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HAS MAIZE OVERTAKEN OUR REALITY?

A PERSONAL BRIEFING, BIOCHEMICAL COMPARISON, AGRIGENOMICS, AND HISTORY OF MAIZE

By

Nader Pahlevan

A thesis submitted to the faculty of The University of Mississippi in partial fulfillment of the requirements of the Sally McDonnell Barksdale Honors College.

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ABSTRACT

Nader Pahlevan: **HAS MAIZE OVERTAKEN OUR REALITY? A PERSONAL BRIEFING, BIOCHEMICAL COMPARISON, AGRIGENOMICS, AND HISTORY OF MAIZE**

Maize (*Zea mays ssp. Mays*) is a revolutionary cereal grain that has raced to the world's most popular staple crop, transforming societies and impacting history. This paper aims to build and portray the story maize has created through its journey to world domination. The important details that encompass this literature are maize's cultural significance in my life's story, the comparison of various starches broken down into amylose and amylopectin ratios, a summative historical account on maize's spread throughout numerous parts of the old world, and the genetical analysis of maize that explains the key features that have led it to be the king of crops. By the end of this paper, it should help explain how maize has sprung to the current level of importance and why its current position has caused human societies to be fully dependent on this crop.

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CHAPTER 1

My Maize Through Life

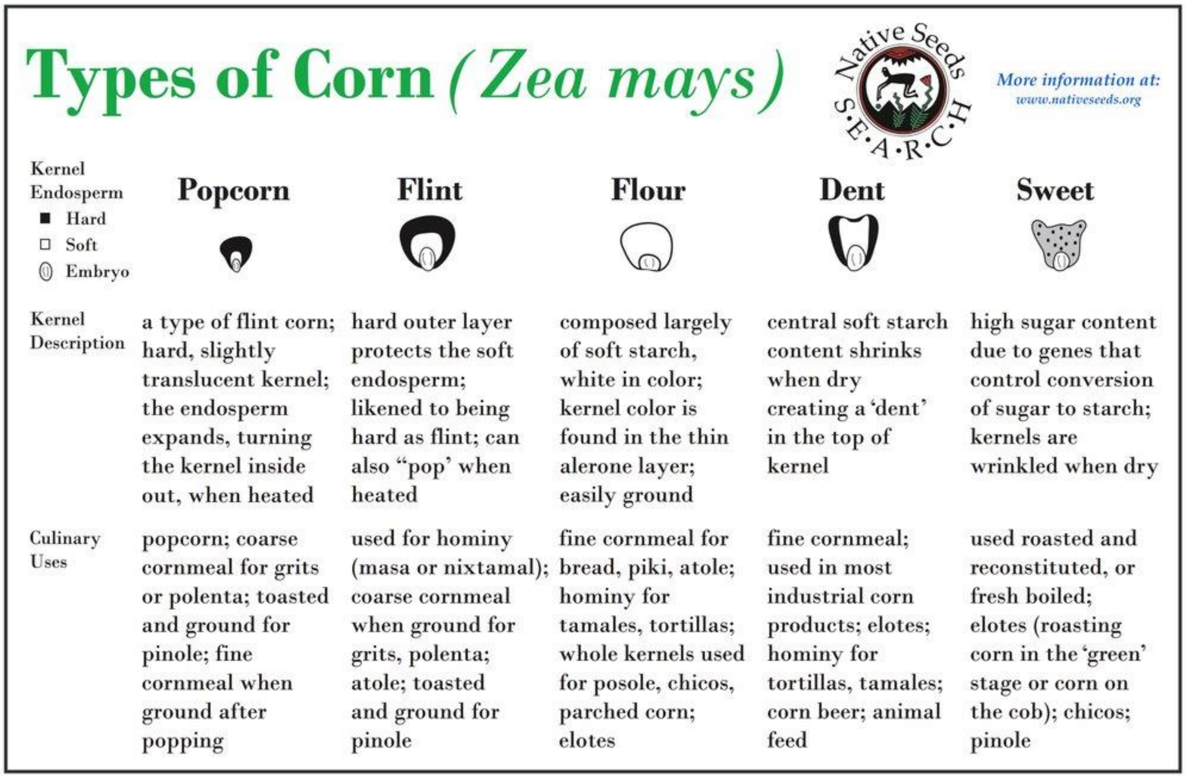
When you think of maize, what images come to mind? Certainly, an all-time favorite food for many, corn on the cob, would be a guaranteed guess. Maize, or better known as corn, is a slender cylinder covered with papery husks that when peeled back reveal neat rows of tightly packed succulent seeds, known as kernels. In its natural form, it can come in a variety of colors and have a range of uses, ranging from a household ornament during the winter seasons to a delicious comestible thrown on a grill during a family barbeque. Corn can also be found in altered forms in many products in our daily lives; some of those things include the popular high fructose corn syrup found in many foods as a sweetener, ethanol that makes up a large portion of gasoline, and cornstarch, which is found in cosmetics, adhesives and other household products. These examples are just a small fraction of products that are composed of the carbons of corn.

Corn is so intrinsic to myself and my culture that it might as well be etched into my DNA. As a Romanian child, I had a natural preference for the foods of my homeland and family. When I immigrated to the US, of course that preference remained, and those foods were available in only one place. To this day, there are specialty cultural foods that are etched into my very being through my mom's exquisite skill and effort, such as cabbage rolls and stuffed bell peppers. Imagine what happened when the younger version of me was faced with cheese pizza, hotdogs, and chicken nuggets instead of my mom's Romanian delicacies. Out with the old, in with the new. With that said, there was one Romanian food that pushed past even my picky childhood palate, mămăligă.

Mămăligă, also known as polenta, is a simple dish made from corn flour—better known as cornmeal—and water, and you can add a pinch of salt if you are feeling fancy. If I had to compare it to a familiar American dish, its consistency would be quite similar to grits. This cultural dish was the main component of my life as an immigrant, and it kept me connected to my Romanian heritage so much so that it was the very first meal I learned to cook. Now every time I cook it, I associate it not only with the taste of my mother’s kitchen, but it is also the keystone that unites me to the past generations of Romanians before me.

