Improving Population Nutrition by Enhancing the Nutrient Content of Staple Crops: Wheat, Corn, Potatoes, and Rice

Foundation for Innovation in Healthy Food

August 2026

Preamble

This report was convened by the Foundation for Innovation in Healthy Food, as a first step towards systematically identifying the most promising concepts towards continuous improvement of commodity foods. The focus of this process is to identify 'mainstreamable' traits. These traits or processing changes lead to no material cost increase to the farmer or others across the supply chain; and they maintain consumers' enjoyment of their foods without food reformulation or major processing changes. At scale, the most attractive options provide material public health benefits for people of all socioeconomic groups globally.

Acknowledgements

Special thank you to Daniel Gallaher, Stanley O. Samonte, and P. Stephen Baenziger for playing key roles in drafting the report. Also thank you to the team developing the concepts and their reporting, including: Katie Brown, Brett Carver, Kirsty Crews, David Douches, Mike Gore, Aymeric Goyer, Mary Guttieri, David Holding, Alison Lovegrove, Seth Murray, Roy Navarre, Shannon Pinson, Peter Shewry, Brad Seabourn, Sunish Sehgal, Endang Septiningsih, Ed Souza, Tadesse Teferra, Isabel Vales, Rod Wallace, Jennifer Yates, Xiaofei Zhang and other scientists who provided insights into this report.

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1. Introduction

The human population is beset by numerous deficiencies of essential nutrients, resulting in billions of people experiencing preventable health maladies that increase morbidity and mortality. Beyond the personal hardships these deficiencies can impose, there is also a significant drain on public resources, both direct and indirect, that countries experience in grappling with their societal impact. Preventing or overcoming deficiencies of essential nutrients is straightforward at its most basic level; we simply need to consume greater amounts of the nutrient(s). Obviously, however, the reality is quite complex. Poverty, conflict, cultural practices, and lack of education, among other factors, result in a failure to consume adequate levels of essential nutrients, leading to debilitating deficiency diseases. Further, food contains many compounds that do not fit the classic definition of an essential nutrient but that are almost certainly highly beneficial to health, such as dietary fiber, polyphenolics, flavonoids, and glucosinolates. Although there is great uncertainty about the optimal intake of these compounds, their consumption is certainly greatly reduced in Westernized cultures from that of more traditional cultures, and this reduced intake has been implicated as an important factor in the greater prevalence of many chronic diseases in Western societies.

There have been notable successes in reducing nutritional deficiencies within populations. Fortification of salt with iodine has reduced goiter and enrichment of refined wheat flour with folic acid has reduced neural tube defects. However, efforts to improve nutritional status by persuading people to change their diets have been largely unsuccessful. The National Cancer Institute/FDA 5-A-Day campaign to increase vegetable consumption to reduce cancer risk was not successful, nor have efforts promoting dietary fiber intake resulted in a meaningful increase in consumption of dietary fiber. A major lesson learned from these efforts is that increased awareness of a benefit does not translate into behavior change. A way to circumvent this lack of behavior change is to modify a plant, either by traditional breeding or by genomic approaches, such that the plant either produces or incorporates a greater amount of a nutrient, a process known as biofortification. This differs from traditional fortification, in which a nutrient is simply added to the food.

Recently, Wallace et al. have advocated for a public health approach to improving nutritional status by biofortification of staple foods.[1] They refer to this approach as HEAL, for Health Embedded Agriculture and Logistics. These authors argue that by making nutritional improvements in the four most consumed plant commodities – wheat, maize, potatoes, and rice – significant decreases in several prevalent chronic diseases can be achieved.

The objective of this report is to briefly describe these four commodities and then discuss examples of possible biofortification targets for each.

1.1 Wheat

Wheat can be categorized into classes. There are red wheats and white wheats. Each of these exists as hard or soft wheat. It is this latter characteristic that primarily determines what wheat is used for. Hard wheats are used for bread making, whereas soft wheats are used for cakes, cookies, and pastries. A final wheat category, durum, is used exclusively for pasta products.

Wheat kernels can be milled into three major fractions – a starchy endosperm (80-85%), a fiber-rich bran (12-17%), and a nutrient-rich germ (the embryo) (3%). Refined flour is almost entirely the endosperm fraction, which contains low concentrations of essential nutrients and dietary fiber. Consequently, in the United States, refined flour is enriched with four vitamins (thiamin, riboflavin, niacin, and folic acid) and one mineral (iron).

Wheat provides about 20% of food calories worldwide and therefore is an obvious choice for modification to increase nutrient intake.

1.1.1 Targets for Biofortification

Iron and Zinc

Iron deficiency is the most common essential nutrient deficiency on the planet and represents a major public health problem. Iron deficiency results in severe fatigue, resulting in a dramatically reduced work capacity, and iron deficiency anemia is a risk factor for death in patients with chronic heart failure,[2] cognitive impairments,[3] and depression.[4] Zinc deficiency is likely also very common, perhaps almost as common as iron deficiency. Zinc deficiency leads to increased birth defects, reduced stature and delayed sexual development in children, and impaired immune responses.[5]

Since about 1960, there has been a decrease in the concentration of iron, zinc, copper, and magnesium in wheat that has correlated with large increases in grain yield that began at that time,[6] a phenomenon referred to as the dilution effect. Traditional breeding approaches may be able to increase iron and zinc content of wheat kernels, although increasing the concentration of these minerals in the endosperm has proven difficult.[7] However, genomic approaches show promise.[8]

Magnesium

Magnesium is an essential macronutrient that is the fourth most abundant element in the body and the second most abundant intracellular cation. It is widely distributed in the body, being found in high concentrations in bone, muscle, and soft tissues. Magnesium is necessary for maintaining bone health, cardiovascular integrity, and normal neurological functioning. Even modest magnesium deficiency can result in lethargy, muscle cramps, muscular irritability, and electrocardiogram abnormalities.[9]

Unfortunately, there are no reliable biochemical methods to determine marginal magnesium deficiency. However, dietary data indicate that half the population of the United States has a magnesium intake below the Estimated Average Requirement.[10] A modelling study suggests that a third of the global population may be deficient in magnesium.[11] Thus, magnesium is clearly a nutrient of concern and an important target to consider for biofortification.

Magnesium is present in the wheat kernel (and other cereal kernels) largely bound to phytic acid in structures called globoids.[12] Globoids are in the bran and germ of the kernel. It is likely that the amount of magnesium in the globoids can be increased, either by traditional breeding or by genetic methods. This could increase the magnesium content of wheat products using flour that contains some bran and germ (i.e., whole wheat or high-extraction flour). However, in refined flour, in which essentially all the bran and germ is removed, this strategy would be ineffective.

Although the amount of magnesium in the endosperm is low, it is not zero. It may be possible to increase the number or activity of transporters that move magnesium into the endosperm. Increasing the magnesium content of the endosperm even modestly could be impactful, given the large amount of endosperm in the kernel, relative to the amount of bran and germ. Whether to focus on increasing magnesium in the endosperm or in the bran and germ (via globoid modification) would depend on the way the wheat would likely be consumed.

Dietary Fiber, including Indigestible Oligosaccharides

Greater dietary fiber intake has been associated with lower chronic disease mortality.[13] In the United States people consume far less than the recommended amount of dietary fiber.[14] Although the wheat kernel is a good source of fiber, it is concentrated in the bran layer, and refined flour contains little dietary fiber. Increasing the dietary fiber content of the endosperm could significantly increase the population fiber intake. This may also improve the gut microbiome, as wheat-containing diets have been shown to change large intestinal bacterial abundance in a positive way in animal models.[15]

There are two dietary fiber targets of particular interest. One is arabinoxylans, which are the major dietary fiber in cereals. Most of the arabinoxylans in wheat are in the bran layer and thus refined flour would have a relatively low concentration. However, because refined flour contains a small proportion of bran, increasing the total arabinoxylan content of wheat will also increase arabinoxylans in the endosperm, thus increasing fiber intake.

Fructans, which are fructosyl polymers, are another fiber type of interest in wheat. Fructans have been shown to have a prebiotic effect; that is, they promote the growth of beneficial bacteria in the gut and provide a health benefit to the consumer.[16] Although fructans are in greatest concentrations in the bran, white and whole wheat breads contain about the same concentration of fructans.[17] Thus, similar to the situation with arabinoxylans, increasing the fructan content of wheat would increase the fructan content of bread, even white bread, and thus increase prebiotic fiber intake.

Bioactive Phytochemicals

Wheat contains many compounds that are suggested to have beneficial health effects.[18] These include alk(en)ylresorcinols, phytosterols, sphingolipids, lignans, phenolic acids, carotenoids, and flavonoids, among others.[19,20] These compounds are concentrated in the germ or bran layer of the wheat kernel. The potential health benefits have almost always been demonstrated using either purified compounds or extracts of the bran, that are then fed at high concentrations unattainable in a normal diet or examined only in cell culture. This has created uncertainty about their physiological importance. An additional challenge is that some of these compounds are chemically bound within the bran or germ in ways that resist digestion, resulting in very low bioavailability.

With these caveats in mind, there are several groups of phytochemicals that may be attractive targets for biofortification. Phytosterols are relatively abundant in wheat and are well-established to have cholesterol-lowering effects in humans.[21] Further, and importantly, phytosterols act within the small intestinal lumen to inhibit cholesterol absorption.[22] That is, they act without being absorbed, thus bypassing the issue of bioavailability. Finally, as noted by Shewry and Hey,[23] phytosterols in wheat show high heritability (>50%) and thus would be amenable to being increased by breeding.

