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Nanotechnology-Enhanced Biofortification: A Review on Advancing Nutrient Bioavailability

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Abstract: Biofortification is a practical way to improve nutrient levels in crops as they grow, helping to reduce micronutrient deficiencies worldwide. This paper examines innovative solutions, including biofortification through nanotechnology, to address the issue of "hidden hunger." It is easier to deliver key nutrients, such as iron, zinc, calcium, and vitamins, more effectively by adding nanoparticles and utilising techniques like nanoencapsulation and nanocarriers. This enhances the absorption of essential nutrients in the body, thereby increasing crop yield and nutritional value. Due to their small size, nanoparticles are highly effective in delivering nutrients precisely where they are needed. However, their overuse raises concerns about safety for both health and the environment. These risks can be reduced by implementing safer production methods and controlling the quantity of nanoparticles used. Nanoparticle-based biofortification is now being applied in farming, food products, and nutrition programs. Side effects, such as toxicity, still exist and need careful testing and proper handling to avoid long-term problems. By combining the concept of biofortification with nanotechnology, this paper outlines a practical and sustainable approach to fighting malnutrition and building healthier societies.

Keywords: Micronutrient Deficiency; Biofortification and Nanotechnology; Nanoparticles Work, Sustainable Nutrition; Fighting Malnutrition; Building Healthier Societies; Nanoencapsulation and Nanocarriers.

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1. Introduction

Nutrient bioavailability refers to the amount of nutrients from food or supplements that the body absorbs and utilises for various functions. It involves multiple steps, including digestion and transport [1]; [17]. This process determines how effectively the

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body can benefit from nutrients. While macronutrients are typically absorbed easily, micronutrients vary in their absorption methods, which depend on factors such as nutrient forms, how they are incorporated into foods, and whether other meal ingredients ease or hinder absorption [2]; [19]. Understanding these factors is crucial for developing dietary guidelines and fortification strategies to improve nutrient intake and combat deficiencies that affect people worldwide [101]. Micronutrient deficiencies are a global issue, affecting billions of people, particularly in low- and middle-income countries. Deficiencies in essential micronutrients lead to serious health consequences [3]; [13]. Approximately 5 billion people (61.14% of the world's population) lack essential nutrients, including iodine, vitamin E, calcium, iron, riboflavin, folate, and vitamin C.

Women are more likely to experience deficiencies in iodine, vitamin B12, iron, and selenium. At the same time, men are more likely to be deficient in magnesium, vitamin B6, zinc, vitamin C, vitamin A, thiamin, and niacin. Among children under five, over half are deficient in iron, zinc, or vitamin A, and two-thirds of women aged 15–49 are deficient in iron, zinc, or folate [4]. Nutritional intake is crucial for supporting overall health and protecting the body from stress and other physical and mental challenges [97]; [114]. Due to the lack of essential nutrients, both physical and mental well-being can suffer. Essential nutrients play important roles in supporting mental wellness [80]; [109]; [110]. As age and experience increase stress, cognitive health can be enhanced by consuming a well-balanced diet rich in specific micronutrients [5]; [95]. Overcoming malnutrition depends on how well the body can absorb and use the available nutrients. One innovative approach to address this challenge is biofortification. Biofortification is a method used to enhance the micronutrient content of food [99].

This approach is particularly beneficial for combating nutrient deficiencies in countries where a variety of foods is lacking, supporting a healthier lifestyle. Therefore, biofortification makes crops more nutritious every day and offers a straightforward way to address malnutrition, helping people achieve better health [9]; [94]. The enzymatic hydrolysis technique employs enzymes to break down nutrients into smaller particles that can be more easily absorbed by the body, such as papain, which breaks proteins into smaller peptides [8]; [102]. In recent years, micronutrient deficiencies have become a significant global issue. Many countries have started incorporating micronutrients, such as vitamins and iron, into their food to enhance nutritional quality and meet the body's nutrient needs, thereby providing health benefits [12]; [15]. This is more effective than consuming direct nutrient supplements. Although food fortification is practised worldwide, it is becoming less effective as the added nutrients are present in trace amounts, and it has several drawbacks. Hence, food fortification through nanotechnology has been implemented, where nano-sized particles are incorporated into food, as these offer a greater advantage over the incorporation of bulk particles [10]; [96].

2. The Vital Role of Micronutrients

Micronutrients are essential in small amounts, as they play a crucial role in supporting metabolic processes, maintaining tissue function, and enhancing the immune system. Vitamin D is essential for maintaining bone health and immune function, as it regulates calcium levels [7]; [20]. Vitamins A, C, and E, along with minerals such as selenium and zinc, are crucial for supporting physical barriers in the body, including the skin and mucous membranes, and for strengthening immune cell function [81]. Similarly, vitamin B12 and iron are essential for the production of RBCs and the transfer of oxygen [6]; [57]. They are important in preventing anaemia and supporting overall vitality, especially in populations having high metabolic demands, such as athletes [106]; [113]. From an agricultural perspective, biofortifying crops with essential micronutrients such as zinc and iron has proven to enhance the nutritional value of food and its role in addressing global nutrient deficiencies [14]. However, insufficient intake of nutrients due to dietary gaps or poor bioavailability remains a global challenge affecting the population; thus, ensuring an adequate supply of micronutrients through balanced diets and fortified foods is essential for preventing deficiency-related diseases and improving human health [11]; [16].