This cornmeal dish led to the inspiration that gave rise to the beginning of my journey down the path of researching maize. I came to the University of Mississippi with scientific research in mind but nothing specific enough to begin investigating. The interdisciplinary emphasis of coursework through the Honors College and my introduction to Biochemistry brought me to a path I had no idea was possible: the intersection of culture and food Biochemistry. This had my mindset onto a goal of researching a consumable food that was intertwined in our human framework and was culturally significant to me. No better food stuck out more to me than maize. The idea of maize—a new world crop—being so widespread in Europe, and specifically Romania, made me wonder how this was possible. With corn just being introduced to the old world a little over 500 years ago, it is a very short time period in retrospect of other comparable foods that were then either replaced by corn or did not flourish as well as corn. Furthermore, it is worth mentioning that the amount of genetic variation—a term coined genetically modified organisms (GMOs)—that corn has been subject to and the manipulation of its contents over the past centuries have been unprecedented when compared to other food products. With these modifications, it has turned into fuel for cars, fuel for animals, and fuel for humans.[1]

My curiosity was piqued as I believed that replacing previous food sources for calories that have been there for generations with corn must have some transverse effects. I was especially intrigued by the idea that many of the proteins we consume have had some traces of corn, whether that is beef, poultry, and even fish if it is farm-raised. To further add to this notion, corn is not the original plant that it was 500 or even 50 years ago. It has now been severely modified to yield different varieties with a range of culinary and industrial uses. These corn types include: popcorn, which is the oldest type of domesticated corn and is ground for use in grits and polenta; flint, which has a glassy outer appearance and is ground for use in masa and nixtamal; flour, which is composed largely of soft starch and used for fine cornmeal used in breads; dent, which is better known as “field corn” and is commercially used in the United States for animal feed, processed foods, and ethanol; and last but not least, sweet, which is the standard corn used for human consumption due to its low starch and high sugar content.[2] More information about these varieties are found in **Figure 1**. Furthermore, most corn products even add a crucial vitamin known as niacin, which is absent in corn. This vitamin is important for preventing one from acquiring pellagra, which is caused by a high corn diet without the supplement. Even with all this knowledge known to us about the Frankenstein that maize is, people around the world are not slowing down in consuming it and some, even if not by choice, use it as a primary food source. This then brings me back to the centralization of the corn product, mămăligă, in Romania.



Types of Corn (*Zea mays*)

Native Seeds S.E.A.R.C.H

More information at: www.nativeseeds.org

Kernel Endosperm ■ Hard □ Soft ◎ Embryo	Popcorn	Flint	Flour	Dent	Sweet
Kernel Description	a type of flint corn; hard, slightly translucent kernel; the endosperm expands, turning the kernel inside out, when heated	hard outer layer protects the soft endosperm; likened to being hard as flint; can also “pop’ when heated	composed largely of soft starch, white in color; kernel color is found in the thin alerone layer; easily ground	central soft starch content shrinks when dry creating a ‘dent’ in the top of kernel	high sugar content due to genes that control conversion of sugar to starch; kernels are wrinkled when dry
Culinary Uses	popcorn; coarse cornmeal for grits or polenta; toasted and ground for pinole; fine cornmeal when ground after popping	used for hominy (masa or nixtamal); coarse cornmeal when ground for grits, polenta; atole; toasted and ground for pinole	fine cornmeal for bread, piki, atole; hominy for tamales, tortillas; whole kernels used for posole, chicos, parched corn; elotes	fine cornmeal; used in most industrial corn products; elotes; hominy for tortillas, tamales; corn beer; animal feed	used roasted and reconstituted, or fresh boiled; elotes (roasting corn in the ‘green’ stage or corn on the cob); chicos; pinole

Figure 1: Index of maize (*Zea mays*) varieties[1]

From what I know historically, this dish was originally popularized for its price-to-calorie ratio. It was cheap and could fill up a lot of people. It was mainly thought of as a “low class” meal. However, over many years, it began to integrate into my culture. It was still a cheap meal that could be eaten, but now it was much more than that. Many more people from different economic statuses would indulge in this cornmeal dish. However, it would now be paired with yogurt, cheese, butter, and other classical Romanian dishes, such as cabbage rolls and stuffed bell peppers. It has even gotten to a point where the dish can be turned into something similar to a casserole and thrown in the oven. With these “new” modifications, it would even constitute a more well-rounded diet. It easily became a signature staple served at basically all gatherings and at almost every restaurant you stop at. I would even go as far as to say that the dish is so signature to my Romanian heritage that it is burned into Romanian DNA. Of course, I say that figuratively, but it does seem like something similar.

As mentioned before, I was a very picky eater but for some reason, I immediately fell in love with mămăligă. I can even say the same thing for my younger sister who was born in the United States. However, in contrast, when I share it with my friends that have different ethnicities, as is a tradition for me at this point, I have noticed some interesting correlations. My Hispanic and Asian friends enjoy it so much that they ask for seconds, my White-American friends tend to like it enough to finish the first bowl, and my African-American and Asian-American friends usually cannot finish a bowl. While this could be genetic, based on how one’s taste buds react to the cornmeal, it seems to me that the culture and environment a person is raised in shapes their palate towards certain foods. My sample size is by no means large enough to be concluded as proof but is an interesting correlation that I noticed in my diverse cohort.

Corn is a mystical plant that has traversed thousands of miles and hundreds of years. With this distance and time, it has managed to gather quite a historical reputation in our human societies. Also, it has aided in propelling our knowledge in the world of agrigenomics that has allowed us to implement corn in so many different variations. Furthermore, with its easily manipulated genome and plentiful yield, it has integrated itself as a key component in structuring our society, both directly and indirectly. The major thesis of this document is that from its humble beginnings, the conquest of corn across the globe has transformed societies and imprinted social structure and practice. I will begin by discussing and comparing the chemical breakdown between cornstarch and other starches in regards to their amylose and amylopectin ratios, and why the high abundance of this readily digestible, soluble carbon has made corn king. Then I will delve into my recollection of maize concerning my cultural background and how it has impacted me. Also, an overall history of corn will be discussed with a specific note to its current socioeconomic benefits and disadvantages. Finally, the agrigenomics of the maize plant will be explored to expand upon the changes it has experienced over the years and how humanity has used this crop to spawn robust research in genetic modification.

CHAPTER 2

Carbohydrates

Maize has become so intertwined into our society that it would be fair to proclaim ourselves as maize people, quite literally. Because of maize's unique preference to the ^{13}C isotope over the ^{12}C isotope, it lets us easily estimate the amount of carbon in a typical American dinner.[2] Jahren, et al. reported that of the 160 food products that they tested from Wendy's restaurant from across the United States, every person could be traced back to a maize source.[3] In general, those who consume more C-4 plants such as maize in their diet will have a larger amount of ^{13}C present in their body. For example, a study in Alaska showed that the younger population, who consumed more market foods, had much larger ^{13}C levels when compared to the older population, who were strictly on a marine-based diet.[4] This shows that the products of C-4 plants found in processed foods is enough to influence what types of carbon is found in a specific individual.

With the villainization of fat, we became a carbohydrate-crazed society with a decades-long focus on carbohydrate-rich foods.[5] Why do we love our sugar-rich food and drink so much? Perhaps an important factor to maize's rise in popularity among the majority is the connection between carbohydrates and serotonin, a neurotransmitter that is considered to "reward" behavior.[6] With the increased consumption of carbohydrates, an increase of serotonin also follows.[7] This mechanism is caused by the increase of blood glucose from ingesting carbohydrates, which then leads to an insulin response. Insulin, in turn, triggers an increase the amount of tryptophan, a common amino acid, found in the blood. Since serotonin is a product created from tryptophan, eating carbohydrates indirectly stimulates your body to feel rewarded. As this sensation can become addicting, the subconscious hunger to satisfy this need for reward is quickly filled by maize-containing because of their ubiquity in processed foods and soft drinks. Further, maize is a crop with large yield production and compatibility with most soil types.