Another group of phytochemicals of interest are flavonoids, which are abundant in wheat and are notable for existing almost completely in an unbound state.[24] Importantly, the concentration of free flavonoids was shown to be equal in the refined flour and whole grain flour.[25] However, whether the concentration of flavonoids is sufficiently high in wheat to impart physiological benefits remains unclear.[26] Preliminary work to address this concern would seem prudent.

1.2 Corn (Maize)

Most maize in the United States is used as animal feed or for ethanol production, with only about 12% used for food, seed, or industrial uses.[27] However, the proportion of the maize crop consumed as food varies widely across the globe, being highest in Sub-Saharan Africa, where it exceeds 50%, and followed by Central America and the Caribbean.[28] The high degree of human consumption of maize in these regions makes it an attractive target for biofortification.

The maize kernel consists of about 82% endosperm, 12% germ, 5% pericarp, and 1% tip cap (the part of the kernel attached to the cob). The maize endosperm is mostly starch, as is the case with most cereals. The dietary fiber content of fresh sweet corn is approximately 2 to 2.4 g per 100 g. However, the dried maize grain has a fiber content of about 7.3 g per 100 g, most of which is insoluble fiber.

1.2.1 Targets for Biofortification

Iron and Zinc

As discussed in the section on wheat, iron and zinc are important targets for biofortification due to widespread global deficiencies of these two minerals. Increasing the iron and zinc content of maize would be desirable, and maize with a greater zinc concentration has been developed by HarvestPlus.[29] However, an issue that is present for all cereals, but more so for maize, is the presence of phytate, or inositol hexaphosphate. The phosphate groups of phytate will bind (chelate) metals, particularly iron and zinc, preventing their absorption. Maize is particularly high in phytate, typically higher than wheat and much higher than rice.

It will likely be quite challenging to develop maize varieties with concentrations of iron and zinc sufficiently high to overcome the absorption inhibiting effects of phytate. However, it appears possible to incorporate a gene for phytase, the enzyme that dephosphorylates phytate and reduces its mineral binding capacity, into a cereal. This has been with barley [30] and maize.[31] Further, iron absorption was greater in a tortilla made from genetically engineered low phytate maize compared to normal maize,[32] indicating the feasibility of such an approach.

Most corn is used as animal feed. Therefore, an attractive biofortification target would be to modify corn such that when the corn is fed to animals, the animal products have a greater health benefit. Conjugated linoleic acid (CLA) is a family of polyunsaturated fatty acids that occur naturally in milk and have been found to reduce inflammation, obesity, and cancer risk in animal models, with some evidence that they also do so in humans.[33] The quantity of CLA in milk can be increased by increasing certain fatty acids in the cow's diet, specifically linoleic acid, linolenic acid, and the so-called fish oils. Thus, increasing the linoleic acid content of corn fed to dairy cows will increase the CLA content of the milk. However, corn oil is already rather high in linoleic acid (54%), so it is not clear that an increase to the level found in sunflower oil (70%) would be impactful. A somewhat different approach would be to modify the corn plant to produce trans-vaccenic acid, which is an immediate precursor to CLA.[34] This would likely greatly increase CLA production in the milk.[35]

1.3 Potatoes

Potatoes, which are indigenous to the Andean highlands, are grown worldwide, with China and India as major sites of production. Potatoes are a tuber and therefore differ fundamentally from cereal crops such as wheat, corn, and rice. They are considered the world's most important non-cereal crop.

Starch is the predominant carbohydrate in potatoes, as it is in cereals. Potato starch is 70-80% amylopectin, with the remaining amount as amylose. Dietary fiber is concentrated in the peel, but there is a notable amount of fiber in peeled potatoes (flesh), much more than in white rice for example. Although the concentration of trace minerals is greater in the peel than the flesh, with the notable exception of iron, the far greater amount of flesh than peel leads to the flesh actually providing more of the trace minerals.[36]

1.3.1 Targets for Biofortification

Glycemic Index (GI)

The GI is a value that indicates to what degree different food carbohydrates increase blood glucose concentrations relative to a standard such as glucose or white bread. A high GI indicates a more rapid digestion and absorption of a dietary carbohydrate. While the validity of the GI as an indicator of disease risk has been debated, there is now strong evidence indicating that a high GI is associated with an increased risk of several diseases, including type 2 diabetes and heart disease.[37]

Baked potatoes have often been described as being like candy, due to their very high GI value. Although potatoes are obviously more nutritious than candy bars, this does point to the GI of potatoes as a clear target for biofortification. Increasing the amylose content would seem to be the most direct way to accomplish this. Although some high amylose varieties have been identified,[38] there does not seem to have been a systematic effort to determine the GI in these or other high amylose varieties. Regardless of the approach, creating high amylose potato varieties with acceptable agronomic and sensory characteristics would seem highly desirable.

Protein

Several factors suggest that increasing the protein concentration of potatoes may be a good biofortification target. First, potato protein is high quality, one of the highest quality plant proteins.[39] Notably, it is high in lysine, the limiting amino acid in cereals. Second, it appears to be an allergy-free protein in humans.[40] Finally, work has been published identifying a large number of single nucleotide polymorphisms (SNPs) associated with the protein and amino acid content of potatoes,[41] which should aid in developing higher protein cultivars of potatoes.

1.4 Rice

Rice, like wheat, and unlike corn, is overwhelmingly grown for human consumption. It is a major commodity crop, as rice consumption accounts for almost one-fifth of global energy intake. Rice consumption is greatest in China, India and Southeast Asia, followed by Brazil and Nigeria.

Rice, like other cereals, is composed of a starchy endosperm (87-90%), a bran layer (8-11%), and a germ (2-3%). Milling removes the bran and germ layers, yielding white rice, which accounts for approximately 90% of rice consumption. White rice contains approximately 85-90% digestible carbohydrate (essentially all starch), 7-10% protein, and only 1-2% dietary fiber, as well as traces of vitamins and minerals.

1.4.1 Targets for Biofortification

GI

While rice has traditionally been considered a high GI food, a recent review of clinical trials feeding high amylose rice suggests that this type of rice has a lower GI value.[42] Given the huge number of rice consumers across the world, developing high amylose rice varieties that are high yielding and well accepted by consumers shows considerable promise for improving health.

Iron and Zinc

As discussed in the sections for wheat and corn, increasing intake of iron and zinc is of great importance. White rice has a low concentration of these essential trace minerals, which are concentrated in the bran and germ. White rice also has little phytate, as this anti-nutrient is mostly present in the aleurone layer, part of the bran.

A high zinc biofortified rice has been developed and the zinc within it was found to be well absorbed in children.[43] A worthwhile goal would be to increase the zinc content of the biofortified rice. A second goal would be creation of a high iron biofortified rice. In both cases, testing functional outcomes would be a priority, for example, increased growth in children in the case of zinc biofortified rice and increased hemoglobin concentrations in the case of iron biofortified rice.

1.5 Toxic Metals

Agricultural soils are under threat of accumulation of toxic metals due to pollution by human industrial activities. These metals, primarily arsenic (As), cadmium (Cd), lead (Pb), and mercury (Hg), sometimes also called heavy metals, pose serious health risks to humans consuming grains from plants grown in metal-contaminated soils. Further, toxic metals can decrease the yield of the crops themselves. Thus, creating cereal crops and potatoes that would incorporate less of these toxic metals from the soil would be of great benefit, both to consumers and to farmers.

An obvious approach to reducing the content of toxic metals in the grains or potatoes is to reduce or delete the transporters of the toxic metals, such that less of these metals are taken up into the plant. However, toxic metal transporters commonly cotransport essential minerals such as iron, zinc, manganese, and phosphate. For example, arsenic and phosphate are chemically similar and therefore it is difficult for plant transporters to distinguish them. However, there is evidence that differences in expression of toxic metal transporters in the roots correlate with differences in the accumulation of the toxic metal in the cereal grain.[44] Thus, there are opportunities to reduce toxic metal uptake by reducing the metal transporters, or the affinity of toxic metals for the transporters, and thereby reduce the toxic metal content of the grains or potatoes. This could be done by traditional breeding techniques or by using molecular biology approaches.

Another approach to reducing the toxic metal content of grains or potatoes from plants grown in contaminated soil is by increasing the sequestration of the metals in the plant vacuoles.[45] This would prevent the transport of the toxic metal within the plant and limit its incorporation into the grain or potato. The multistep toxic metal detoxification pathway is quite well understood [46] and offers multiple points where interventions could be explored. For example, it has been demonstrated that overexpression of a transporter involved in sequestration (OsHMA3) reduced cadmium accumulation in the rice kernel.[47]

Given the continued environmental contamination of agricultural soils, and the devastating impact on human and animal health of toxic metal intake, developing cereal crops and potatoes that accumulate less toxic metals would be a major public health success.

2. Next Possible Applications for Improved Nutritional Qualities in Wheat

2.1 Background

Wheat (*Triticum spp*.) is frequently evaluated or classified according to growing season (winter, facultative, spring), kernel color (red or white), kernel hardness (hard or soft), protein level (high [~16%] to low [~8%]), and grain volume weight which relate to their food uses.

Bread and Pasta Wheat

The two main commercial wheat types are "bread" or common wheat (*Triticum aestivum* L.) and "pasta" or durum (*Triticum durum* L.) wheat.

Winter vs Spring Wheat

Winter wheat is grown over the winter and requires vernalization (a cold period) to produce grain. A facultative wheat is also grown over the winter in mild climates so there is rarely winterkilling (e.g. California and Arizona or some European countries). Spring wheat is grown in the spring to summer in regions with harsh winters that would otherwise kill winter wheat.