3. The Role of Biofortification in Enhancing Micronutrient Intake

Biofortification has become a powerful tool in combating micronutrient deficiencies in areas where food options are limited. Biofortification enhances the nutritional value of staple crops, including rice, wheat, and maize, during their growth by employing methods such as agronomic practices, traditional plant breeding, and genetic engineering. A well-known example is ‘Golden Rice,’ enriched with beta-carotene, which has been developed to tackle the widespread vitamin A deficiency in communities that depend on rice as a staple food. Techniques like agronomic biofortification, which use nutritionally enriched fertilisers and beneficial soil microbes, have successfully increased the levels of zinc and iron in crops [21]; [22]. These methods naturally enhance the nutrient content of food without affecting its taste and appearance. Genetic engineering has enabled the introduction of specific genes into crops to enhance the bioavailability of essential nutrients, such as zinc in barley and iron in cassava. Programs like HarvestPlus highlight the practical application of biofortification by providing nutrient-rich sweet potatoes and maize in Asia and Africa [104]. These effects show how biofortification is scaled up to improve global nutrition. Innovative techniques, such as gene editing and omics, are paving the way for the quicker and simpler development of these nutrient-rich crops. In a world grappling with climate change and food shortages, biofortification helps enhance diets and make crops more resilient to environmental challenges, offering a solution to the issue of “hidden hunger” [19]; [23].

4. Methods of Biofortification

Biofortification is a sustainable approach that focuses on improving the nutritional content of crops during their growth. The primary methods employed for biofortification include agronomic techniques, conventional plant breeding, and transgenic approaches. Each of these has unique characteristics and applications [79]; [111]. The primary biofortification techniques—agronomic practices, conventional breeding, and transgenic methods—are illustrated in Figure 1.

Agronomic Biofortification: This method involves the application of nutrient-rich fertilisers or soil treatments to increase the concentration of micronutrients in the crops. These fertilisers include traditional options and advanced alternatives, such as chelated or nano-fertilisers, which are applied to the soil or directly sprayed on plants to enhance nutrient uptake. This method provides quick solutions but is short-term. The success of this technique depends on the soil quality, the type of fertilisers used, and the specific plant cultivated. Nano-fertilisers, with their small size and precise delivery mechanisms, have shown potential in improving nutrient absorption by plants [24]; [25].

Conventional Breeding: This technique uses the conventional method of plant breeding to develop nutrient-enriched crop varieties [100]. It involves the selection and crossbreeding of plants within a species that have genetic differences and higher micronutrient levels. This approach has led to the development of crops such as beans with increased iron content and rice with enriched zinc. Although it is often practised and successful, the approach is terribly slow and depends heavily on the crops' genetic variability [25]; [26].

Transgenic Approaches: Genetic engineering enables scientists to directly insert specific genes or foreign DNA into crops, thereby enhancing their nutritional value and potential health benefits. This approach is particularly beneficial for crops that lack the natural genetic variability necessary for certain essential nutrients. One such example is the "golden rice," which is developed by introducing genes that aid in the production of vitamin A. This method still struggles with social acceptance and regulations, although it is more helpful than conventional practices. The latest advancements in omics technologies have enhanced the precision of these methods, making it easier to identify and modify specific genes [24]; [27].

Gene Editing: Advancing gene-editing methods, such as CRISPR/Cas9, TALENs, and zinc-finger nucleases, are powerful tools that enable scientists to directly alter the genome of crops to enhance their nutritional value. These techniques have been used to develop rice varieties enriched with β-carotene and wheat with high zinc content. CRISPR/Cas9 is more precise, cost-effective, and versatile, but is also known for off-target effects [24].

Microbial Fortification: This approach involves the application of beneficial microorganisms, such as rhizobacteria and mycorrhizal fungi, that enhance the absorption, transportation, and storage of nutrients in crops. For instance, fluorescent Pseudomonas enhances the plant's iron absorption.

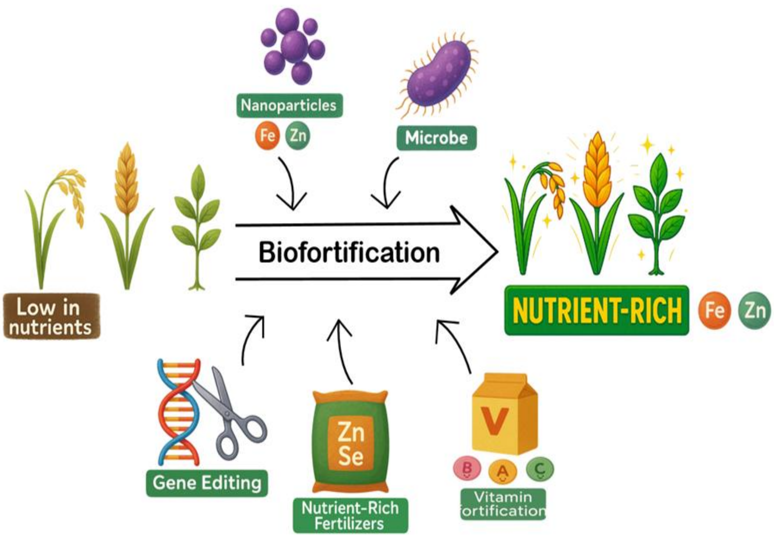

Figure 1: Mechanisms of biofortification to enhance nutrient content in crops

Genes from Aspergillus species have been used to increase the lysine and phytate concentrations in crops such as rice and wheat [24]; [25]. A comparative overview of various biofortification approaches is depicted in Figure 2.