Maize, as a crop, separates itself from other carbohydrates by its diverse varieties that can be applied in many different fields such as cooking, commercial engineering, and genetic research. It is a crop that has quickly risen to the top in a relatively short time period. What exactly could explain this boom? Several factors are at play that depend upon carbon fixation. During photosynthesis oxidized carbon in the form of CO_2 is fixed in a reductive process into carbohydrates that are then used for the purposes of growth, storage and readily available energy. It derives almost all of its CO_2 from the atmosphere (97%),[8] hence its suitability for even poor soils. Most importantly, it is a C-4 plant. C-4 plants form a 4-C compound, oxaloacetate, rather than follow the typical C-3 pathway that leads to 3-phosphoglycerate (or the C-2 pathway that does not even have O_2 as a product).[9] When looking at the C-4 pathway of maize, apparently this means that the mechanics of the CO_2 uptake require less water. Further, the uptake and fixation of atmospheric CO_2 by maize means that this cereal grain is an excellent target crop to be used to reduce atmospheric CO_2 pollution caused by the combustion of fossil fuels.[10]

Specific grains are used for specific purposes, and there are even specific maize varieties for specific purposes. We had interest in distinguishing the properties of relative to other grains such as rice, wheat, potato, and tapioca. Likely, its versatility is due to the ratio of amylose and amylopectin in its starch component. To this end, we spent some time developing an assay to measure this ratio as compared to carbohydrates mentioned previously using an amylose/amylopectin assay kit.

Materials

This assay works by taking advantage of the fact that amylopectin has many non-reducing ends and amylose only has one. In a reagent that complexes with non-reducing ends, the amylopectin will precipitate, leaving behind the amylose in solution. Amylose is the cleaved into individual monosaccharides. Their concentration is then determined calorimetrically. The starches used in this experiment were tapioca, wheat, potato, and rice. The actual enzymes and reagents are described below. The Amylose/Amylopectin kit contained 6 bottles of materials that were imperative for the assay to work. Bottle 1 contained freeze dried concanavalin A (Con A) that when used under defined conditions of pH, temperature, and ionic strength, complexes branched polysaccharides based on α-D-glucose units at multiple *non-reducing* end-groups (C-4) with the formation of a precipitate. Con A effectively complexes the amylopectin component of starch but not the primarily linear amylose component.[11] Bottle 2 contained amyloglucosidase [200 U on ρ-nitrophenyl β-maltoside] plus fungal α-amylase, which is an enzyme that hydrolysis α-1,4 and branching α-1, 6 linkages (glucose units) in liquefied starch.[12] Bottle 3 contained Buffer (50mL, pH 7.4), ρ-hydroxybenzoic acid, and sodium azide which is the Glucose Determination Reagent (GO-POD Reagent). Bottle 4 contains Glucose Oxidase plus Peroxidase and 4-aminoantipyrine (GO-POD Reagent Enzymes) that is used to analyze D-glucose, which is enzymatically hydrolyzed from the specific starch's amylose. Bottle 5 contains D-Glucose standard solution in 0.2% (w/v) benzoic acid. Bottle 6 contains a starch reference sample that has a specified content of amylose. Buffers and solvents not supplied by the kit were also necessary. These included: Sodium Acetate Buffer (100 mM, pH 4.5), Concentrated Con A Solvent (600 mM, pH 6.4 sodium acetate buffer), and Dimethyl Sulphoxide (DMSO).

Procedure

The first part of the procedure is performed to isolate the starch sample from lipids or other possible contaminants. The first step is weigh 20-25 mg of starch sample into a 10 mL screw capped tube. Then add 1 mL of DMSO to each tub while gently stirring it at low speed on a vortex mixer. After this heat the contents in a boiling water bath for approximately 1 min until liquid is homogenous. Allow tubes to cool for 5min and then add 2 mL of 95% (v/v) ethanol with continuous stirring on a vortex to mix. Further add 4 mL of ethanol to each tube, then cap and invert to mix, which will cause a starch precipitate to form at the bottom. Let tubes stand for 15 min, then centrifuge at 2,000 g for 5 min. This will cause mixture to separate and allow one to discard the supernatant that has formed. Next, dry the tubes with tissue paper for 10 min, making sure all ethanol from tubes are drained. Then add 2 mL of DMSO to the starch pellet and gently vortex the mixture. Place the tubes in a boiling water bath for 15 min and mix occasionally to ensure there are no gelatinous lumps. Once tubes have been removed from water bath, immediately add 4 mL of Con A solvent and mix thoroughly. Then quantitatively transfer contents of each tube by washing with Con A solvent to a 25 mL volumetric flask. Dilute to volume with Con A solvent. This will be used as Solution A (one for each starch sample) in the next part of the procedure. This finishes the starch pre-treatment procedure.

The next part will be the procedure that forms the Con A precipitation of amylopectin and allows for determination of amylose. Begin by transferring 1 mL of Solution A to a 2 mL microfuge tube. Add 0.50 mL of Con A solution to each Salutation A starch mixture and gently mix by repeated inversion without frothing. Allow all tubes to rest for 1 h at room temperature. Once time has passed, centrifuge at 14,000 g for 10 min in a microfuge. A supernatant will form for each tube, and 15 mL of the supernatant will need to be transferred to centrifuge tubes. After, add 3 mL of 100 mM sodium acetate buffer (pH4.5) to each tube, causing the pH to drop to approximately 5. Then mix the contents and heat in a boiling water bath for 5 min to denature the Con A. Equilibrate the tubes for 5 min in a 40°C water bath. Add 0.1 mL of amyloglucosidase/α-amylase enzyme mixture and incubate at 40°C for 30 min. While this is happening, prepare the Reagent Blank, D-Glucose Controls, and Total Starch aliquot. The Reagent Blank is prepared by adding 1 mL of 100 mM sodium acetate buffer to 4 mL of GOPOD Reagent. The D-Glucose Controls comprise 0.1 mL of D-glucose stand solution (1 mg/mL), 0.9 mL of sodium acetate buffer, and 4 mL of GOPOD Reagent. The Total Starch aliquots are made by mixing 0.5 mL of Solution A with 4 mL of 100 mM sodium acetate buffer (pH 4.5); then, adding 0.1 amyloglucosidase/α-amylase solution to solution and incubating the mixture at 40°C for 10 min; and finally, transferring 1 mL aliquots of this solution to glass test tubes and adding 4 ml of GOPOD Reagent, making sure to mix well. Once sample tubes are finished incubating, centrifuge the tubes at 2,000 g for 5 min. Aliquot 1 mL of the supernatant and add 4 mL of GOPOD Reagent to it. Then incubate sample tubes, Reagent Blank, D-Glucose Controls, and Total Starch aliquots at 40°C for 20 minutes. Finally, read the absorbance of each sample, the D-Glucose Controls and Total Starch aliquots at 510 nm against the Reagent Blank.

