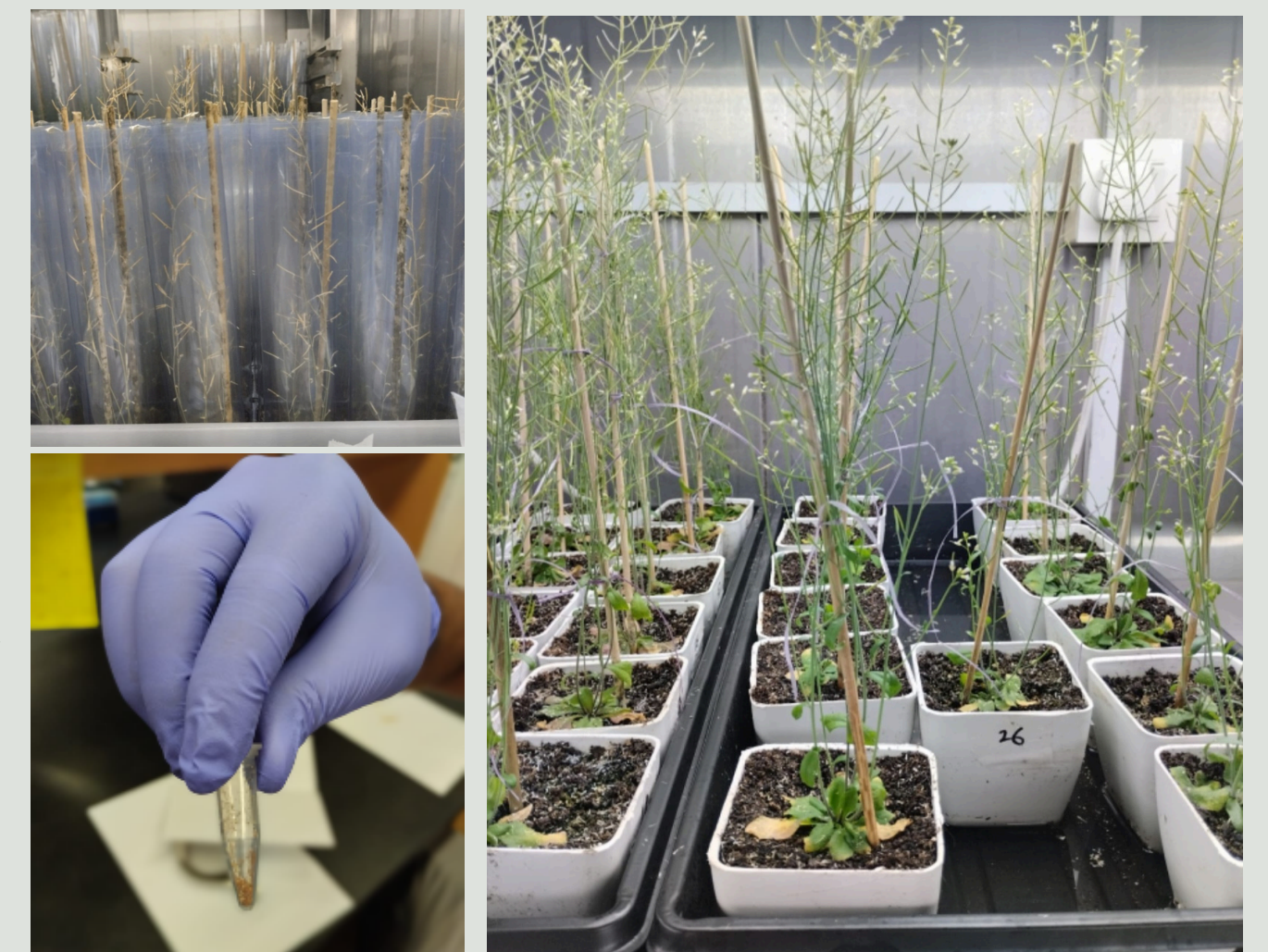
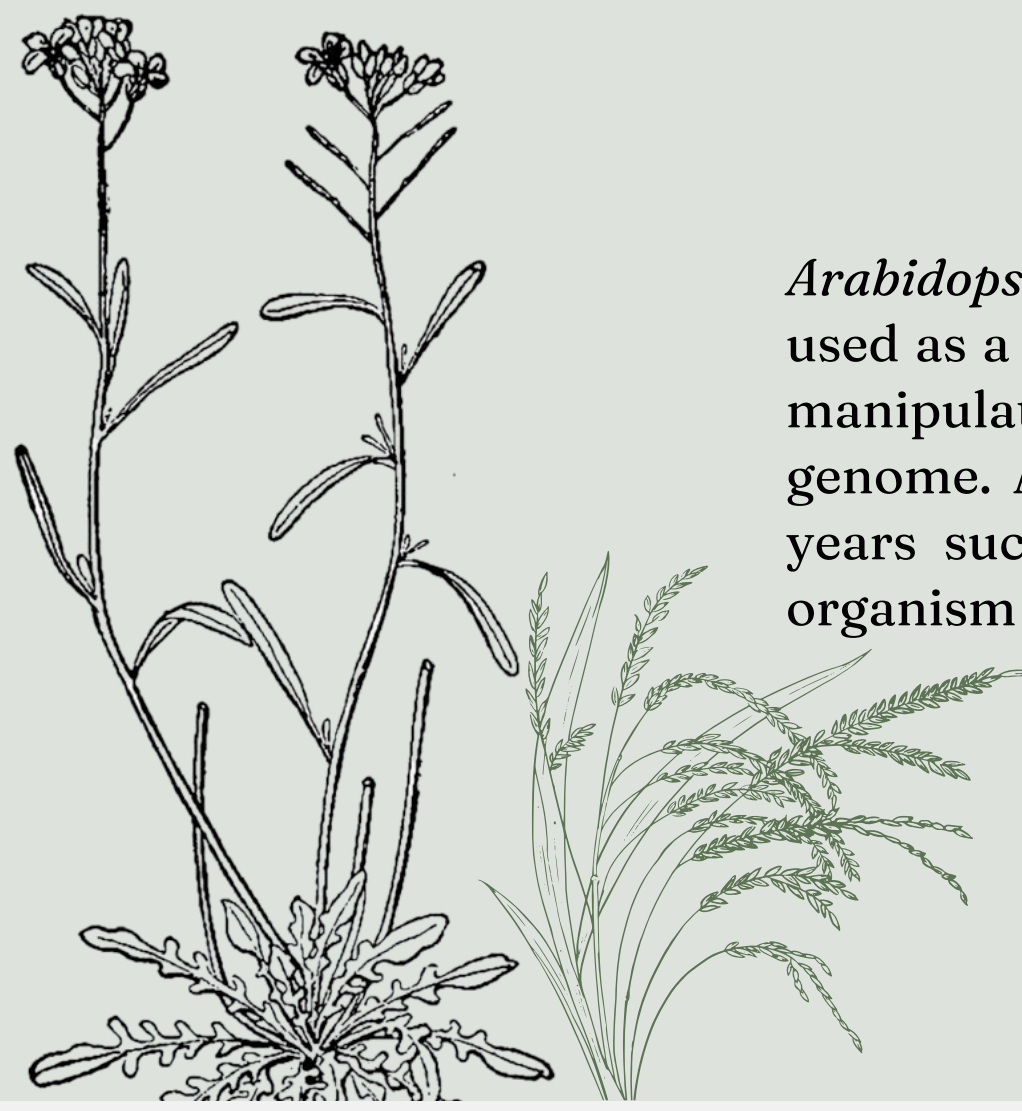
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Introduction & Objectives