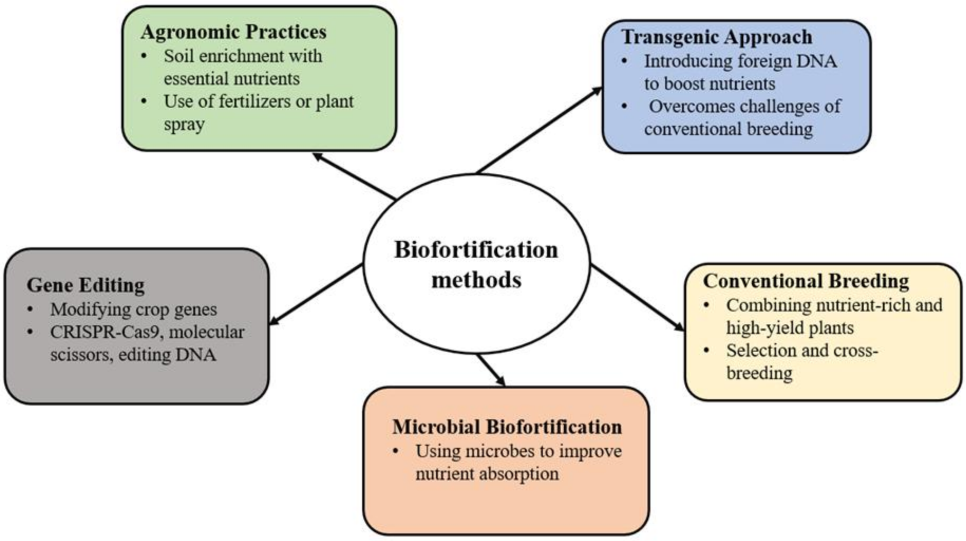

Figure 2: Biofortification approaches and methods to increase nutritional bioavailability

5. Fundamentals of Nanotechnology in Food Systems

Nanotechnology refers to the manipulation of materials at the nanoscale level, as they have distinctive features compared to bulk particles [28]. Nanoscale materials tend to have a greater surface area relative to their volume. The use of this technology in the food industry helps improve the delivery of nutrition through methods such as nano-emulsions [29]. These nanoparticles enhance stability and bioavailability, ensuring efficient absorption of essential micronutrients by the body. Malnutrition can be addressed using advanced technologies integrated into food processing and product development, as they enable more efficient nutrient delivery. Nanoencapsulation, a key application of nanotechnology, is a process that enhances the delivery, stability, and bioavailability of micro compounds by enclosing them within a protective nanoscale covering, thereby shielding them from degradation [30]; [31]. Enhanced control is provided over the release of active compounds [32]. It improves the shelf life of particles via nanopacking. This technology helps improve the quality of food by controlling the release and targeted delivery of nutrients [33]. The potential advantages of nanoparticles in enhancing nutrient delivery and product stability are summarised in Figure 3.

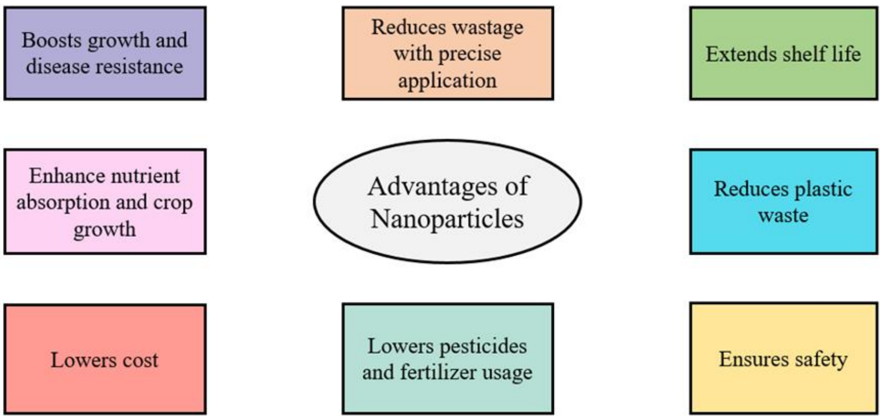

Figure 3: Pros of nanoparticles in the production of bio-fortified food products

Another important aspect of nanotechnology is the use of nanocarriers, which are nanoscale materials designed to enhance the delivery, stability, and bioavailability of bioactive compounds [35]. They help sustain the quality of the compounds through effective transport. Their small size enables easy modification, which makes them durable for packaging solutions [33]. An example of a nanocarrier, α-Lactalbumin, is used to deliver bioactive compounds, which have shown therapeutic benefits [36].

Similarly, liposomes are another type of nanocarrier, which are tiny spherical structures made up of natural fats, that can hold both water-soluble and fat-soluble substances [34]. They improve the effectiveness of pesticides and fertilisers by releasing them gradually in a controlled way [37]. In medicine, it can carry larger amounts of therapeutic agents and avoid being broken down by the body's immune system, which makes treatments more effective [38].

Nanoparticles (NPs) are widely used in biofortification as a key application of nanotechnology. These particles range in size from 1 to 100 nm, encompassing both metallic and metal oxide materials [40]. These can efficiently deliver essential micronutrients to plants, boosting their nutrient content and overall yield. This method helps address food nutrient deficiencies while supporting sustainable farming practices [39]. However, it is essential to focus on environmentally friendly methods for producing these nanoparticles to ensure their safety and minimise any potential harmful effects, thereby maximising their benefits in the food industry [41]. The bioavailability of vitamins and minerals plays a significant role in food fortification. However, there are challenges with conventional delivery methods. Some vitamins and minerals are poorly absorbed in the gastrointestinal tract due to factors like dietary interactions or their chemical forms. Additionally, these nutrients can be degraded during storage or cooking. For example, vitamin C and certain B vitamins are sensitive to heat and light, while minerals like iron can form compounds in food that are hard for the body to absorb. To overcome these issues, there is a need for improved delivery methods that protect nutrients and make them more readily available for the body to utilise.

6. Synthesis of Nanoparticles for Biofortification

The production of nanoparticles for agricultural biofortification relies on a variety of techniques, broadly categorised into physical, chemical, and biological approaches. Each method offers distinct advantages and disadvantages, which in turn influence the resulting nanoparticle characteristics and environmental impact.