Results and Discussion

Once all the absorbances of each sample and standard were collected (**Table 1**), we used **Formula 1** to calculate the amylose percentage. The final calculations for the starch samples and standard are showcased in **Table 2**. By measuring the percentage of amylose found in a specific starch sample, it provided insight that could be used as a mode of comparison on how different starch ratios possibly affect the physical properties.

Starch is constituted by two types of polysaccharides: amylose, a linear α(1-4) linked glucose residues, and the more branched amylopectin, an α(1-4) linked glucose residues with many α(1-6) branch point linkages. By finding the amount of amylose in a specific starch, it gives the ability to create ratios for these samples. Normally, starches contain 20-30% amylose while the remaining 70-80% is amylopectin.[13] This ratio can also vary by whether the starch is waxy which could have <1% amylose, while certain high amylose maize starches have >70% amylose.[13] These values allow for the use of broad amounts of applications based on the desired ratio.

Furthermore, we wanted to test if the difference in gel characteristics of our starch samples correlated with the amylose/amylopectin ratio. We began by testing corn starch which began to gelatinize at 80°C and had a white, viscous, glue consistency, Next, tapioca starch turned white at 80°C and then turned clear and viscous as it cooled. Wheat starch began to turn into a thick cloud white liquid at 91°C, but once it cooled to 80°C, it started separating into white and clear with very low viscosity. Potato starch was added to 80°C and immediately turned into a high viscosity, clear gel once the temperature reached 55°C. Finally, rice starch was added to 80°C water and began to thicken in a clumpy, white mush at 86°C. While these results do not give a conclusive answer as to whether the amylose/amylopectin ratio predicts gel characteristics, the results could have been affected by other starch characteristics such as granule size, crystalline structure, and amylopectin chain length.

Starch Samples:	Con A Supernatant Absorbance (nm):	Total Starch Aliquot Absorbance (nm):
Standard	0.19359	0.20180
Tapioca	0.03692	0.18037
Potato	0.04982	0.14805
Wheat	0.11738	0.13374
Rice	0.08435	0.25879

Table 1: Absorbances for Starch Samples and Total Starch

$$\frac{\left(Absorbance\left(Con\ A\ Supernatant\right)\right)}{\left(Absorbance\left(Total\ Starch\ Aliquot\right)\right)}\left(\times\left(\frac{6.15}{9.2}\right)\right)\left(\times 100\right)$$

Formula 1: Calculation of Amylose Percentage Content. Where 6.15 and 9.2 are dilution factors for Con a and Total Starch extracts, respectively.

Starch Samples:	Amylose (%)	Percent Error (%)
Standard	68.0	3.9
Tapioca	16.5	2.8
Potato	20	2.5
Wheat	25	2.0
Rice	20	1.8

Table 2: Absorbances for Starch Samples and Total Starch

The starch reference sample was reported to have 71.9% amylose. Thus, based on our assay, we have a 3.9% relative standard error in the measurement. Further, we find that high amylose maize, produced as a byproduct from the creation of so many variates, has the least amylopectin in comparison to the other high starch foods that were tested.[13] This high amylose content is the reason why corn starch is such a good thickener in gravies and sauces.

CHAPTER 3

History of Maize

The maize plant as we are used to seeing it with its thick cylindrical structure lined with bright chunky kernels looked vastly different at its first conception. Even the name that we give this plant has changed over the centuries; the name that it has currently adopted in the United States and Britain is corn, a word that was used to represent all types of grains.[14] From its earliest known form—about 8 to 9 thousand years ago[15]—to how it has cultivated itself as the pinnacle of the all-purpose entity of the present, this section aims to discuss the widespread history of maize throughout time.

Origins

Using modern DNA evidence, experts have found that maize's ancient ancestor was a wild grass called teosinte, or *Zea mays ssp. Mexicana*;[15] however, there has also been discussion that it could have evolved from an earlier Mesoamerican maize variety called Chapalote.[16] Even if the genetic makeup of modern maize and teosinte have distinct similarities, according to plant geneticist John Doebley, maize and teosintes differ profoundly in terms of vegetative characteristics and inflorescences architecture, which is defined as how the flowers form on branched stems and the branching pattern of the plant.[16] Even with these distinctions, Doebley agrees that the Mexican teosintes and modern maize are variants of the same biological species.

As for maize's reproductive pattern, it has not changed much as it still resembles a primitive concept. The plant includes both male and female reproductive organs which allow for self-pollination and self-reproduction. It disperses the pollen from the tassels to the silk of the maize ears, female inflorescences, where the reproductive process activates. Through the process of self-fertilization, it is difficult for the maize plant to naturally acquire genetic diversities since it does not require having 2 parents with distinct chromosomes; however, it currently has 25 primary races around Mesoamerican from the use of genetic modifications, which are far from pure to the original plant.

The earliest form of maize that has currently been discovered is an ancient preserved cob dating to 5,310 years ago. The DNA sample of the ancient cob, known as Tehuacan162, was used to map its genome and compare it to modern maize through molecular analysis. The ancient cob is pictured in **Figure 2** below. The specimen is a 10th of the size of modern maize cobs and about 2cm long. Its cob only contained 8 rows of kernels, half the amount found in modern maize. Surprisingly, the genome sequence shows that the ancient plant is more similar to modern maize than to its wild grassy ancestor, teosinte. This reveals that even though maize has recently changed drastically due to genetic modifications, the natural mutation that occurred centuries prior created a plan with soft and palatable kernels that would lead to its explosive expansion in the New and Old Worlds.[15]

Figure 2: Tehuacan162, the ancient ancestor of modern maize.
It contains eight rows of kernels, half the number in modern maize[1].

Maize Throughout Parts of the Old World

Maize has been a crop that seems to have been stained into our human history for as long as one could remember. The crop itself was not even rediscovered by European explorers until Columbus' first voyage to the Americans in 1492. Even for the first individuals that were fortunate enough to lay their eyes on maize, they were astonished about the similarities and differences it had with the "corn" they were familiar with, known as millet. Its first description in Old World literature was recorded in a letter of Pedro Martyr of Anghiera that dates back to November 13, 1493. However, this crop had not made its way over to the Old World from the Americas until some ships from Columbus' second voyage returned to Spain on May 3, 1494, initiating the start of the Columbian exchange.[17] Through the Columbian exchange, maize quickly diffused through European and Asian countries, such as Spain, Portugal, Italy, Turkey, and China, at a rapid rate.