Arabidopsis thaliana is a small dicotyledonous species native to both Europe and Africa, used as a model organism for plants. As it possesses a relatively small genome, it can be manipulated through genetic engineering more rapidly and easily than any other plant genome. An extensive toolkit to mold its genes have been developed over the past 20 years such as DNA, RNA and metabolite isolation detection methods. This model organism is an invaluable tool to understand the genetic foundation of plant response to abiotic stresses and can help agriculture workers understand, in addition to deal with the unpredictable and extreme weather changes in Pakistan that affect crops.



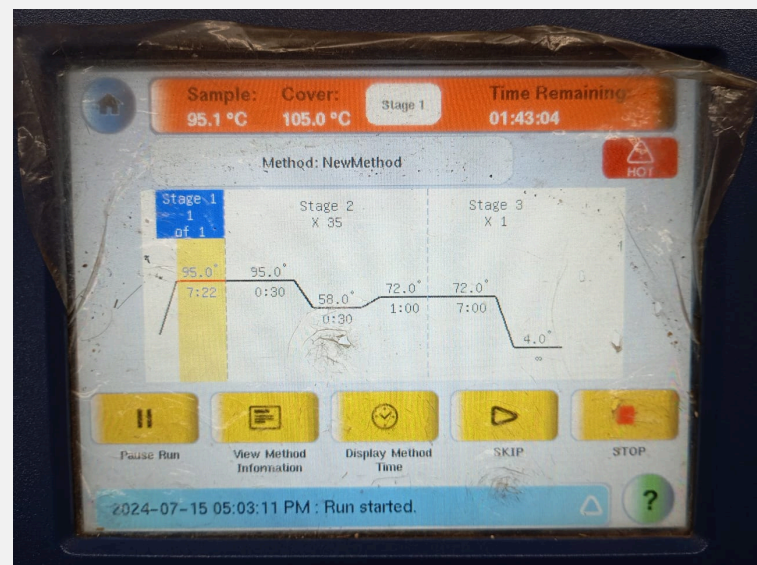
CTAB Method for DNA extraction

DNA extraction begins by breaking down plant tissue, typically using liquid nitrogen to freeze the sample, which is then ground into a fine powder. This powder is mixed with a CTAB buffer to facilitate cell lysis, and RNase A is added to digest RNA. DNA is separated from other cellular components through phenol/chloroform extraction, which creates distinct aqueous and organic phases after centrifugation. The aqueous phase, containing the DNA, is extracted multiple times until clear. The DNA is then precipitated with isopropanol, pelleted by centrifugation, and washed with 70% ethanol to remove salts. After decanting the ethanol and evaporating any residual solvent, the DNA pellet is resuspended in a suitable buffer for downstream applications like PCR or next-generation sequencing (NGS).

RT-PCR

(Reverse Transcriptase-Polymerase chain reaction)

Polymerase Chain Reaction (PCR) is a technique used to amplify a small sample of DNA, making it suitable for analysis. The process involves several key steps: denaturation, where DNA is heated to 95°C to separate the strands; annealing, where the temperature is lowered to 55-72°C to allow primers to bind to the single-stranded DNA; and extension, where Taq polymerase operates at 75-80°C to synthesize new DNA strands by adding nucleotides to the primers. Taq polymerase, derived from the heat-resistant bacterium *Thermus aquaticus*, is essential for this process, requiring Mg²⁺ ions as a cofactor. Typically, 30-40 cycles of these steps are performed to exponentially amplify the target DNA. The PCR setup includes primers, nucleotides, a template strand, a buffer, and nuclease-free water.



NGS (Next Generation Sequencing)

Next Generation Sequencing (NGS) is a powerful method for analyzing genetic material, enabling rapid sequencing of large amounts of DNA or RNA. Unlike PCR, which can only detect known sequences and is cost-effective for small target numbers, NGS can identify both known and unknown sequences with higher sensitivity (down to 1%) and greater mutation resolution, making it suitable for sequencing millions of small DNA fragments. The NGS process involves several key steps: nucleic acid extraction, library preparation (where DNA/RNA samples are fragmented and specialized adapters are added to create sequencing libraries), clonal amplification, and sequencing by synthesis (SBS), where the library is loaded onto a sequencer and each nucleotide is read individually. Finally, the raw sequencing data undergoes processing, analysis, and interpretation to provide meaningful insights into the genetic material.