6.1. Physical Synthesis

Physical methods involve energy-intensive techniques. The upside is that these techniques often yield nanoparticles of exceptional purity, minimising chemical contamination. For instance, pulsed-laser liquid ablation is an eco-friendly technique that creates stable nanoparticles in colloidal form. However, this approach requires a significant energy input and specialized, often expensive, equipment [42]; [43]. It is a highly controlled but energy-intensive method for creating nanoparticles from source material.

6.2. Chemical Synthesis

Chemical approaches use reduction reactions to convert high-valence compounds into elemental nanoparticles. Stabilisers, such as polysaccharides and surfactants, are used to manage the particle size and stability of the nanoparticles, as well as to prevent clumping. This method is cost-efficient and allows precise control over nanoparticles. But the downside is that the use of toxic chemicals during the process can pose environmental risks. It is a balancing act between achieving desired nanoparticle characteristics and minimising harmful by-products [42]; [43].

6.3. Biological Synthesis

Biological synthesis presents an environmentally friendly alternative. Microorganisms, such as fungi, algae, bacteria, or plant extracts, are used to reduce metal ions and stabilise nanoparticles. This method is gaining traction because it produces nanoparticles with high bioactivity and low toxicity, making them particularly attractive for biofortification. For example, selenium nanoparticles synthesised from plant extracts or microbial systems have enhanced stability and bioactivity, making them suitable for biofortification applications [25]; [43]. Various nano-delivery systems and their synthesis methods for nutrient biofortification are detailed in Table 1.

Table 1: Nano-delivery mechanism for nutrients with methods and examples

Mechanism	Description	Example nutrient	Synthesis method	Reference
Nano-fertilizer	Slow release for better absorption	Nano-Fe, Nano-Zn	Precipitation method, Vacuum Arc deposition, Chemical Absorption	[44]
Nanocarriers	Encapsulated nutrient for better release	α-Lactalbumin	Enzyme-assisted self-assembly & cross-linking	[45]
Nano-chelates	For improved intake	Nano-Fe-EDTA	Self-assembly, Ultrasonification	[45]
Nano-emulsions	Stable nano-sized nutrient dispersions	β-carotene	High-pressure homogenization, microfluidization, Ultrasonification, & spontaneous emulsification	[46]

Nanostructured systems	Targeted nutrient release at plant sites	Vitamin D	High-pressure homogenization	[47]

The precursors of Zn and Fe were added with sodium hydroxide dropwise under stirring, and white precipitates of nanoparticles were formed, which were filtered, washed, dried, and calcinated at 400-500°C [44]; [18]. α-Lactalbumin is dissolved in distilled water with the pH adjusted to 7.4, and the protein is desolvated by adding ethanol drop by drop while being stirred. As ethanol is added, nanoparticles start to synthesise [112]. To stabilise the nanoparticles, glutaraldehyde is used as a cross-linking agent. The nanoparticles are then centrifuged and rinsed to remove excess chemicals, and subsequently dissolved in water [36]. A solution of ferric chloride hexahydrate was prepared, and EDTA was added to this solution, which was then continuously stirred to facilitate chelation and the formation of an EDTA complex. Fe-EDTA nanoparticles were formed by ultrasonication of the chelated solution, which facilitated particle size reduction [45].

β-carotene was mixed with the oil phase, water, and an emulsifying agent to prepare a coarse emulsion; the mixture was then homogenized using high energy to produce nano-emulsions. Little droplets are created by forcing the emulsion through tiny valves. These droplets are then sent into an interaction chamber, where two streams meet under high pressure, producing strong disruptive forces that further reduce the particle size. The size is additionally reduced to the nano range by ultrasonification [46]. Vitamin D is nanostructured, encapsulated in an oil-in-water nano-emulsion using high-pressure homogenization as the synthesis method. A coarse emulsion with larger droplets is first created and then forced through a small valve under high pressure, resulting in the breaking of the larger droplets into smaller ones, achieving nano-sized droplets [47].

7. Nanotechnology in Bio-Fortification

Nanotechnology in biofortification utilises nanomaterials to enhance crop nutrient absorption and increase their nutritional value. Delivering nutrients to plants at a molecular level helps enhance crop quality and protect against environmental stresses, including drought and pests. This innovative approach would lead to the development of healthier and more nutritious crops, providing a practical solution to combat malnutrition and enhance global food security.

7.1. Minerals

7.1.1. Iron

Iron is an essential micronutrient critical for supporting overall health and preventing deficiencies. It plays numerous vital roles in the human body, including binding and transporting oxygen through blood cells, supporting energy production, regulating body temperature, and more [48]. Ferritin, a key iron-storage protein, shows high metabolic activity and can store thousands of iron atoms [49]; [50]. Iron homeostasis involves recycling, where specialised cells break down aged blood cells and generate new ones. Excess iron is stored in liver cells. The body tightly regulates iron absorption from the intestine, where iron is stored as ferritin and released into the bloodstream when needed. This process is mediated by receptors on the surface of cells that recognise and bind to ferritin for further transport and use [50]; [51]. Iron deficiency is a widespread condition affecting billions of people worldwide [73]. The body cannot produce iron; it is introduced into the body through food [118].

This nutritional disorder has multiple causes, including inadequate food supplements, poverty, and a lack of knowledge about the importance of iron-rich nutrients. Thus, the food supplements are being fortified with iron. Iron-fortified foods were found to be more effective than direct iron supplements; however, the world is facing difficulties in overcoming nutrient deficiencies due to major drawbacks [98]. Thus, iron is made to be effectively absorbed by developing nanoparticles, as their smaller particle size provides a large surface area for absorption. These nanoparticles possess anticancer properties and exhibit both magnetic and electrical properties, rendering them effective for targeted delivery [52]. Several studies illustrate the potential of NPs based on iron fortification methods. One of them is pre-milling fortification of wheat grains with iron and zinc, which increased their content by 54% and 62%, respectively. This method did not affect the quality of the flour [53]. It is a cost-effective way to tackle public health issues. Fortifying wheat grains ensures that nutrients are evenly distributed, improving diets for large populations.