The fast expansion of maize can be showcased through the vast amounts of artforms that have depicted the plant within just a few years that it was introduced to the Old World. The earliest art form is traced to 1517 in the frescoed festoon painted by Giovanni Martini in the Vatican (**Figure 3A**). As more time progressed, maize seemed to make a common appearance in other artworks for different purposes. An example of this is found in Giuseppe Arcimboldo's paintings *Summer* and the *Portrait of Rudolph II as Vertumnus* (**Figure 3B**) where the artists use maize for ears, most likely inspired by the term "ear" used as a synonym for the husk of maize. Furthermore, maize was not restricted to only becoming popular in Europe as it held strong significance in Asian countries as seen by a humorous depiction of maize in Japanese woodcuts (**Figure 3C**).[17] At just an early stage, this crop was already forming its reputation as a potential dominator in human cuisine.

Figure 3: Early Old World and Asian images of maize in Art.

(A) Images of maize from the Loggia of Cupid and Psyche, Vialla Farnesina, 1515-1518.[17]

(B) Maize as ears in paintings of Guiseppe Arcimboldo: (left) Summer, 1573; (right) portrait of Rudolph II as Vertumnus, 1590.[17]

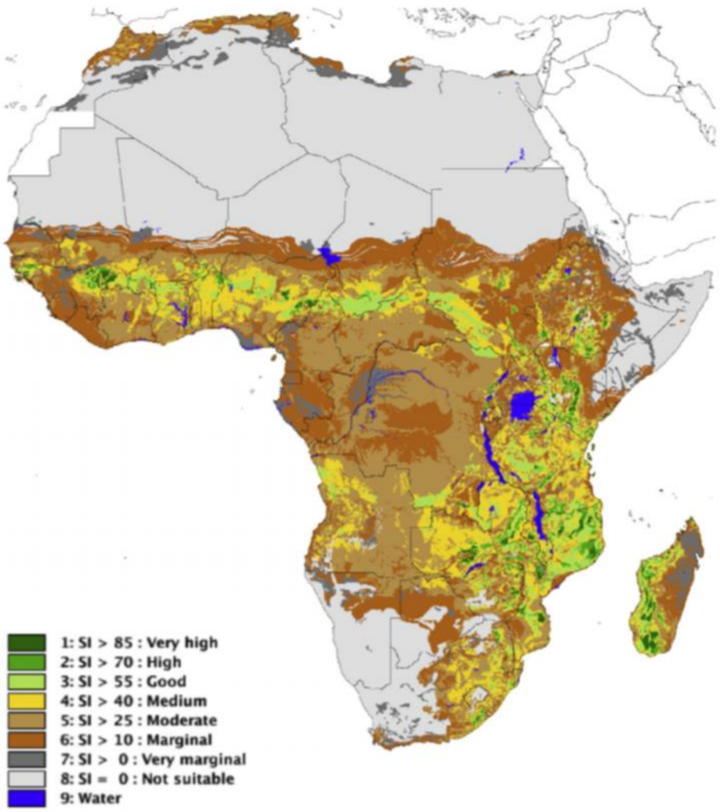
(C) Humorous maize anomalies in Japanese woodcuts.[17]

However, even with maize's increase in global popularity, it did not begin its journey with a bright uprising. The vast majority of people during that time formed mixed feelings about this new rising crop that went by names such as Turkish wheat, Turkish grain, Spanish wheat, and

Indian Corn.[17] These perceived biases came from the rumors that maize was less nutritional when compared to grains that were already produced and sold in Europe. This was, most likely, falsely spread around just as quickly as maize was by millers, who did not want to hinder their profits, in hopes of tarnishing the New World crop's name and stopping any rising competition in the grain industry. Even though maize could be seen to have a greater yield with a shorter growing season, only adding to the benefits that it could contribute to society, it was labeled as a foodstuff that should only be served to animals and the poorest of the peasantry. As the majority of Europeans were Christian, the importance of the religious significance of wheat, the grain of life, could not be replaced by an imitation grain.[18] This blasphemous notion stamped maize as being practically worthless. For the unfortunate peasants that had to consume the unholy grain out of necessity, they would grind up the kernels of the plant into a fine white powder, which would at least resemble white flour, and create a mush by mixing it with water. This, eventually, would be how the northern Italian dish of polenta, or known in Romania as mămăligă, would come into existence.

While Europeans were attempting to stop the revolutionary changes maize would soon cause in human society, it was already creating a huge impact in the continents of Asia and Africa. In Asia, maize was quickly adopted so that it could be cultivated in rotation with other crops, such as rice and millet, increasing the food supply available for the populations. By reaping the benefits that maize provided, it allowed the increase of growing seasons which then caused an exponential growth in populations. Similarly, in Africa, maize came to dominate the economics and society in many countries. **Figure 4** shows the varying suitability of the land in growing maize, which shows most of Africa was able to produce the grain at moderate levels. While this meant more consumers and workers for a specific country, this also led to some very negative outcomes by fueling, what would later be known as, the transatlantic slave trade.[19]

A



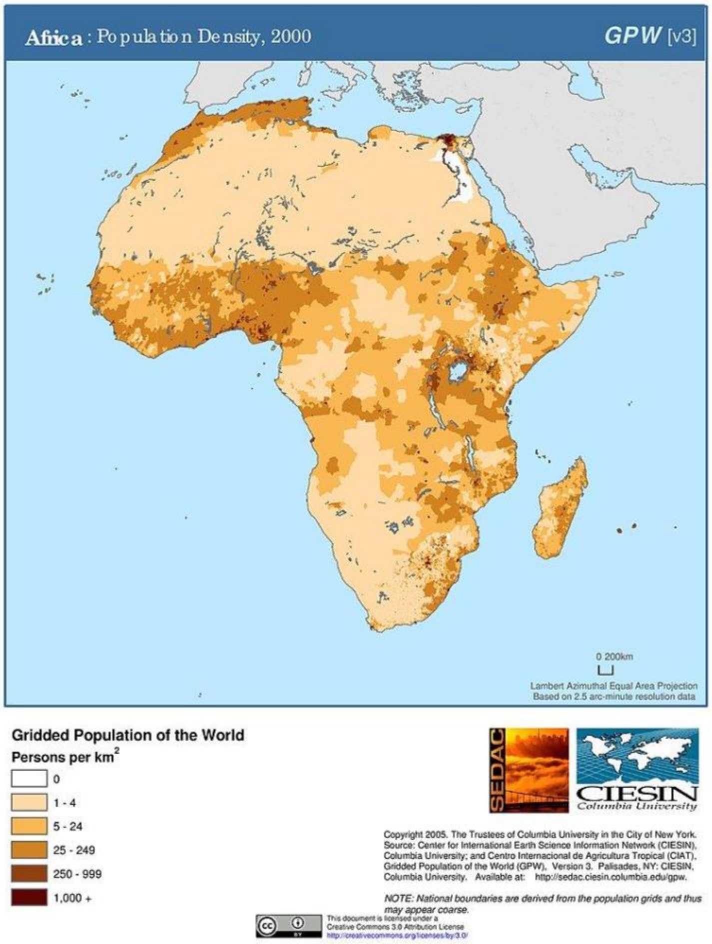
B

Figure 4: African Maps

(A) The Suitability of land for the cultivation of maize in Africa.[19]