Designing Primers

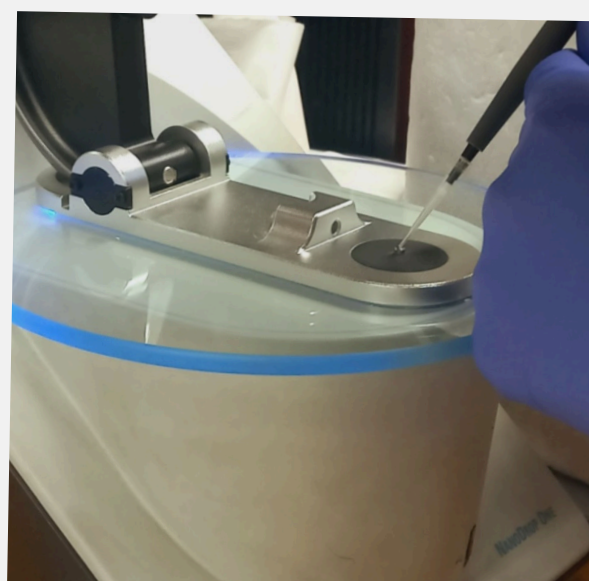
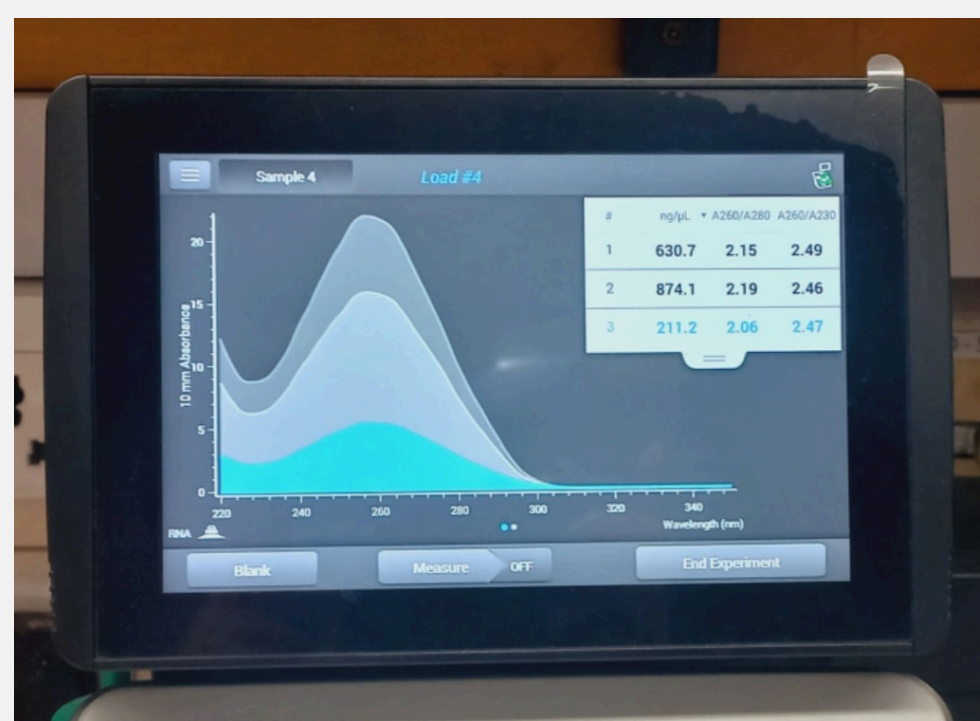
Primers are short single-stranded DNA sequences that serve as starting points for DNA synthesis in PCR. There are two types of primers: the forward primer, which synthesizes the upper strand (5'-3') using the lower strand (3'-5') as a template, and the reverse primer, which is reverse complementary to the upper strand and synthesizes the lower strand (3'-5'). The melting temperature (T_m) of primers is calculated using the formula (A+T) x 2 + (G+C) x 4, and it is crucial for both primers to have similar T_m values to ensure complementary binding. Primers are typically designed to be 18-22 nucleotides long with a GC content of 35%-65% for stability, avoiding sequences that may bind to themselves. Additionally, GC-rich regions are preferred at the 3' end to promote binding, while high GC content at the 5' end can interfere with fluorescent signals in TaqMan probes.

Nanodrop

The NanoDrop is a laboratory instrument used to measure the concentration of DNA, RNA, or proteins using a sample volume of 0.5-2 µL. To use it, clean the sensors with tissue, perform a blank measurement with water, and then place the sample on the lens. The instrument provides key purity ratios:

- 260/280 ratio: indicates nucleic acid purity, with 1.8-2.0 for pure DNA and 2.0-2.2 for pure RNA.
- 230/260 ratio: assesses nucleic acid purity, with ideal values between 2.0-2.2.

The readings are influenced by the specific absorbance of nucleotides, with values such as 1.15 for guanine, 4.50 for adenine, and 1.51 for cytosine. The NanoDrop is essential for assessing the quality and concentration of nucleic acids and proteins in molecular biology.