Germination studies revealed that Allium cepa seeds treated with superparamagnetic iron oxide nanoparticles exhibited an enhanced seed success rate of 62%, compared to 70% in water, 58% with iron(II) ions, and 44% with iron(III) ions. This showed potential for targeted delivery [54]; [103]. This superparamagnetic property enables the direct delivery of nutrients into the seeds, thereby improving efficiency. This method can enhance farming practices and increase nutrient levels in crops, leading to higher crop yields. Another method of iron NP fortification involves spray drying, where iron gluconate achieved 60% bioavailability and increased iron absorption by 38% in intestinal cell models [55]. Spray-drying NPs reduced toxicity and enhanced iron nutrients. This is a great solution for iron deficiency, especially anaemia. This method is used in food fortification to enhance overall health and nutrition. These studies demonstrate that iron is essential for various bodily functions, yet deficiency remains a significant global health challenge. NPs-based iron fortification methods enhanced bioavailability and reduced toxicity. These novel approaches could be highly effective in tackling iron deficiency and boosting public health.

7.1.2. Zinc

Zinc is a vital micronutrient that plays a crucial role in maintaining human health. It acts as a catalyst and regulatory factor in various biological processes, playing a particularly important role in homeostasis by supporting the immune system and regulating cell death. It is necessary for growth, development, and metabolic functions in several organisms. Zinc particles play a catalytic role in metabolism, a structural role in gene expression related to protein stability and DNA processes, and a regulatory role in zinc homeostasis. The primary sources of zinc to meet the body's requirements are dietary zinc and supplements. Zinc deficiency occurs when the body does not receive enough zinc for its normal metabolism, resulting in retarded growth and, in some cases, mortality. The children of growing age do not get sufficient nutrients from the food they consume due to their poor background and poverty. However, the amount of zinc obtained from food is significantly lower due to its low absorption rate. Supplements are available in organic, inorganic, and biozinc forms, but they have been found to exhibit potential side effects. Thus, zinc nanoparticles are being used as they are a desirable choice to meet the body's need for zinc.

ZnO nanoparticles have higher solubility, stability, and biocompatibility. They also have a smaller particle size and a large surface area, making them suitable for high absorption [56]; [58]. One such study showed that spraying ZnO NPs on mushrooms increased zinc absorption by 10.46 mg/2 g with 5000 mg of ZnO NPs. Optimum efficiency was seen at 200 mg of ZnO NPs per kg of substrate [59]. This research demonstrates how nanotechnology can help address zinc deficiencies by enhancing the nutritional density of food. It also shows that the right amount of nanoparticles should be used, as excessive use can be wasteful and damage the environment. In another study on the use of fertiliser, adding ZnO NPs at 5 kg per hectare with a Zn-biofertilizer of 20 g/kg enhanced the growth of the Triticum aestivum plant. The plant height increased by more than 14%, and protein content increased by 30.74%. The zinc uptake rose by more than 66% [60]. This method can enhance crop production, resulting in more nutritious and healthier food. It reduces the need for chemical fertilisers, which can harm the soil and water. It can also ensure that crops receive enough zinc to combat deficiencies. Zn fortification is used to lessen toxicity.

In an investigation, the 1% Zn spray on the PS-2015 wheat variety increased grain yield and reduced cadmium levels in wheat in the field near Peshawar, Pakistan [61]. Zinc is essential for plant health as it helps in growth, enzyme activity, and stress resistance. Zinc sprays on crops can enhance productivity and nutritional value, particularly in soils with poor or polluted conditions. This method mitigates the adverse effects of cadmium and enhances the quality of wheat, potentially benefiting food security and promoting more sustainable farming practices in areas contaminated with cadmium. Zinc fortification is a crucial approach to overcoming zinc deficiency and improving nutrition. ZnO nanoparticles enhance zinc absorption, boost plant growth and yield, reduce the dependency on chemical fertilisers, and promote sustainable farming. They also help to lower heavy metal toxicity and ensure safe food. In the regions with high nutrient deficiencies and soil contamination, these approaches are crucial for enhancing food security and public health.

7.1.3. Calcium

Calcium is a key mineral essential for various body processes, such as growth and maintaining structural strength [62]. It is crucial during life stages such as childhood, pregnancy, lactation, and after menopause. Calcium is available in reactive forms. Calcium oxide is one of the most used forms of calcium [63]. In the human body, calcium metabolism maintains stable blood levels through dietary intake, and bone regulation is controlled by parathyroid hormones [65]. A deficiency in calcium in the body can lead to health issues, such as bone loss and hypertension during pregnancy [66]. This highlights the crucial need for calcium intake, particularly during pregnancy. The role of calcium is vital in agriculture, where calcium deficiency negatively affects plant health and yield [67]; [108]. Calcium is crucial for cellular and structural functions in plants, and its deficiency limits growth and yield, causing a risk to food security. To address calcium deficiencies in the population, calcium fortification has emerged as a viable solution [64]. This method enriches calcium in commonly consumed food products, helping the low-income population, for whom access to calcium-rich foods is limited [68].

It supports bone health and reduces the dependency on calcium supplements. By incorporating calcium fortification and emphasising its importance in agriculture and nutrition, both human health and crop productivity can be enhanced. A study examined the impact of adding nanocalcium from catfish bones to noodles with 30% seaweed. The study evaluated nanocalcium at various concentrations, and a 1% concentration showed the best results, with 1.5% calcium in the fortified seaweed noodles [69]. This study demonstrates the potential for enriching everyday foods and utilising by-products from fish processing. Fortified seaweed noodles are a popular option in the food market, appealing to consumers who are concerned about making healthier food choices in their diets. Hen eggshells have natural calcium content. One such study incorporated calcium from eggshells into a traditional Polish bread spread. The study demonstrated that enriching bread spread with eggshells and other nutrients led to a 2.5-fold increase in calcium [70]. This innovative approach offers health benefits and reduces calcium deficiency. It also promotes the usage of by-products. This calcium-enriched bread spread meets consumer needs for nutrition and adds value to the market.