Wheat Color

White kernel types are generally preferred because the milling extraction is higher (you can mill closer to the outer layers without red flecks often assumed to be insect contamination). Red kernel types are grown in wetter climates at harvest to reduce pre-harvest sprouting (seed germinating in the head before harvest). The red color is from tannins in the testa layer which may be responsible for the reduced pre-harvest sprouting. Hence white wheat tends to be grown in drier climates.

Hard vs Soft Wheat and Strong vs Weak Dough

Hard wheat differs from soft wheat by their physical hardness. The major genes controlling hardness on chromosome 5 D encode two puroindoline proteins (*Pina* and *Pinb*), with soft being the wild type and hard wheat resulting from mutations in one or both genes. The *Pin* genes appear to act by determining how the gluten protein matrix adheres to starch granules, affecting release during milling. In hard wheats, the protein matrix adheres tightly to the starch granule, while in soft wheats there is less adherence. Hence, the granules are more readily released from soft wheats with less damage. Hard wheats are preferred for breadmaking because the damaged starch results in higher water absorption. The gluten proteins present in flour form a continuous matrix in dough which confers a unique combination of properties: elasticity (strength) and extensibility. In broad, high protein wheats which form strong doughs are required for breadmaking and low protein wheats which form weaker more extensible doughs for cakes and cookies. Crackers are made from higher protein soft wheat or blends of hard and soft wheat. Durum wheat is very hard (because it lacks *Pin* genes) and is used to make the semolina flour used in pasta (such as spaghetti and couscous). It should be noted that in countries with little durum wheat production, noodles were often made from bread wheat or other cereals (e.g. rice noodles). In some European literature, durum wheat was described as hard wheat and all bread wheats (both hard and soft, currently) were considered soft wheat. In Europe the two major classes are hard wheats with high protein and strong gluten for breadmaking and low protein wheats, which may be either hard or soft, for livestock and poultry feed. Some soft wheats with lower protein and more extensible gluten are also used for cakes and cookies.

Grain Volume

Grain volume weight (also called specific weight) is also used for purchasing wheat with the standard set at 74.83 kg/hl (60 lbs/bu). Lower grain volume weights are often considered less desirable for milling (lower milling extractions); however, grain volume weight is a function of how kernels pack into a unit volume and can be misleading for predicting milling extractions.

Commercial Classes

The common commercial classes of wheat are (based upon the USDA grain standards): For bread wheat: Hard Red Winter Wheat, Hard Red Spring Wheat, Soft Red Winter Wheat, Soft White Wheat, and Hard White Wheat. For pasta wheat: Durum Wheat.

In the USDA grading system, there are two other classes of wheat; namely unclassed wheat (any wheat that is not classifiable into the above classifications) and mixed wheat (a mixture of wheat that contains less than 90 percent of one class of wheat and more than 10 percent of another class or classes; for example, 85% hard red spring wheat and 15% hard white wheat).

Components of the Wheat Kernel (Bran, Germ, and Endosperm)

The wheat kernel can be separated by milling into three fractions: the bran (outer layer and aleurone), the germ (the embryo), and the starchy endosperm which gives white flour. The bran and germ may be sold separately in health foods or as livestock feed.

Refined Flour and Whole Grain Processing and Composition

Refined flour is roughly 86% digestible carbohydrates; 10% protein; and 4% dietary fiber. In whole grain flour, the kernel is often milled as in refined flour, but the germ and bran are added back to the flour in the exact percentage of kernel. In smaller milling operations, the whole kernel can be milled without separation. The bran and germ are higher in protein, vitamins, lipids (which can lead to issues of rancidity), and dietary fiber. For example, 12% protein grain will yield a refined flour that is usually 1 to 1.5% lower in protein (10.5 to 11% protein). However, the protein present in the germ is less functional in making bread as it lacks gluten. As mentioned earlier, red kernels include tannin in their testa layer, so whole grain flour from red wheat often has an astringent taste compared to refined or whole grain flour from white wheat. An important difference between refined flour and whole grain flour is that refined flour is often fortified by adding nutritionally important B vitamins (like folic acid to prevent birth defects) and minerals, whereas 100% whole wheat cannot contain fortifying or other additives. As such, many nutritionists recommend blends of refined flour with whole grain flour to optimize the benefits of both flours. Currently refined flour is about 83% of the flour market in the USA.

Wheat Consumption by Humans, Animals

Approximately 70% of all wheat produced is used for human consumption, 20% for animal feed, and 10% for other uses (e.g. seed, ethanol production). Additional information about USA consumption can be found at https://www.ers.usda.gov/topics/crops/wheat/wheat-sector-at-a-glance and for the global wheat sector at: https://link.springer.com/chapter/10.1007/978-3-030-90673-3_4. For human nutrition, wheat provides approximately 20% of the calories, 20% of the protein, and is a major source of dietary fiber and other nutrients.[23] Serbia and Central and West Asia have the largest per capita wheat consumption while Guinea-Bissau and other sub-Saharan countries in Africa or rice-based food systems in Asia have the lowest per capita wheat consumption.[48]

2.2 Science-Based Nutrition Targets for Human Consumption

The targets are broadly grouped by the trait they are affecting.

2.2.1 Fiber

Fiber related traits to build upon the ongoing work on increasing arabinoxylan dietary fiber (Coalition for Grain Fiber) and also the existing product of high amylose wheat (Health Sense® high fiber wheat). It is currently estimated globally that 80% of the people (6.2 billion) do not consume the recommended daily amount of fiber. In the USA, it is estimated that 95% of the people do not consume the recommended daily amount of fiber.

Elevated Fructan Target

Elevated fructan composition with appropriate polymerization (prebiotic activity) or reduction thereof to conciliate fructan intolerance (1 in 15) which is part of the FODMAP (*Fermentable Oligosaccharides, Disaccharides, Monosaccharides, and Polyols*) diet. These are short-chain carbohydrates that the small intestine struggles to absorb, often causing bloating, gas, and digestive distress in people with Irritable Bowel Syndrome (IBS). Approach: Bi-selection (meaning selection for increased or decreased fructan levels) of white flour or wholemeal fructan concentration through breeding.

Increased Inulin Target

Increased inulin, a soluble fiber and polymer of fructose (a fructan) which is sometimes used as a bread additive. Natural inulin in wheat is primarily found in the aleurone layer (part of the bran) which means that its level is highest in whole wheat flour and greatly reduced during milling to make refined flour. Approach: Look for natural variation supplemented by EMS mutation of known genes (6FST and 1FEH) affecting inulin concentration. These mutations and natural variation would be used in breeding programs to develop higher inulin (fiber) lines.

Increased Beta Glucan Target

Increasing beta glucan to increase dietary fiber. Approach: Increase dietary fiber through natural variation and selective breeding. Expected to have a smaller impact than with arabinoxylans.

Increased Amylose Target

High to Medium levels of amylose. Approach: Because amylopectin and amylose are the major types of starch in wheat endosperm, it may be possible to select lines through breeding with higher levels of amylose that have superior milling and baking characteristics. Starch is the major component of wheat flour, and it is expected that major changes in its composition may have major effects on milling and baking, as well as grain yield.

Improved Sensory Bran Target

Bran hydrolysates. Approach: Processing bran into hydrolysates and adding back to flour to increase dietary fiber intake with improved sensory acceptance. Basically, making a higher fiber "bran" flour. Approach: Using chemical, heat and enzymatic hydrolysis of bran under different pretreatments to form the hydrolysates which would be added back to the flour.

2.2.2 Bioactive Components

Could not find an estimate of the number of diets that are deficient in bioactive compounds, but highly/ultra processed food is low and the Mediterranean and plant- based diets tend to be high in bioactive components.

Increased Phenolics-Anthocyanins in Whole Grains Target

Increased phenolics-anthocyanins in whole grains. Approach: Using natural variation for pigmentation (e.g. blue or purple seed color including introgressions from wheat relatives) to increase the phenolics-anthocyanins in the bran or wheat whole grain flour (kernel color is usually determined by the bran layer). It will not affect refined flour as the bran is milled off. This approach is genetically independent of dietary fiber, hence it can be combined with increased fiber wheats in whole grain products.

2.2.3 Protein and Lipids

It is estimated 12-14% of the world does not consume enough protein (~1 billion people). Lysine and tryptophan (essential amino acid) tends to be deficient in cereal-based diets.[49] In addition, some wheat proteins may have deleterious effects in those with gluten sensitivity (~10%) or celiac disease (~1%). Lipid is a diverse class of molecules with specific activities. For example, it is estimated that most of the world does consume enough omega-3 fatty acids (76%) or polyunsaturated fatty acids (70%), but consumes sufficient total fat.

Higher Lysine Target

Like more cereal grains, wheat is limited in the essential amino acid (those amino acids that humans cannot synthesize and must be consumed) lysine. By increasing lysine in wheat, the protein would become more balanced and available for human use. The most recent successful example of elevate lysine level is in quality protein maize. Approach: Germplasm screening and conventional breeding. It is expected that this approach will have to be supplemented by biotechnology because in the 1970s and 1980s there was a largely unsuccessful germplasm screeding and breeding effort to increase lysine.

Reduced 33mer Protein in Gliadins Target

The 33-mer peptide is a 33-amino-acid fragment derived from alpha2-gliadin, a protein found in wheat gluten. It is the most potent immunogenic sequence known to trigger celiac disease, characterized by its extreme resistance to gastric and pancreatic digestive enzymes. Key Characteristics: Structure & Sequence: The 33-mer peptide (sequence: LQLQPFPQPQLPYPQPQLPYPQPQLPYPQPQPF) contains six overlapping copies of three distinct T-cell epitopes. Toxicity Mechanism: Because it resists proteolytic cleavage, the peptide passes through the gut epithelium, stimulates T-cell immune responses, and can form supramolecular oligomers that increase gut permeability. Approach: germplasm screening, mutations, and selective breeding supplemented by biotechnology.[50] Note gliadins and glutenins are multi-gene families, so while reduction is possible, elimination would be more difficult.