Gel Electrophoresis

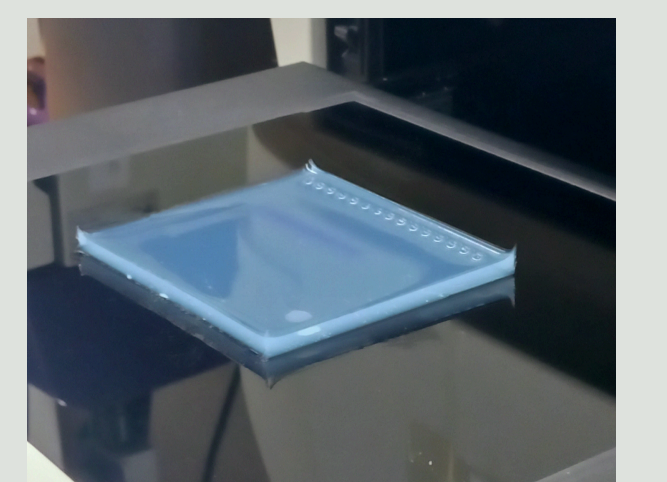
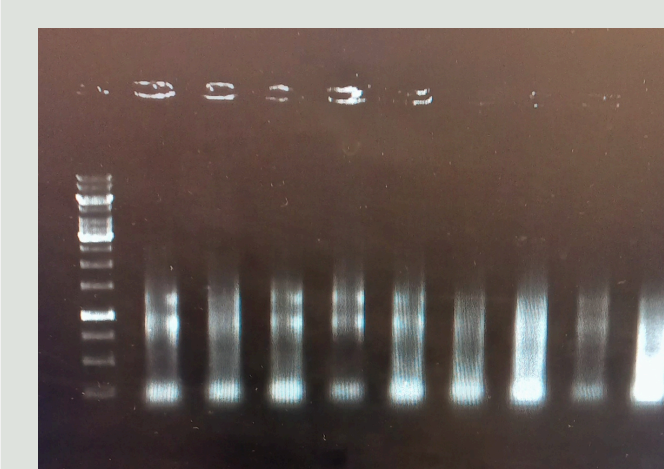
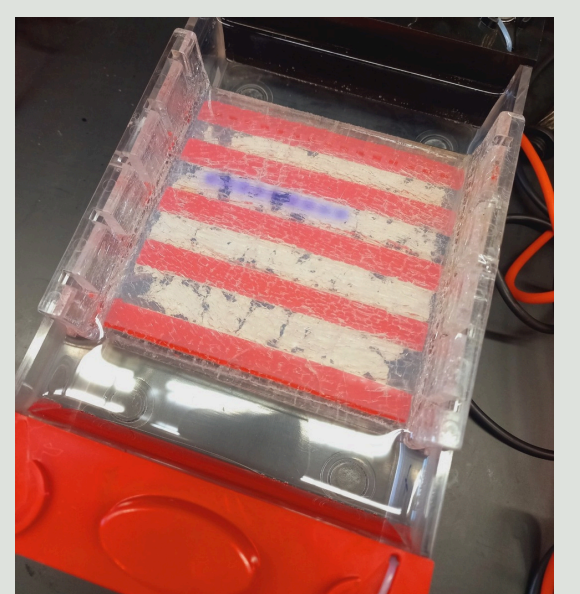
Electrophoresis is a technique for separating DNA and RNA based on their size and electrical charge, and it is commonly employed after PCR or DNA cloning to visualize the amplified products. This method is essential for several analytical purposes, including determining whether nucleic acids are intact or degraded, confirming the presence of RNA or DNA, assessing the purity of the samples, and verifying that the nucleic acids are of the desired size for subsequent analyses.

The separation process utilizes agarose gel, a polysaccharide derived from seaweed. When agarose is polymerized, it forms a network of pores that acts as a molecular sieve, allowing smaller nucleic acid fragments to migrate more quickly than larger ones. The gel is prepared by heating it with a buffer solution, which can be either TAE (Tris-acetate-EDTA) or TBE (Tris-borate-EDTA). TAE is particularly effective for separating larger DNA fragments, making it ideal for cloning applications, while TBE is better suited for smaller fragments and produces sharper bands.

To visualize the DNA, ethidium bromide (EtBr), a DNA-binding stain, is often incorporated into the gel. EtBr fluoresces under UV light, allowing for the detection of DNA bands.

The samples are mixed with a loading dye, which contains glycerol and a tracking dye (such as bromophenol blue). The loading dye increases the density of the sample, allowing it to sink into the wells of the gel, and provides a visual marker to track the progress of the electrophoresis. A DNA ladder, which is a reference sample containing DNA fragments of known lengths, is also loaded into one of the wells to facilitate size estimation of the separated bands.

An electric current is applied across the gel, with the negatively charged nucleic acids migrating towards the positive electrode due to the presence of negatively charged phosphate groups in their backbone. As the current flows, smaller DNA fragments navigate through the agarose matrix more rapidly than larger fragments, resulting in the formation of distinct bands. The gel is then examined under UV light.



Conclusion

Techniques such as PCR, gel electrophoresis, nanodrop and various others have revolutionized our ability to decipher genetic information with unprecedented speed and accuracy enabling significant developments in crop improvement, plant disease management etc. It is the role of our future scientists to adhere to sustainability principles and focus on the Sustainable Development goals such as climate action, life on land and various other from which we can build a resilient and equitable future.