Fortifying infant milk formulas with calcium is essential to meet their nutritional requirements. One such study investigated the impact of differing ratios of soluble and insoluble calcium in infant milk formula. The formulation contained a total calcium concentration of 1500 mg/L. The results showed an increase in calcium level from 1.44 mm to nearly 7 mm [71]. This method helps in increasing calcium availability, which is crucial for healthy bone development in infants. This research carefully developed a fortified infant nutrient product that fulfills the dietary requirements while ensuring safety and appeal. Thus, calcium fortification plays a crucial role in enhancing human nutrition. By utilising innovative methods, calcium deficiencies can be addressed by enhancing the nutritional value of everyday food items. These strategies enhance public health, reduce the need for supplements, contribute to food security, and satisfy consumer preferences for healthier options, ultimately.

7.1.4. Magnesium

Magnesium is an essential mineral required in trace amounts to maintain physiological activities in the body. Magnesium appears in the body as a cation and is involved in various bodily functions, including bone development, muscle contraction, nerve function, and enzymatic reactions. Magnesium is also crucial for energy production [72]; [74]. Magnesium deficiency is worldwide. The human body faces insufficient magnesium levels due to improper dietary intake and poor absorption. Magnesium deficiency is particularly noteworthy because it can lead to various health issues, including heart disease and other complications [75]. This necessitates dietary management. To overcome such deficiencies, magnesium fortification has been implemented, which involves adding magnesium to specific foods or beverages to enhance their nutritional value. This method is initiated in areas where dietary habits promote optimal magnesium consumption. By this fortification method, health authorities aim to reduce the risk of deficiency and improve health outcomes.

The University of Toronto developed a method for creating double-fortified salt that employs titanium dioxide (TiO_2) to mask its reddish color. Due to health concerns about TiO_2, researchers are exploring magnesium carbonate as a safer alternative. Their study found that higher levels of MgO_3 effectively masked the colour. A 41% MgO_3 coating in a gum Arabic or soy stearin blend effectively improved colour, also making it a replacement for TiO_2 and supporting efforts to combat micronutrient deficiencies [76]. This shift to magnesium carbonate not only enhanced food nutrition but also had an application in food fortification and public health. Refined wheat flour is low in magnesium, making it suitable for fortification. This study investigated the effects of adding magnesium ions from gluconate and lactate salts, i.e., magnesium salts, on bread quality, focusing on physical characteristics. Optimal results were found with 100 mg/100 g of magnesium from lactate salt and 150 mg/100 g from gluconate salt.

Increased magnesium levels resulted in denser crumb structures and greater bread hardness, and gluconate produced a darker colour than lactate. In conclusion, both gluconate and lactate salts effectively enhanced the nutritional quality of bread through magnesium fortification [77]. This study revealed significant benefits and nutritional improvements in bread quality through the use of specific magnesium salts. It also supports the public health sector, which aims to increase magnesium intake. It opens up opportunities for product development in the food industry to create bread products with added nutrients, making them more appealing to health-conscious consumers. Magnesium fortification is a crucial way to fight deficiencies that affect global health, which many people face today. This not only helps individuals meet their daily magnesium needs but also encourages food manufacturers to create innovative products that appeal to health-conscious customers. Prioritising magnesium fortification can be a powerful way to improve public health and support overall well-being in the communities, and helps ensure that everyone has access to the nutrients they need for a healthier life [105].

7.1.5. Iodine

Iodine is the heaviest micronutrient that the human body requires in trace amounts. It is essential for producing thyroid hormones; this element helps regulate metabolism and maintain bodily functions. Iodine is absorbed in the small intestine as iodide and transported by the bloodstream to the thyroid gland, where it is oxidised to iodine. The production of T4 and T3 hormones takes place. Excess iodine is excreted through urine. Iodine deficiency occurs when the food does not deliver sufficient iodine to the body. This deficiency leads to various effects termed Iodine Deficiency Disorders (IDDs), affecting both low-income and wealthier regions. The addition of iodine or iodides to food was futile, and so maintaining iodine in processed food has not been effective enough to eradicate the IDDs. Thus, the WHO recommended food fortification with iodine. Salt iodisation has been the best way to reduce IDDs. Potassium iodide is the commonly used iodine in fortification processes [78]; [82]. A study on food fortified with iodine-rich macroalgae and selenized yeast showed a sixfold increase in iodine and a threefold increase in selenium. The fortified fish increased iodine levels by more than 12% and also boosted other micronutrients [83]. The increase in iodine and selenium content can help address deficiencies without compromising the fish's quality, allowing consumers to enjoy the same taste with enhanced nutrition.

This solution offers a solution for the aquaculture sector to enhance the nutritional quality of fish products. Nutritionists can recommend these fortified fish as part of a healthier diet, helping people consume more nutritious food. An iodine fortification for bread salt was introduced in Australia in 2009 to address mild iodine deficiency. A study was done to find urinary iodine levels in pregnant women. Samples were collected in later years, and it was found that the urinary iodine level increased from 88 μg/L to 150 μg/L [84]. This study demonstrates that iodine fortification of bread has successfully increased iodine levels in pregnant women, thereby meeting their iodine needs. Enhanced iodine intake can support maternal and fetal health by reducing the complications that lead to nutrient deficiency [107]. This program would have increased awareness among pregnant women about the importance of iodine in their bodies for maintaining good health. Iodine fortification is a powerful way to combat iodine deficiency and support thyroid health, which is crucial for the metabolism and overall well-being of the human body. Adding iodine to everyday foods helps people obtain the necessary nutrients and reduces the risk of health issues associated with iodine deficiency. It is an effective solution to improve public health and ensure better nutrition for everyone.