2.2.4 Vitamins and Minerals

Globally, it is estimated that large numbers of people are deficient for iodine (68%), vitamin E (67%), vitamin C (66%), iron (17% to as high as 4 billion people),[51] Zn (17%), vitamin A (7%. Figures usually are given by affected populations, e.g. 30-33% of children 6 to 59 months or 190 million children, 19 million pregnant women), and folic acid (syn. folate; 54%).

Increased Iron and Zinc Target

Increasing mineral content of wheat flour for minerals that most of humanity is deficient in (e.g. Fe and Zn). Approach: Germplasm screeding and selective breeding. HarvestPlus has major programs in this area which could be extended further.[52]

Increased Magnesium (Mg) Target

Mg is among the most widely under-consumed nutrients — roughly half of U.S. adults fall below the Estimated Average Requirement — partly because it is found in the bran and germ, hence milling removes up to about 80% from the whole grain. Refined flour has ~20–25 mg per 100 g compared to ~137 mg in whole-grain flour and Mg is not added back at enrichment. In a dose-response meta-analysis of 40 cohorts and over a million participants, each extra 100 mg per day of magnesium was associated with a 7% lower risk of stroke, 22% lower heart failure, 19% lower type 2 diabetes, and 10% lower all-cause mortality (though not with total cardiovascular or coronary heart disease, and the associations are observational, so less certain than for potassium salt). Approach: Grain Mg can be improved by breeding with parental lines having major genes for Mg accumulation. Mg content varies widely between genotypes and the first QTLs have been mapped [52] — but it is polygenic and environmentally sensitive (low heritability)[53] in some wheat gene pools. It could be stacked with other nutritional traits like higher fiber.

Increasing pro-Vitamin A Target

Increased pro-vitamin A. Like rice-based diets, many wheat-based diets are deficient in pro-vitamin A. Approach: Use EMS mutations or biotechnology to increase the variation of pro-vitamin A in wheat followed by conventional breeding to create higher levels of pro-vitamin A in wheat cultivars. This approach would be similar to “golden rice”.

Increased Folic Acid Target

Increase folic acid in wheat to reduce birth defects. In the developed world, folic acid is commonly added to enriched wheat flour, but it is less common in the developing world and cannot be added to 100% whole wheat flour. Approach: Germplasm screening and selective breeding supplemented by biotechnology.[54]

2.2.5 Reducing Deleterious Compounds

Cadmium is a toxic heavy metal with no known beneficial effect in plants including wheat or humans. Cadmium is taken up from the soil and can be found naturally in some regions or as the result of industrial pollution. Twenty-seven % of cadmium dietary exposure is from consuming grains and vegetables. Phytate (the salt of phytic acid) is a phosphate rich chelator of beneficial minerals (Fe, Ca, Zn) that is largely indigestible. Hence it reduces beneficial minerals and phosphorous in the human diet (see above comments on mineral deficiencies). Phosphorous deficiency is rare in humans and most the effects of phytate are due the reduction of beneficial minerals and phosphate pollution from human and animal waste. Acrylamide is a compound formed during cooking (baking or frying) from free asparagine chemically reacting with reducing sugars. Acrylamide, at very high doses, can cause cancer in animals, hence there is a concern that lower doses may harm humans.[55]

Reduced Cadmium Target

Reducing Cadmium (Cd), a known toxic heavy metal. Approach: Use known genes in bread wheat to reduce Cd uptake and transport within the plant. This approach was successfully used in durum wheat, though it may be the durum alleles are better at reducing Cd than those in common wheat.[56]

Reduced Phytate Target

Reducing phytate. Approach: Use natural variation and mutations followed by selective breeding to reduce phytate concentrations in wheat.[57]

Reduced Acrylamide Target

Reducing acrylamide in cooked wheat products. By reducing free asparagine or reducing sugars in wheat flour, you can reduce acrylamide in the cooked products.[58] This work is very well advanced in the UK by gene editing and mutations where acrylamide was greatly reduced in baked products.[59] Approach: Free asparagine is controlled by genetic and environmental factors including sulfur deficiencies and other stresses like salt or drought. Hence free asparagine can be reduced by germplasm screening, gene editing, and mutations [59] followed by selective breeding, by how wheat is grown and fertilized, or a combination of both.

3. Next Possible Applications for Improved Nutritional Qualities in Maize (Corn)

3.1 Background

Maize (syn. corn, *Zea mays* L.) is classified as being vegetable maize (harvested before maturity; e.g. sweet corn, but sometimes immature grain maize) or grain maize (maize harvested at maturity, e.g. dent and flint maize, or popcorn corn).

Growing and Breeding Overview

Maize is a summer annual crop that is diploid (2n=2x=20) and cross pollinated which lends itself to open-pollinated populations (usually grown in the developing world where farmers save seed) or controlled pollination hybrids (grown in the developed and developing world where farmers can afford hybrid seed and purchase seed every year). Modern hybrids are single crosses between two inbred lines and the yield advantage and uniformity of the product more than compensate the grower for the higher cost of commercially bred seed.

Kernel Biology

Considering the grain maize kernel, approximately 82% is endosperm, 12% is germ, 5% is pericarp, and 1% is the tip cap (the kernel part that attaches to the cob). The largest portion of the kernel is carbohydrates (72-73%), protein (8-10%), lipids (4-5%), and fiber (2-3%). Historically, grain maize carbohydrate is about 25% amylose and 75% amylopectin starch. However, by genetic manipulation waxy maize (99-100% amylopectin) and high-amylose maize (between 50% - 90% amylose) have been developed and grown.

Commodity Classification by Color and Other

Grain maize is classified mainly by its kernel color (e.g. yellow maize [not more than 5% maize of other colors], white maize [no more than 2% maize of other colors] and mixed maize [maize that does not meet the yellow or white classification]). The USDA grain standards can be found at: https://www.ams.usda.gov/sites/default/files/media/CornStandards.pdf. The kernel color is important, as is the processing for the type of product that maize is made into.

Nutrition Overview

As mentioned earlier, maize can be separated into sweet corn or dent maize that was harvested before maturity and eaten directly by humans. An ear of roasted sweet corn has approximately 1 gram of fat, 17 grams of carbohydrate, 2.4 grams of fiber, 2.9 grams of sugar, and 2.9 grams of protein. It also contains 10% of the daily Vitamin C and 8% of the daily magnesium requirement.[60]

The other major type of maize is popcorn which is a flint maize and is prized for its ability to "pop" or explode at high temperatures and is consumed as a snack food. "Air-popped popcorn (no salt or other additives) is 4% water, 78% carbohydrates (including 15% dietary fiber), 12% protein, and 4% fat. In a 100 gram reference amount, popcorn provides 382 calories and is a rich source (20% or more of the Daily Value, DV) of riboflavin (25% DV) and several dietary minerals, particularly manganese, phosphorus, and zinc (36–45% DV). B vitamins and other minerals are in appreciable amounts".[61]

Food and Non-Food Applications

Yellow maize is used for livestock feed, refined into ethanol, maize oil, biodegradable starches, and sweeteners (high fructose corn syrup). In the USA, approximately 43% is used for animal feed, 44% for ethanol production, and the remaining 13% for seed and other uses.[27]

Grain maize production (99% or 81 million acres), dwarfs the production of sweet corn (600,000 acres) and popcorn (200,000 acres). To give another perspective on the size of these acres, silage maize (for animal feed) is planted on approximately 6 million acres. When dry milled, grain maize is used for cornmeal (cornbread, hush puppies, coatings, and sometimes polenta—can be made with yellow or white corn meal) and for corn flour. White maize is usually used to make masa (nixtamalized maize dough; a specially treated corn flour in which the kernel is cooked and soaked in an alkali solution to make a dough which can be dried and milled) which is used to make tortillas, tacos, and tamales.

How maize is consumed varies greatly by country and region. For Mexico and Central America, masa is a staple carbohydrate and the main food use. In Sub-Sahara Africa, maize is consumed as a porridge. In the USA, of the ~1.244 billion bushels of human-consumed field corn:

785 (~63%) corn syrup sweeteners (fructose, glucose, dextrose)

245 (~20%) corn starch

214 (~17%) dry milled (for chips, flour (e.g., tortillas), cornmeal, or grits)

Approximately 100 million bushels of the commercial vegetable crop, sweet corn, is consumed in the U.S.

In Asia, maize is primarily consumed as baby ears in stir fried foods or grits. In parts of Europe (northern Italy and Balkan countries), human maize consumption is mainly as polenta. Most European countries including UK and Germany use the majority of maize produced for livestock feed, directly as forage or as silage.

With most of the maize produced being used for animal feed or ethanol production, the targets for human consumption include direct consumption, but also the animal products maize is fed to and the byproducts of ethanol production. For example, maize distillers spent grains are high in protein and total digestible nutrients. Yellow maize is high in carotenoids (precursors of vitamin A), B-complex vitamins, and vitamin E.[62]

3.2 Science-Based Nutrition Targets for Human Consumption

The targets are broadly grouped by the trait they are affecting.