(B) Population Density of Africa.[20]

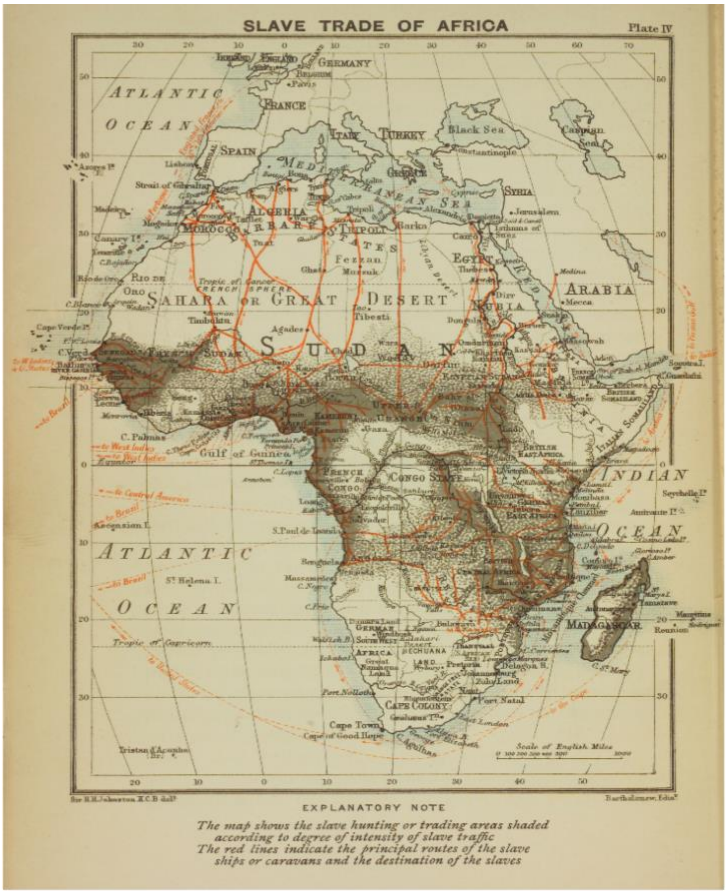
(C) Regions in Africa that showed the most intense slave trade activity.[21]



Source: Center for International Earth Science Information Network (CIESIN), Columbia University; and Centro Internacional de Agricultura Tropical (CIAT), Gridded Population of the World (GPW), Version 3. Palisades, NY: CIESIN, Columbia University. Available at: http://sedac.ciesin.columbia.edu/gpw.

NOTE: National boundaries are derived from the population grids and thus may appear coarse.

C



The integration of maize in African countries made it extremely efficient and cheap for European and Arabic slave traders to transport large amounts of people from there. This was one of the many locations that maize impacted the social structure the most. The rise of commercial cash crop farming was brought in by the colonists with the excuse of creating a plentiful source of food and income for the African people.[22] For example, maize had similar nutritional levels to the indigenous staples millet and sorghum but could be produced in higher yields with lower labor requirements.[19] However, in the end, this led to the colonizer's pocket books filling, whereas leaving the local population socially and physically starved. The wide variety of less commercial indigenous crops that had previously contributed to the economical and nutritional value of African societies were quickly being replaced.[22] This implementation prompted an explosion of both population density and slave exports during the precolonial era. By the 1950s, once most African countries were "freed" from colonist rule, the shrinking indigenous food supply left most of the area in a food drought that forced them to rely on foreign aid to survive.[22]

Furthermore, the introduction of cash crops such as maize did not cause any meaningful effect on urbanization rates, suggesting that the economic growth of the region was not stimulated.[19] This was in part to the European settlers seizing any and all arable land in Eastern Africa, leaving the locals with small segments of desolate land. Unfortunately, the overtaking by these colonizers caused much of the indigenous markets in this area to evaporate, removing one of the main sources of economic, political, social, and judicial activities from the African people. As the Africans did not have any power to withstand the "New World Order" imposed by the invading outsiders, they were forced to watch their traditional way of living be stripped away while having their populations artificially inflated.[23] This made it so the African populations were unable to escape the Malthusian trap, which is when a population increases faster than the production of food that will eventually lead to a shortage and cause famine.

Furthermore, the implementation of maize could have very well increased the amount of conflict seen in Africa. It is indicated that the introduction of the new world crop led to a 7.6% increase in the likelihood of conflict in a specified country. This increase in conflict is seen to have mainly been created due to maize increasing the size of the slave trade.[19] It is also important to note that based on its land suitability for growing maize, each country had an increase or decrease of the factors mentioned earlier.

As the spread of maize became more rapid, as if it had spawned wings and started soaring across the globe, it eventually reached the continent of Asia, specifically affecting China. Similar to Africa, maize did not help China break out of the Malthusian trap and could even be credited to increasing its effects. While the population did sharply rise, especially after the 18th century, the per capita income was barely moved, causing a discrepancy between population and economic growth due to maize. Furthermore, maize diffusion through China was not an overnight phenomenon. It took many generations of farmers to realize the massive benefits of the crops, such as resistance to drought and cold weather, to finally take off in its spread after 1750 as seen in the images below (**Figures 5A** and **5B**).[24]