7.2. Vitamins

Vitamins are essential organic compounds that the body cannot produce on its own, except for vitamin D, which is synthesised through exposure to sunlight [85]. All other vitamins are obtained from the diet to support the body's vital functions [86]. These nutrients are crucial for maintaining a healthy body. It acts as a coenzyme that regulates metabolism and maintains the body's overall well-being [87]. It plays a significant role in energy production, immune defence, and several physiological functions. Vitamins are classified as fat-soluble (D, E, A, and K) and water-soluble (C and B complexes). Vitamins are found in smaller quantities in natural foods, which can lead to vitamin deficiencies that affect people of all age groups and often occur in conjunction with mineral deficiencies [88]. These deficiencies can occur due to a poor diet, health issues that impair the body's ability to absorb nutrients, or increased nutrient requirements during periods such as pregnancy or growth. If they are not properly cared for, they can lead to serious health problems and affect normal bodily function. To overcome these problems, vitamin fortification has become a key solution. By adding essential nutrients to common foods, fortification helps meet nutritional needs, particularly in populations where there is a substantial risk of deficiencies.

This approach improves food quality and acts as a powerful tool to prevent diseases caused by vitamin deficiencies. Fortification plays a crucial role in building healthier communities. Boosting vitamin C content in jam is a vitamin fortification study where chia seeds were added to pineapple jam at distinct levels to increase the protein content. The best combination for increasing vitamin C and protein content was found by adding 50% chia seeds, cinnamon, and lemongrass oil [89]. This study reveals the benefits of incorporating essential oils and chia seeds into jam, enhancing its nutritional quality by increasing protein and vitamin C content. Lemongrass essential oil acts as a natural preservative, inhibiting microbial activity and thereby extending shelf life. This research promotes the development of foods that deliver extra health benefits. By using natural ingredients, the flavour and texture are enhanced, making it more appealing to consumers. Microencapsulation of vitamins is one such study where oil-soluble vitamins, i.e., vitamins A, D, and E, were microencapsulated from raw hen egg whites using ultrasound assistance. The free thiol groups were crucial in creating stable microcapsules with robust shells [90]. This method enhances the nutritional value and bioavailability of staple foods by improving the stability of vitamins. This method supports health-conscious consumers. This may reduce food waste by increasing nutrient retention.

Table 2: Metal and vitamin nanoparticles enhance growth, increase yields, and improve the nutritional quality of products, and proper usage ensures benefits while minimising risks from high concentrations

Metals	Form	Particle size	Optimized conc.	Fortified product	Enhancement	Toxic Trails	Ref.
Zinc Nanoparticles	ZnO	10-30 nm	20 ppm	Lentil seedlings	51-54%	> 20 ppm resulted in a decrease in dry and fresh weights	[92]
	$ZnSO_4$	10 ppm	Soil: 5–25 kg/ha Foliar: 2–5 g/L	Wheat	Grain zinc ↑ 83%	> 0.2 mg/kg Cd conc in soil affects zinc-regulated transporters	[93]
				Rice	27%		
				maize	9%		

	Zano (synthesised using brown algae extract)	31.4 nm	40 ppm	Rice grain	Grain yield ↑ 15.8% ($ZnSO_4$ + ZnO NP (40 ppm))	No significant toxicity observed with ZnO NPs concentrations up to 60 ppm	[94]
					17.4% ($ZnSO_4$ + ZnO NP (60 ppm))		
Zn chelate	Zn-citric, Zn-glycine & $ZnSO_4$	-	8 mg/kg	Lettuce	↑ 21% yield by Zn-GLY	The stem diameter increased	[95]
Iron Nanoparticles	Fe_3O_4 (synthesised from Glycyrrhiza glabra)	23.8 nm	150 ppm	Rice seeds	Grain yield ↑ by 204.05%	-	[96]
	Nano-chelated iron fertiliser	80 nm	2.5 g/L	Rice	↑ in yield by 27%	-	[97]
	Fe_2O_3	<50 nm	75 mg/L	Cucumber fruit	38.99%	100 mg/L, ↑ in Fe conc of leaves and fruits	[98]
Combination of Fe and Zn Nanoparticles	Zn-Bac	77 nm	250 ppm	Broccoli Microgreens	Fresh weight ↑ by 178%	No toxic symptoms identified	[99]
	Fe-Bac	68 nm			156.10%		
	Zn NPs	77 nm			150.40%		
	Fe NPs	68 nm			112.10%		
	Fe Bac + Fe NPs	68 nm			156.10%		
	Zn Bac + Zn NPs	77 nm			178%		
Calcium Nanoparticles	CaO	160 nm	75 ppm	Canola plants	↑ in yield by 35.18% ↑ in germination by 30%	100 ppm, slight ↓ in benefits without causing toxicity	[100]
Magnesium Nanoparticles	Mg $(OH)_2$ (synthesised from Aspergillus Niger)	50-100 nm	500 ppm	Zea mays	100% ↑ in germination, highest GRI (3 seeds/day)	>500 ppm affects the plant negatively	[101]
	$MgCl_2$ and $MgSO_4$	-	100 ppm	Snap beans	$MgSO_4$ has 78% Scavenging activity	>100 ppm increases toxins	[102]
	NanoMg (from Aspergillus brasiliensis)	<5.9 nm	200 ppm	Wheat plant	Grain ↑ by 38.7%, dry matter yield ↑ by 63.2%	>200 ppm disrupts plant physiology	[103]
	NanoMg	500-2000 nm	50 ppm	Green beans	25% ↑ in biomass	> 200 ppm showed	[104]