3.2.1 Protein Quantity and Quality

Maize is generally low in two essential amino acids (lysine and tryptophan). An essential amino acid is one that the human body cannot synthesize and must obtain by consuming food that contain it. Quality Protein Maize (QPM) improved the amino acid composition and is currently grown in 32 developing countries and on over 3.5 million acres. Also, while higher protein may be useful in some human diets, lower protein in industrial maize could be of value to reduce nitrogen pollutions from industrial uses.

QPM in Popcorn and Sweet Corn

Approach: Because QPM has already been accomplished in grain maize, the genes and modifiers (the original gene source for higher lysine increased opacity and reduced kernel hardness which could be overcome through modifier genes to increase hardness and vitreousness) could be transferred to popcorn. The major storage protein in maize is zein which is low in lysine and tryptophan. There is good proof of concept using proprietary lines from ConAgra that conventional breeding, marker assisted selection and validated with biochemical assays will work without sacrificing affecting popping and taste. Similarly, the genes for QPM can be transferred to sweet corn using conventional breeding, marker assisted selection and validated with biochemical assays. Another approach would be to use CRISPR gene editing to delete the 19kD alpha zein loci (a family of genes) which reduce zein possibly without increasing opacity and not require (or require fewer of) the hardness modifier genes. CRISPR can also be used to directly modify the zein proteins to increase their content of lysing and tryptphan. CRISPR technology has less regulatory hurdles than conventional transgenic lines. Multiple new CRISPR edited maize lines which have already been made are awaiting funding for testing and subsequent breeding.

Low Protein Maize to Reduce Nitrogen Pollution for Industrial Uses

This is part of a circular economy model which aims to shift synthetic N further along the food chain (away from fertilizer and towards synthetic amino acids) for greater overall efficiency. Approach: Search for natural variation and use conventional breeding, marker assisted selection and validated with biochemical assays to select for low protein maize lines. Some of the longest running experiments in maize recurrent selection involve selected for higher (tended to increase the germ to endosperm ratio) and lower (tended to decrease the germ to endosperm ratio). Another approach is to employ CRISPR to selectively reduce the abundance of abundant proteins that are deplete in limiting essential amino acids and selecting for lines with reduced total protein amount but of optimized amino acid quality.

3.2.2 Vitamin and Mineral Concentrations

Increased Vitamin Concentrations

Approach: Maize is already a rich source of pro-vitamin A and other biologically important carotennoids, vitamin E and vitamin B complex. Building upon this foundation and with the knowledge that many of the pathway genes have been identified, conventional breeding and marker assisted selection should be successful. If genetic variation is lacking, CRISPR editing of the known genes should increase the level of useful variation. These genes can be transferred to grain, sweet, and popcorn.

Increased Mineral Concentrations

Like many crops, increasing iron and zinc would reduce "hidden hunger". Approach: Higher zinc maize has already been developed by HarvestPlus that provides up to 70% of the required daily required zinc needs. The lines were developed by conventional breeding and marker assisted selection. Though less progress has been made for increased iron, the opportunity remains [63] with new knowledge and approaches being developed. While it is expected that conventional breeding will be successful, CRISPR technology can add to the genetic diversity if needed.

Increased Fiber

Like many crops, increasing amylose fiber content is an option in maize that could enhance ability to reduce chronic disease risks.

Reducing Phytate (Salt of Phytic Acid)

Phytate chelates beneficial minerals such as Fe and Zn and is phosphorous rich which can lead to phosphorous pollution. Approach: Use natural variation and mutations followed by selective breeding to reduce phytate concentrations in maize.[64] The alternative approach relies on processing, for example the nixtamalized maize dough process reduces phytate's ability to chelate minerals.[65]

3.2.3 Bioactive Compounds: Antioxidants

Increased Antioxidants (including Anthocynanins)

Approach: Few crops have the diversity of seed (or cob) colors as maize (consider the diversity in an ear of Indian maize). Antioxidants can be readily increased in grain, sweet, and popcorn through conventional breeding and marker assisted selection should be successful.[66]

3.2.4 Corn to Improve Animal Food Products

Soft Starch (Non-Waxy) Dent Corn for Dairy Cows Feed, Increasing CLA, Reduced Palmitic Acid

Approach: Soft starch dent corn when fed to dairy cattle appears to be digested differently in the rumen (as opposed to the third stomach), potentially changing the period of fermentation in the rumen. This change has been suggested as shifting the fatty acid profile of the milk, although more information is required.

4. Next Possible Applications for Improved Nutritional Qualities in Potatoes

4.1 Background

Most cultivated potatoes belong to the species *Solanum tuberosum* ssp. *tuberosum* L. (2n=4x=48), an autotetraploid part of the Solanaceae (commonly referred to as the nightshade) family which includes other important economic plants like tomatoes, egg plants, bell peppers, tomatillos, and tobacco. Due to the economic importance of these crops, potatoes and its family members have extensive genomic (see: https://solgenomics.net/) and biotechnology resources. The Solanaceae family also includes plants like deadly nightshade. Hence the Solanaceae family includes plants with a wide range of bioactive compounds.

Potatoes are classified as being a starchy vegetable and due to evolution and plant breeding the anti-nutrient compounds found in the ancestors of potato have been bred out of modern commercial potatoes which are highly nutritious and safe to eat. The edible part of the potato plant are its tubers which are produced underground.[67]

Food and Non-Food Applications

Potatoes are the fourth largest staple food (third largest for human consumption and first vegetable) and together with wheat, rice, and maize provides most of the caloric intake for humanity. Potatoes provide more calories for human consumption than any other high-moisture vegetable—notably outstripping energy provided by sweet corn.

Approximately 50% of all potatoes are consumed as fresh food or processed into human food products. The remaining 50% is used for livestock feed, industrial starch manufacturing, to produce ethanol and alcohol products (e.g. vodka), or as seed potatoes. Potatoes are used in cooked food products including baked, fried, boiled, in soups, mashed, and chipped. They are rarely eaten raw.

Nutrition Overview

While best known for its starch, potatoes are also an important source of minerals, high quality protein, and dietary fiber which are retained after cooking. Potatoes are a rich source of vitamins (C and B_6), but vitamin C is reduced during cold storage, and both vitamins are reduced after cooking.

The outer layer of the potato (the skin) is nutrient dense and is high in fiber,[68,69] minerals, and antioxidants. Both the skin and flesh (the inner part of the potato) are high in vitamins, but the skin can prevent the vitamins from leaching into cooking water. In this way, whole potatoes vs. peeled potatoes are like whole grain wheat vs. refined flour. To understand the nutrient value of the potato, it is important to know if the nutritional value is determined from whole or peeled (skin removed) potatoes, as well as the effect of genotype, growing location, stresses (biotic, abiotic), storage conditions, cooking methods, and serving temperature on the chemical composition of potato tubers.[70]

Importance as a Food Crop

Potato is a very important crop from a world food security perspective due to its broad adaptability, high yield, highest food energy/area and nutrition/heath properties. Potatoes originated in the Andean highlands but are extraordinarily broadly adapted and grown in 160 countries, in 17.1 million ha, with a total production of 390.4 million metric tons. Currently, the majority of potato production is in Asia (205.4 million metric tons; mainly in China and India, the world's top potato producers), followed by production in the Ukraine, Russia, the United States, Germany, Bangladesh, France, Pakistan, and the Netherlands.[71] In addition to their broad adaptation, potatoes are a water efficient crop and produce more calories per liter of water than many other crops.[72] Potatoes are grown under irrigation and in rainfed environments with irrigated production often providing the more uniform product.

Plant Breeding and Production

Potatoes are unusual among the major crops that contribute to food security because they are sown from seed potatoes (tubers) or cuttings of seed potatoes (hence vegetatively/clonally propagated) which means that unique genotypes can be mass produced without the concerns of segregation, inbreeding, or open pollinations. Clonal propagation of edible plants can be done from tubers (e.g. potatoes), cuttings (done in fruit trees), and through apomixis. As mentioned above, the tuber is the commercial product and provides potatoes with its ability to grow and survive in harsh and diverse climates (it's being grown in 160 countries). In the center of diversity, there are many related species which provide a wealth of genetic diversity and novel traits for potato breeding including nutritional improvement.

In breeding new potato cultivars, a common procedure is to make crosses between two parents (both genotypically unique and highly heterozygous) and harvest the F1 seeds (full-sibs, highly heterozygous) which are sown to develop plants. The plants and their tubers are evaluated for advancement and if selected, the tubers are used for multiplication and inclusion in replicated testing and further evaluation. There is an increased interest to develop commercial diploid potatoes. If successful, potatoes propagation could be either through true seed or vegetatively through tubers, as desired. The Dutch have had success with diploid specialty lines. Diploid potatoes would make breeding for higher nutritional value much easier. For tuber propagation and breeding, as the number of tubers required for advanced testing in multiple locations increases, germplasm storage and maintenance are critical. Tubers are bulky, grown in soil with diseases and pests which can affect seed tuber quality and longevity. For this reason, potato breeders and germplasm curators have developed ex situ and in vitro methods of storage. Potato tissue cultures are disease free and commonly used to start the limited-generation seed certification system [73] and in germplasm exchange to avoid spreading diseases and pests with the germplasm. Cryopreservation, though expensive, is the most efficient method for long term preservation.[74]

An excellent resource for understanding potatoes is https://link.springer.com/chapter/10.1007/978-3-030-28683-5_6.

Commodity Classification

Potatoes are categorized into two main market classes, based on the amount of starch and use: fresh market and processing (high starch is required for potatoes used for processing as French fries and potato chips). Potatoes are also described based on visual categories [75] which include: red (often used for stews, soups, and salads), russet (often used for baking and making mashed potatoes), white (often used for grilling, steaming, boiling, and in mashed potatoes), yellow (often used in baking, roasting and mashing, and pan frying), purple/blue (often used for the bright colors), fingerling (visually interesting and often used in roasting, and petite (bite-sized versions of larger categories of potatoes that can be quickly cooked).