A

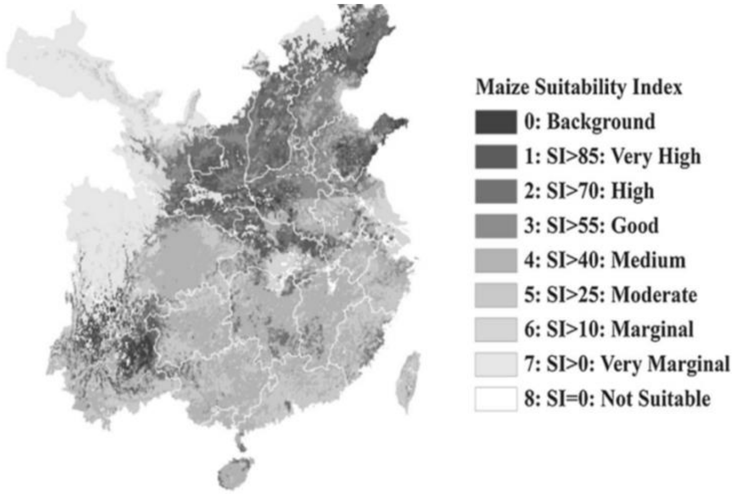
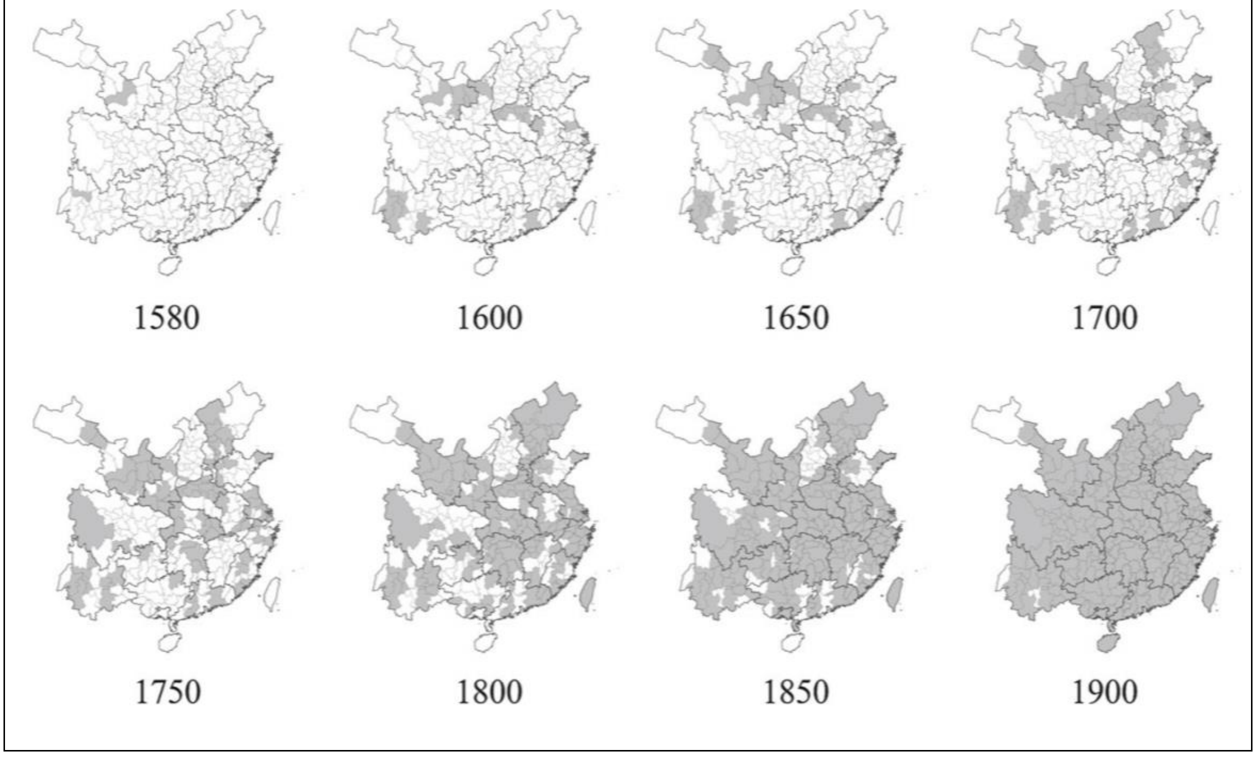


Figure 5: Foothold of Maize in China

(A) Maize Suitability Index indicates the best land for growth of maize crops.

(B) The introduction of maize agriculture into China occurred over many generations, but in a 300 year period, virtually the entire subcontinent was producing maize products for human or animal consumption.

B



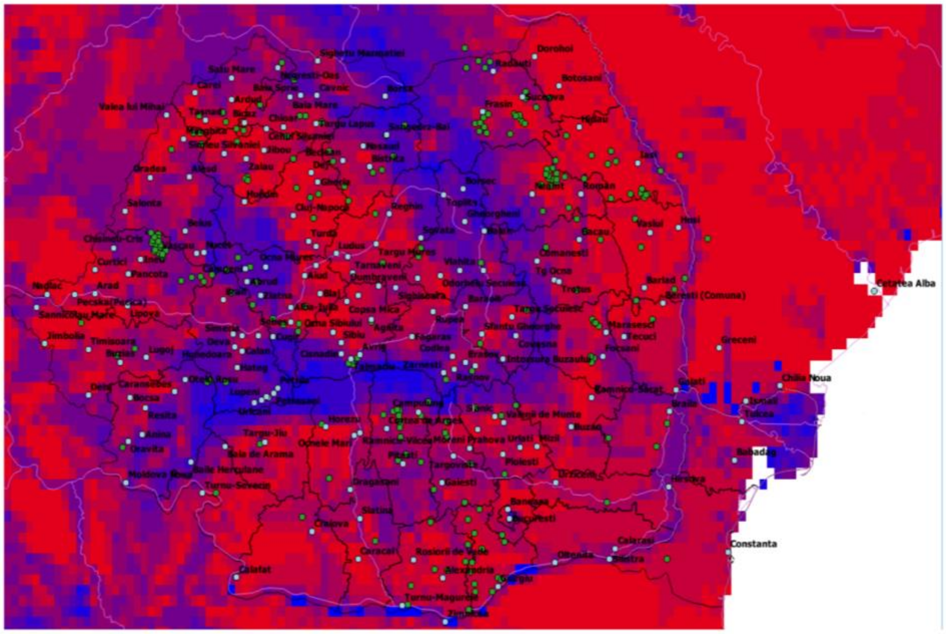
While more sustainably grown in some parts of China better than others, maize used up 55% of the available land. This massive monopoly of land could have been contributed to the taste, usability in local cuisine, and yield of maize produced compared to other new world crops.

As for most of Europe, even though millet and wheat farmers attempted to slow down the dispersion of maize, it began spreading to many countries starting from Western Europe, such as Spain, Italy, and England, and eventually reaching Eastern Europe, in particular Romania. Similar to the continents of Asia and Africa, populations in this region were trapped in the Malthusian regime. This meant, while the population was increasing, it was not leading to an increase in urbanization, causing these countries' development to stagger. However, unlike the boost in maize production in Africa and Asia, this new world grain turning into an important staple crop aided in freeing Western Europe from its shackles, even if it took 300 years after its initial introduction.[24] Romania, specifically, was the country that led this maize movement since countries in Western Europe saw it as no more useful than for livestock fodder. Since its first introduction to Transylvania, a northern region in Romania, during the 17th century, maize has become the number one harvested crop in the country. Furthermore, Romania is the leading producer of this new world cereal in all of the EU.[25] Between Romania's temperate-continental climate and a vast amount of fertile soil, the combination was a perfect mix of growing maize as it quickly replaced millet and wheat production. This maize suitability of Romania can be seen in **Figure 6A**. The red regions represent more suitable land, whereas the blue regions represent non-suitable land due to the mountain ranges depicted in **Figure 6B**. As can be seen, a large portion of the land is colored red making most of Romania suitable for maize. However, with maize being a labor-intensive crop, it has decreased urbanization in the country by 10% and reduced its potential for economic growth.[25] This evidence shows that Western Europe was able to reap the benefits of having a plentiful food source from Romania through trade and allowed for more populations to urbanize, hence increasing social and economic development in those countries.

Figure 6: Romanian Maps

(A) Cultivation of Maize in Romania. Suitable (red) land for cultivation dominates the unsuitable (blue) land in the country of Romania.[25]

(B) Geography of Romania, including predominant mountain ranges that hindered maize production and water systems that enhanced maize production.[26]



A

B

CHAPTER 4

Agrigenomics

Maize domestication and evolution have been in the process of constant change ever since its first discovery thousands of years ago in Mesoamerica. At that time, maize can be genetically traced back to when it was a simple grass-like plant known as teosinte. From then, it went through countless mutations and experienced the raths of natural selection due to changes in environment, including climate, soil type, and interactions with new chemicals, which was induced by mankind and mother nature alike. Furthermore, these changes could be categorized as natural, methods that followed the normal laws of the world. However, with our increase in technology and understanding of genetics, this natural approach has quickly shifted into a Frankenstein experiment, with the plucking and pushing of different genes to modify maize's variability artificially. This section looks to delve into the domestication of maize and how it has become the center of research for genetically modified organisms in modern time.

Firstly, it was quite astonishing for geneticists to attempt to trace maize back to its common ancestor. This cereal grain has been modified to such an extreme in the last few decades that even when experts found teosinte to be the original plant there was much debate about the matter since the ancient plant shared very little phenotypic characteristics (**Figure 7**), other than its abundant variations, to the modern grain. Nevertheless, Indigenous Americans used the gift of variation in teosinte to bring rise to the crop that is now being ingested or utilized by almost the entire human population, directly or indirectly. Furthermore, scientists have now began to question whether the domestication of maize arises from a selection of few loci with large effects, many loci with small effects over a large amount of time, or possibly a combination of two.[27]

Through the journey of tracing maize back to teosinte, numerous hypotheses have been formed in hopes of discovering the mysteries behind the cereal grain. One hypothesis is the Tripartite Hypothesis, which states that maize was domesticated from a now-extinct wild maize from South America, while teosinte was produced from a cross between maize and a grass-plant known as *Tripsacum*. Another is George Beadle's Teosinte Hypothesis, which states that artificial selection by ancient humans that created mutations with large effects could have caused teosinte to form into maize. While these and numerous other hypotheses only fueled controversy, eventually, H.G. Wilkes helped relieve some debate by creating the first thorough monograph on teosinte. This then paved the way for Hugh Iltis and John Doebley to produce a system of classification that made it possible to analyze evolutionary relationships between *Zea* taxa. Furthermore, the teosinte-maize dilemma was further mediated through the discovery of chromosome morphology and number, which revealed that the *Zea* species have 10 chromosomes, while *Tripsacum* can have either 18 or 36. These and many other analyses proved that maize (*Z. mays ssp. mays*) and teosinte (*Z. ssp. parviglumis*) were related through their genome; however, it also showed that the variations in morphologies can evolve drastically in these subspecies in a relatively short period of time.[27]

Figure 7: Teosinte (*Z. ssp. parviglumis*) and maize (*Z. mays ssp. mays*)[27]

Genetic analysis of these two plants found that they are directly linked. Teosinte (*Z. ssp. parviglumis*) (left) is the ancient ancestor of modern maize (*Z. mays ssp. mays*) (right).

As seen in this image, teosinte has 2 interleaved rows of 6-12 kernels enclosed in a hard case, while a typical ear of maize has horizontal rows of soft, uncovered kernels. These large differences have led to much debate about the credibility of these two plants being ancestors.

To further emphasize the variability of the most popular cereal grain to date, *Z. mays ssp. mays*, the population genetic theory predicts that a joint function of mutation rate and effective population size affects the level of gene selectivity, which maize satisfies both by a great margin. Furthermore, just 2 maize varieties differ in their DNA in silent sites by 1.4%, which can be equivalent to the difference between humans and chimpanzees. What predefining factors could have caused this grain to produce such variability? This again traces back to its progenitor, *Z. mays ssp. parviglumis*. Grass-type plants tend to experience large amounts of diversity, but teosinte, specifically, produced larger than normal genetic diversities coupled with the fact that maize did not experience a severe domestication bottleneck. For example, if the cereal grain did experience a genetic bottleneck, it could have lost approximately 95% of its genetic diversity like how tomatoes did during their migration from the Andes to Europe.[27]

As mentioned earlier, endless debates ravaged on between scientists on whether maize was derived from teosinte through a combination of small genetic mutations over a 10,000-year period or through just a few large meaningful mutations. Fortunately, with the use of new genetic technologies, scientists have been able to begin uncovering the mysteries behind maize's unique genome. By using quantitative trait loci (QTL) analysis, it has provided evidence that there are few regions in the genome that have been exploited and used to branch *Z. mays ssp. mays* from *Z. mays ssp. Parviglumis.* Through QTL, as few as 5 loci have been identified to largely affect maize's basic morphology. Two loci of interest that have been extensively studied are *teosinte glume architecture1 (tga1)*, a locus that controls the expression of the hard protective layer surrounding kernels, and *teosinte branched1 (tb1)*, a locus that determines the plant branch architecture.[28] The phenotypic effects of these two loci help reveal a possible association between early humans selecting plants that benefited food supply rather than them being naturally selected since these mutations provide poor survivability for the crop. However, it is important to note that QTL analysis has advanced. Geneticists have now discovered more regions that contribute to the morphology of domesticated maize, which brings back the notion that small numerous genetic mutations are still in play. With this new information, there seems to be a correlation forming where large significant genes were selected by humans for specific needs such as *tga1* and *tb1*, while smaller less significant genes were naturally occurring over time to help increase crop yield and assist in the rapid adaptation of local environments.[27]

CHAPTER 5

Conclusion

The story of a simple grain has managed to breach through history, genetics, and cultural identities. The domestication of maize, whether by genetic accident or destiny, is a long process that seems to have taken hold of our world. In our current society, especially in the United States, corn is etched into almost everything we produce so that we would be ruined if the crop spontaneously disappeared. I would not say it is as important as the air we breathe, but it is constantly chiseling its way to that point. Even with other New World crops being favored around the world and many countries reducing corn product consumption, the crop has refused to stop its spread in conquering our world.

This paper has aimed to reveal and compile the journey of maize and give us a better understanding of how it has managed to reach its current standing. It is fascinating to analyze the different aspects that needed to come together to produce such a worldwide successful crop. Furthermore, the recollection of the impact of corn on my life through mămăligă looks to provide a more personal approach in understanding how a simple grain can affect an individual within any society. I aim to further my research into maize by branching out to the implications it may have on human health. With the rapid increase of corn products, that are excessively genetically modified, the health effects still have not been fully studied and could lead to avenues of clinical research in the coming years.

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