4.2 Science-Based Nutrition Targets for Human Consumption

The targets are broadly grouped by the trait they are affecting.

4.2.1 Dietary Fiber

Potato is a starchy vegetable and a major source of calories in the human diet. Most calories are from starch, hence developing potatoes with improved nutritional starches would be of value. Currently potato starch varies between 17 to 21% amylose (resistant starch) and 79 to 83% amylopectin. Resistant starch is a class of dietary fiber and improving the fiber content of potato would impact diabetes by reducing blood sugar spikes after eating potatoes and cardiovascular disease. Cooking and chilling can also increase the levels of resistant starch.[76]

Increased Amylose (Resistant Starch) Content

Increasing the level of amylose (resistant starch) in all classes of potato, with an emphasis on fresh market potatoes. Approach: Ongoing research using conventional plant breeding has identified potato lines ranging from 17% amylose to 42% amylose (Vales, personal communication, unpublished results). These lines need to be tested in more environments to determine how stable and repeatable the higher amylose content is in diverse growing environments. Approach 2: Potato lines with higher levels of amylose may be possible through gene editing. Approach 3: As mentioned earlier, cooking and chilling can increase resistant starch. It may be possible to develop potato lines or recipes which have higher levels of resistant starch after cooking, after cooking and chilling (e.g. in potato salad), or retain higher levels of resistant starch after reheating.

4.2.2 Increasing Potato Bioactive Compounds

Potatoes have a wide range of beneficial bioactive compounds which can be increased. For example, kukoamines have anti-inflammatory, antidiabetic, neuroprotective, treating nonalcoholic fatty liver disease, anticancer activity, and are μ-opiod receptor agonists. They are found mainly found in potato skin. Potatoes can be a rich source of phenylpropanoids which are a high value target as they positively affect gut health (more important) and are antioxidants.[77] Other than anthocyanins, many of these compounds are colorless and can be present in high amounts in white potatoes which increases real world impact. For example, white potatoes can have high amounts of chlorogenic acid, which has numerous health promoting properties. It should be noted that flavonols are colorless, highly desirable in the diet, and present in high amounts in some parts of the potato plant, but not in tubers.[78] Potatoes with blue, red or purple flesh (due to anthocyanins) typically have much higher amounts of colorless chlorogenic acid, but also abundant amounts of anthocyanins which are efficient antioxidants. Consumer education can increase the consumption of color-flesh potatoes that have exceptionally high amounts of antioxidants, rivaling kale. Baby potatoes often have increased amounts of bioactive compounds, as well as vitamins and are eaten with the skin on.

Increased Kukoamines (all classes)

Approach: Use conventional plant breeding or gene editing to increase their level in potatoes that are processed with the skin retained or possibly enhanced levels in the potato flesh.

Increased Phenylpropanoids (all classes)

Approach: Use conventional plant breeding or gene editing to increase their level in potatoes. It is understood that a consumer education effort will be needed to have consumers eat pigmented (high anthocycanin/antioxidant) fleshy potatoes.

Baby Potatoes (a specialty class)

Approach: Breeding and gene editing to produce varieties that produce a large amount of golfball sized potatoes. This high set tuber trait is important to keep the price down and maintain profitability for growers. Baby potatoes cook faster, are eaten with skin on, are more exotic which appeals to many consumers, are prized for their taste, smaller portions. These are all factors that appeal to especially younger consumers. These immature potatoes have higher amounts of protein, antioxidants, carotenoids and more than mature potatoes. Increased consumer education is needed so they realize what the advantages are.

4.2.3 Vitamins and Minerals

In addition to the bioactive compounds mentioned above, potatoes are also a rich source of vitamins and minerals including provitamin A (carotenoid), vitamin B1, C, B6, and B9, potassium (K), phosphorus (P), magnesium (Mg), zinc (Zn) and manganese (Mn). The major minerals (macrominerals, need more than 100 mg/day) in human diets are: calcium, K, sodium

(Na), Mg, P, chloride (Cl), and sulfur (S). The trace minerals (microminerals, need <100 mg/day) are iron (Fe), Zn, iodine (I), selenium (Se), copper (Cu), fluoride (F), Mn, and chromium (Cr).

Increased Carotenoids (pro-Vitamin A and other Bioactive Compounds; all classes)

Approach: Yellow flesh potatoes have the highest amounts of carotenoids (the amount is proportional to the color intensity). Potatoes have carotenoid types that are known to be important for eye health. Carotenoid amounts can be increased further with breeding and biotechnology, but not in white-flesh potatoes. In the United States, white-flesh potato sales dwarf those of specialty potatoes. So for the biggest impact one either has to change consumer eating preferences or focus on white-flesh potatoes. Higher pro-vitamin A trait could be stacked or identified in purple, red, blue flesh potatoes that also have high carotenoids and higher anthocyanin compounds. High carotenoid potatoes often have a reputation of also having higher sprouting (short dormancy), which is an issue that would need to be solved in order to have the highest amounts of carotenoids possible.

Increased Vitamin B1 and B9 (Folic Acid); all classes

Of the vitamins mentioned, folic acid is probably the most important target. Its daily allowance is often not met in the diet and its deficiency is a leading cause of birth defects. Approach: Conventional breeding or using biotechnology to increase the amount above its current level of ~10% of the recommended daily value are likely to succeed.

Increased Vitamin C, and B6; all classes

Approach: Conventional breeding or using biotechnology to increase the amount above its current level of ~35-45% of the recommended daily value. Recipes or processes that reduce vitamin loss in processing may need to be optimized. Potato is already a good source of these vitamins, but it could be improved.

Increased Minerals (P, K, Mg, Zn, Mn); all classes

Potassium may be especially important as Americans often do not intake the recommended amounts. Furthermore, potassium content is often a major component of how nutritionists rank diets. Potato is a generous source of K already and this is one of its most important nutritional contributions. Approach: Conventional breeding or using biotechnology to increase the amount above their current level, as well as, crop management.[79] Potatoes are a good source for some minerals.

4.2.4 Protein, Lipids, and Digestible Carbohydrates

While potato is considered a starchy vegetable, it does contain protein and the quality of that protein is high (similar to eggs).

Higher Protein Potatoes

Approach: Conventional breeding to increase the amount above their current level or using biotechnology to exceed natural variation.[41,80]

Research in China and in Texas indicate large genetic and environmental effects on protein and free amino acid content in potato. For example, evaluations for two years and at two locations of >200 advanced (mainly TX) breeding genotypes, have shown variation for protein between 8.3 and 14.6% dry weight basis (based on N measurements; protein= N x 6.25).[79,80] Growing potatoes under high temperature conditions resulted in increases of 10-20% in protein level, possibly due to the decrease in water and starch. The materials were developed using conventional breeding. Many of the genotypes are preserved in tissue culture as disease-free stocks. The effect of growing location should be expanded as protein and free amino acid content (like many other crops) exhibit large environmental effects.

<u>4.2.5 Reducing Deleterious Compounds</u>

Reduced Glycoalkaoids (mainly Solanine and Chaconine)

While these compounds are often considered as toxins, they have strong anticancer activity and other health promoting effects.[81] Approach: Plant breeding has been highly effective in reducing these compounds. All new potato varieties get tested for glycoalkaloids content (must be below 20 mg per 100 g FW).

Reduced Acrylamide in Cooked Products

Approach: Acrylamide can be reduced by selecting potatoes with reduced amounts of its precursors, i.e., free asparagine [80] and reducing sugars (glucose, fructose). Reducing sugars are produced during cold storage from sucrose via the acid vacuolar invertase, a process known as cold induced sweetening. Potatoes can be bred for resistance to cold induced sweetening.

5. Global Rice Improvement for Low Glycemic Index Aromatic and Non-Aromatic Varieties

5.1 General Introduction

5.1.1 Commodity Categories

Rice (*Oryza sativa* L.) is one of the world's most important staple crops, providing nearly one-fifth of global caloric intake. In the United States, long-grain rice production is concentrated in the southern states (Arkansas, Louisiana, Mississippi, Missouri, and Texas), with California contributing significantly to medium- and short-grain types. Rice varieties in the US are commonly classified by grain length and starch composition, which together determine cooking properties and consumer preference. The major categories of rice grain types in the US are listed in Table 5.1.

Table 5.1 Characteristics of major categories of rice grain types in the United States.

Group	Type	Description	Typical Use
Grain form	Rough rice	Unmilled rice with husk intact; also called paddy rice.	Stored or transported before milling; used for processing and seed production.
	Brown rice	Husk removed but bran layer retained; higher fiber and nutrient content.	Consumed as whole-grain rice; popular in health-conscious diets for its higher content of fiber, protein, vitamins, & minerals.
	White rice	Fully milled with husk, bran, and germ removed; polished for smooth texture.	Most common form for cooking and commercial sale; used in table rice and processed foods.
Grain size	Long Grain	Slender kernels (length-to-width ratio [LWR] >3.0). Cooked grains are light, fluffy, and separate easily.	Dominant in US production (Southern states). Preferred for table rice. Typically of intermediate amylose content.
	Medium Grain	Shorter and wider than long grain (LWR ~2.0–3.0). Cooked grains are moist and cling together slightly.	Common in California and Arkansas. Used in risotto, paella, and sushi-style dishes. Typically of low amylose content.
	Short Grain	Nearly round kernels (LWR <2.0). Cooked grains are soft, sticky, and cohesive.	Used for sushi, desserts, and specialty ethnic cuisines. Typically has low to no (waxy) amylose content.

Aroma	Jasmine-type	Long-grain types with natural aromatic compound (2-acetyl-1-pyrroline [2-AP]).	Imported varieties dominate; US breeding programs are developing competitive domestic lines. Typically has low amylose content.
	Basmati-type	Extra-long and slender grains with a natural aromatic compound (2-AP); distinct elongation and fluffy texture when cooked.	Used in Indian, Middle Eastern, and gourmet cuisines; valued for aroma and low glycemic index. Typically has high amylose content.
Amylose content	Waxy (Glutinous)	Very low amylose content (<2% amylose). Grains are highly sticky and cohesive when cooked.	Used in Asian desserts, processed foods, and brewing. Limited US acreage.
	Low-amylose	Typically, 10–20% amylose content (AC). Cooked grains are soft, moist, and slightly sticky.	Preferred for table rice in many Asian markets and used in ready-to-eat rice products.
	Intermediate amylose	Typically, 20–25% AC. Cooked grains are tender, fluffy, and moderately firm.	Most common class for US long-grain rice; widely used for everyday consumption and food service.
	High-amylose	>25% AC. Grains have firm texture when cooked and is associated with slower digestion.	Targeted for health-conscious consumers, parboiling, and export to South America; supports low-GI diets.
Grain color	Red rice	Pigmented bran layer rich in proanthocyanins (potent antioxidants); may be long or medium grain.	Used in specialty health foods and gourmet blends; valued for color and nutrition.
	Purple rice	Deeply pigmented bran with high anthocyanin (antioxidant) content.	Used in desserts, mixed-grain dishes, and functional foods emphasizing antioxidant benefits.

Figure 5.1 Classification of rice based on grain size.

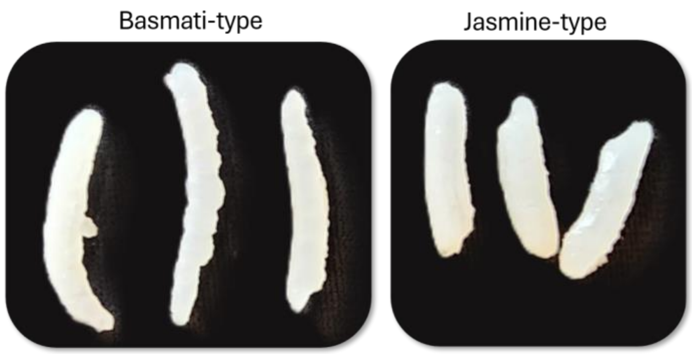

Figure 5.2 Cooked grains of basmati- and jasmine-type rice, with basmati-type exhibiting higher grain elongation ratio than jasmine-type upon cooking.

5.1.2 Production, Processing, and Consumption Patterns

Production and Processing

Rice production and processing involve three main stages: growing, milling, and cooking. After harvesting, rough rice (paddy rice) goes through a milling process to remove the hull and bran layer. This process results in polished white rice, which is the most consumed form worldwide. Milling removes the nutrient-rich bran and germ from the starchy endosperm. The milled endosperm consists of about 85-90% digestible carbohydrates, 7-10% protein, and 1-2% dietary fiber. The bran and germ that are removed contain most of the grain fiber, vitamins, minerals, and antioxidants.[82]

Processing Variants

Processing methods can alter the nutritional profile of rice. Brown rice skips the intensive polishing or milling step, so it retains the bran and germ. The bran provides more fiber, vitamins, minerals, and beneficial oils than the endosperm. However, skipping the milling results in a shorter shelf life since the natural lipids in the bran oxidize quickly in warm environments.[83]

Parboiled rice is produced when rice grains are hydrothermally steamed before milling. This pressure-assisted process moves water-soluble vitamins, particularly thiamin, from the outer layers into the endosperm. As a result, it retains more nutrients than regular white rice even after milling,[84] and it converts some of the starch to resistant starch, thereby lowering its GI.[85] Parboiled rice accounts for only about 20% of U.S. rice consumption because it has traditionally been considered a product for food service and ready-to-eat applications as opposed to home consumption, but consumption of parboiled rice is increasing in the U.S., and it accounts for 50-60% of global rice consumption.[86]

Consumption Patterns

Globally, rice is mainly milled into white or polished rice, the most consumed form, as the nutrient-rich rice bran and germ are removed during the milling process and often used for animal feed or for oil extraction. In the US, rice is used for table consumption, packaged foods, brewing, and pet feed. In Asia, boiled or steamed rice is a staple food that is usually eaten several times a day. Meanwhile, aromatic (jasmine-type and basmati-type) and pigmented (red or purple) rice are becoming popular, while health-conscious individuals target brown rice and/or high-amylose (low GI) and high-anthocyanin rice types. This shows a growing interest in rice that combines tradition with improved nutrition.

Primary Food Applications

Rice plays a significant role in many food traditions. It can be prepared in various ways, including steamed grains, porridge, noodles, rice cakes, and crispy snacks. Its mild flavor absorbs seasonings and sauces easily, making it perfect for savory dishes like curries and stir-fries as well as sweet desserts like puddings. In addition to whole-grain uses, modern food processors mill rice into flour for gluten-free products, brew it into regional drinks, and ferment it for traditional ingredients. This highlights the importance of rice as both a daily staple and a versatile ingredient in cuisines around the world.

5.1.3 Nutrition

Dietary guidelines around the world focus on eating more whole grains and reducing the intake of refined carbohydrates. Milled white rice is low in fiber and micronutrients, so biofortification is a common strategy to enhance its nutritional value.[87] In recent years, GI has gained attention. The Texas A&M Cereal Quality Lab recently developed a new rapid protocol for glycemic load estimation as a proxy for GI response of ready-to-eat foods loaded with quickly hydrolysable carbohydrates. This parameter measures how quickly carbohydrates raise blood glucose after a meal. The new approach follows an *in vitro* digestion for 60 min and quantification of glucose released against a D-glucose standard (9/100g). Conventional white rice usually has a high GI, which causes rapid spikes in blood sugar. In contrast, rice varieties with higher contents of amylose and/or resistant starch digest more slowly and result in a slower glycemic response.[88]

Resistant starch behaves similarly to dietary fiber, resisting digestion in the small intestine and fermenting in the colon, thereby supporting gut health.[89] Breeding rice with naturally higher resistant starch offers a promising way to improve metabolic health while keeping traditional eating habits. This approach supports global nutrition goals to develop staple foods that are both culturally familiar and beneficial to health.

5.1.4 Market Challenges in High-Amylose and Aromatic Rice

Demand for high-amylose and aromatic rice in the US is rising, but domestic varieties struggle to meet grain quality standards and compete in the market. The US imports over $1.6 billion worth of rice every year, with 60% being jasmine- and basmati-types. Domestic aromatic rice accounts for less than 1% of the market because current US varieties lack the aroma, texture, and flavor of imported varieties. US rice exports of high-amylose rice to South America are declining due to competition from Brazil and Paraguay. Globally, the demand and popularity of high-amylose rice are increasing, as it is linked to a low glycemic response (See section 5.2.1).

5.2 Justification: Rice Health Innovation Overview

Improving the nutritional quality of rice has become a national priority as global demand continues to rise alongside growing concerns about diet-related chronic diseases. Cardiovascular diseases are among the leading causes of death worldwide, accounting for approximately 19.8 million deaths annually and nearly one-third of overall global deaths.[90] Unhealthy diets, high glucose levels, obesity, and other metabolic risk factors are major contributors to cardiovascular diseases. Therefore, developing healthier staples is important. To address these challenges, various innovative strategies are proposed to the health benefits of rice, including increasing amylose concentration and modifying starch composition for low glycemic response, fortifying with micronutrients, and maintaining desirable sensory qualities like aroma and texture. These strategies reflect a broader goal of aligning crop improvement with public health needs. A summary of these ideas is presented in Table 5.2, which lists key recommendations and research priorities. The table provides a comparative overview of breeding targets, potential trade-offs, and areas for innovation.

5.2.1 High-Amylose Rice for Improved Glycemic Control and Cardiovascular Health

Dietary strategies that lower GI of staple foods can enhance cardiovascular health. Since rice is the primary source of calories for more than half of the world's population, reducing its postprandial glycemic response is important for public health. One approach is the development and consumption of high-amylose rice varieties. Higher amylose concentration is directly linked to lower GI values and reduced metabolic stress (Figure 5.3).

High-amylose starch resists gelatinization and enzymatic digestion, leading to compact crystalline structures that slow glucose release and lower glycemic load. As a result, high-amylose rice generally produces lower postprandial blood-glucose and GI values than low-amylose varieties.[42,88] Across 17 clinical trials, researchers consistently found that consuming high-amylose rice led to significantly lower postprandial blood glucose and GI values compared with diets based on low-amylose rice.[42]

Low-GI diets improve insulin sensitivity, reduce chronic inflammation, and decrease oxidative stress, endothelial dysfunction, and dyslipidemia, all of which are key factors in cardiovascular disease progression.[91] Therefore, foods that lower postprandial glycemia, including high-amylose rice, support healthier blood vessel function and better long-term health outcomes.

The same principle explains the naturally low GI of basmati-type rice, which has higher amylose concentration and longer, less easily digested starch granules. In contrast, jasmine-type rice has low amylose concentration and is digested quickly, producing higher GI values and larger glucose spikes.

However, global consumption patterns show that the most popular choices do not match these health benefits. Intermediate amylose rice is by far the most widely consumed category, holding a market share of 55 to 60%. Jasmine rice accounts for the second largest share at 12 to 15%, followed by basmati at 5 to 8% and high-amylose varieties at 3 to 5%. Meanwhile, glutinous or waxy rice makes up the smallest part of global consumption, representing just 2 to 3% of the total share.

High-amylose and basmati rice are healthier options compared to typical US varieties with intermediate or low amylose content. Changing these popular preferences could be an effective way to improve diet. Since high-GI diets are linked to a greater risk of type 2 diabetes,[92] and diabetes affects an estimated 40.1 million Americans,[93] encouraging consumers to choose lower-GI staples is a meaningful public health strategy. The rise in obesity, impacting 1 in 5 children and 2 in 5 adults,[94] further emphasizes the need for low-GI dietary options, as these diets promote better weight loss and improved metabolic health.[95] High-amylose and basmati rice thus offer healthier choices compared to typical US varieties with intermediate or low amylose content. Creating high-amylose aromatic rice would extend these advantages to consumers who enjoy the jasmine aroma but desire better health results. This rice would maintain the flavor profile of jasmine while offering a firmer texture and a significantly lower GI due to the higher amylose content, combining sensory appeal with the potential to prevent chronic diseases.

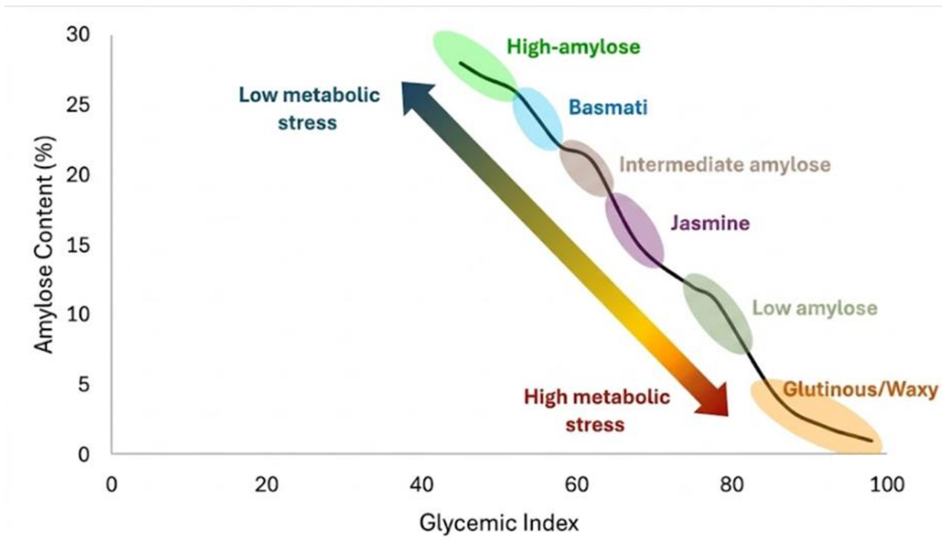

Figure 5.3. Graphical representation of the relationships between rice amylose content and postprandial glycemic response.

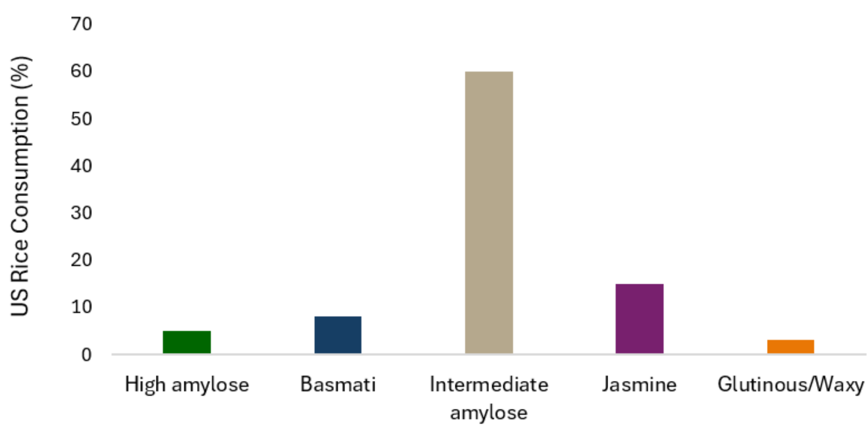

Figure 5.4. Percentage consumption of various rice types in the United States. Intermediate amylose rice constitutes the vast majority of consumption at 55–60%, followed by Jasmine (12–15%), Basmati (5–8%), High-amylose (3–5%), and Glutinous/Waxy varieties (2–3%).

Table 5.2. Targeted traits for healthful rice improvement.

Target Nutrient / Compound	Primary Goal	Category	Core Breeding & Agronomic Approach	Strategic Commentary & Technical Bottlenecks
Inorganic Arsenic (iAs)	Reduce grain concentration	Anti-Nutrients	Gene editing of metal transporter genes; mapping natural variation.	High health concern for all market classes. Editing root uptake transporters is tricky because they also transport beneficial Silica; targeting sequestration in leaves to reduce leaf-to-grain transport is a viable alternative.
Arsenic (iAs & oAs)	Reduce grain concentration	Anti-Nutrients	Alternate-Wetting-and-Drying (AWD) water management; furrow irrigation.	Non-flooded production can cut grain arsenic by 50-90% while conserving water. However, dry periods increase weed pressure and risk of rice blast disease.
Magnesium	Increase concentration	Vitamins, minerals	Marker-assisted selection (MAS), upregulating *OsMGT* transporter genes, and foliar fertilization.	Mg is a vital metabolic cofactor; deficiencies worsen insulin resistance and cardiovascular risk. Milling removes Mg-rich bran; thus, white rice is a poor source of Mg. Breeding should target transporter genes that actively load Mg into the endosperm. Applying foliar Mg sprays at the panicle initiation stage serves as a highly effective agronomic complement, directly enhancing grain accumulation in Mg-deficient soils.
Amylose Content	Increase concentration	Digestible Carbohydrate	Marker assisted breeding targeting the *Wx* haplotype for high amylose concentration	Higher amylose is the primary driver of resistant starch formation, lowering the GI and reducing post-meal blood sugar spikes. High-amylose rice absorbs less water and cooks into a dry, firm, and non-sticky texture that faces

				low consumer acceptance in markets accustomed to soft, sticky rice varieties.
Resistant Starch (RS)	Increase concentration	Dietary Fiber	Breeding (MAS) for *beIIb* and *Wx-a* alleles; gene editing of starch-synthesis pathways.	Converts digestible carbs into dietary fiber, creating low-GI and low-calorie rice to combat type II diabetes epidemics. A major challenge is avoiding hard cooked textures and poor field stand establishment.
Slowly Digestible Starch	Increase concentration	Protein, Lipid, Digestible Carbohydrate	Basic research & gene editing of known starch synthesis genes.	Provides a lower glycemic index and smoother post-meal blood sugar curve without increasing dietary fiber. Requires foundational research to map unknown regulatory genes.
Non-Colored Antioxidants	Increase concentration	Positive Bioactives	Breeding, MAS, and gene editing of pathways for gamma-oryzanol and Vitamin E.	Concentrated in the bran. Increasing these can naturally slow down the rapid oxidative rancidity of brown rice, a major shelf-life limitation in tropical regions.
Colored Antioxidants	Increase concentration	Positive Bioactives	Breeding, MAS, and gene editing of color-bran genes (*Pb*, *Rc*, *Kala3*).	Leverages anthocyanins and proanthocyanidins. Distinct colors (red/black) may increase consumer appeal by eliminating the perceived "dirty" look of standard brown rice.
Lysine Content	Increase concentration	Protein, Lipid, Digestible Carb	Gene editing.	Directly addresses the primary limiting essential amino acid in cereal grains to improve total protein quality.
Oxidation & Lipase	Reduce rancidity in brown rice	Anti-Nutrients	Gene editing to knockdown lipase activity.	Extends the shelf life of highly nutritious brown rice, helping it compete with polished

				white rice in tropical climates where rancidity occurs quickly.
Lipids / Rice Bran Oil	Healthier lipid profile	Protein, Lipid, Digestible Carbohydrate	Gene editing.	Tailors the fatty acid composition of the bran layer to favor heart-healthy profiles.
Total Protein Content	Increase concentration	Protein, Lipid, Digestible Carbohydrate	Screening public deletion mutant collections (e.g., Katy library at GSOR); mutation breeding.	Crucial for developing nations lacking diverse protein sources. Co-consuming protein slows starch digestion (lowering GI). Overcomes the hurdle of current proprietary high-protein germplasm (e.g., Parish rice) being locked out of research use.
Phytic Acid	Decrease concentration	Anti-Nutrients	Breeding or gene editing utilizing known low-phytic-acid (*lpa*) mutants.	Phytic acid acts as an anti-nutrient, binding crucial minerals (Ca, Fe, Zn). Reducing it increases bioavailable phosphorus and essential minerals in the endosperm.
Vitamins (Thiamine / B1)	Increase concentration	Vitamins & Minerals	Post-harvest parboiling (hydrothermal steaming prior to milling).	Effectively pushes water-soluble nutrients from the hull/bran into the endosperm, historically preventing beriberi. Requires energy/water inputs and alters the grain's final color and texture.
Essential Minerals (Zn, Fe, P, K, Mg)	Increase concentration	Vitamins & Minerals	Traditional biofortification (HarvestPlus), MAS for known mineral QTLs, and target-specific soil/foliar fertilization.	Addresses widespread micronutrient deficiencies in rice-dependent populations. Breeding is highly viable due to wide, documented natural genetic variation for mineral accumulation.

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