			100 ppm		62% ↑ in pods yield	phytotoxic effects	
Titanium Nanoparticles	TiO_2	length: 10-190 nm Width: 1-20 nm	1500 ppm	Tomato	↑ in yield by 27%	>1500 ppm causes local toxicity in plants	[105]
		20-30 nm	100 mg/L	Moldavian balm	↑ in fresh weight by 11.72%, ↑ in plant height by 28.87%	>200 mg/l caused aggregation in roots	[106]
Copper Nanoparticles	Cu NPs	21.86 nm	300 mg/L	Wheat	↑ in grain weight by 2.8%	700 mg/L & 950 mg/L, negative effect on physiological attributes	[107]
	Cu NPs	100-1000 nm	3 mg/L	Wheat	43.4% ↑ in yield wrt 60% control	>3 mg/L & >10 mg/L decreases growth parameters	[108]
	Ag NPs	<= 10 nm	10 mg/L				
	NanoCu	30-40 nm	69.4 µM	Maize plants	seed yield ↑ by +40%	No adverse effects reported	[109]
Vitamin Nanoparticles	Vitamin A encapsulated as Vitamin A Palmitate	25 µm	68.6%–68.7%	milk	4.19 ± 0.06% ↑ in dry weight	Unstable at >75% alcohol	[110]
	Vitamin D2 encapsulated as ergocalciferol.		1.2%–1.4%				
	Vitamin C encapsulated in chitosan	300 nm	420 mg/kg	Nile tilapia	206.1% relative growth rate	No health hazards observed	[111]
	Vitamin E encapsulated in chitosan		100 mg/kg				
	Vitamin C encapsulated in oleogel	-	-	Chewable gels	Vitamin C retention by ↑ 96.7%	No toxic traits were mentioned	[112]

	Vitamin K3	-	23.7 mg/kg	Chicken eggs	Egg MK-4 levels ↑ by 55.4%	A dose of 45.7 mg/kg caused a slight ↓ decrease in egg production.	[113]
Vitamin Fortification	Vitamin B12	-	Wheat flour: 50 µg/g of VB12 & VD3 Rice Bran Oil: 250 µg/g of VD3	Wheat flour & Rice bran oil	Maximum retention 91% in Chapatti	No toxicological risks mentioned	[114]
	Vitamin D3				Maximum retention 84% in Cake		
	Vitamin D3 & Vitamin D2	-	150 ng VD3 & VD2/g	Sunflower oil	Retention of 70-99%	No toxic concerns mentioned	[115]
			19.2 g/100 g VD2	Mushrooms	62-88%		
			7.5 g/100 g VD3	Rainbow trout	85-114%		

Research on vitamin A deficiency where yoghurt is fortified with pumpkin flesh to enhance carotene content. Three types of yoghurts were prepared for the study. Pumpkin pie spice and ground pumpkin seeds were added to the yoghurts [91]. The fortified yoghurts showed a prominent level of beta carotene. This method enhanced the nutritional value of yoghurt by increasing essential nutrients and reducing undesirable fats and carbohydrates. As appealing ingredients are added, the fortified yoghurt achieves high consumer demand. This research demonstrates the potential for creating fortified foods as an effective means to enhance public health and address nutritional deficiencies. Vitamin fortification helps improve public health by combating deficiencies and enhancing the nutritional value of food. Several food fortifications through vitamins have boosted nutrition and supported healthier lifestyles. It meets the demand for nutrient-rich products, promoting overall well-being in communities [115]. Table 2 presents a consolidated view of how different metal and vitamin nanoparticles enhance yield, bioavailability, and safety thresholds. The influence of diverse nanoparticles on crop nutrient content and growth performance is shown in Figure 4.

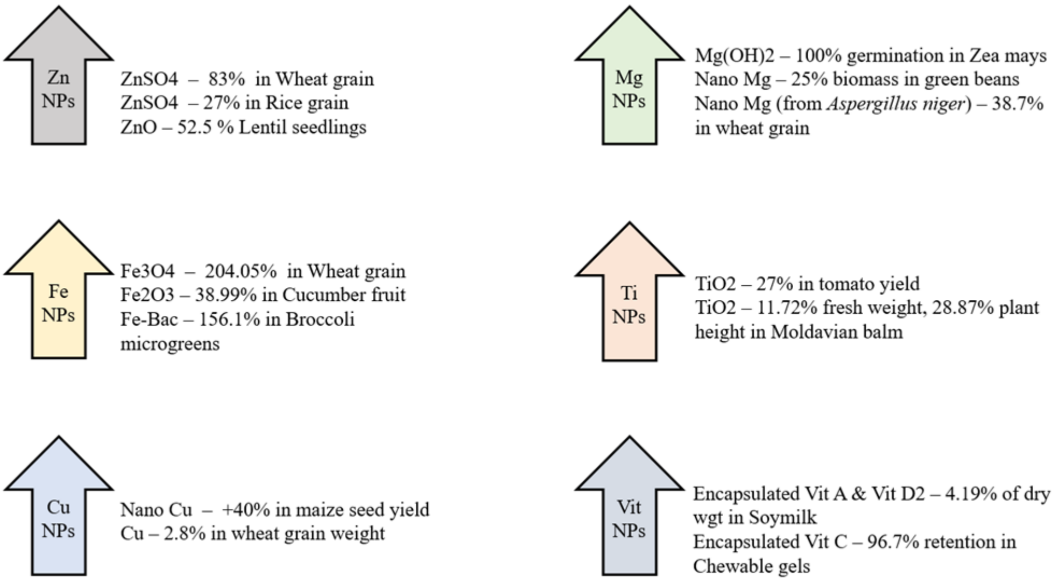

Figure 4: Effect of various nanoparticles on crop nutrient enhancement and growth parameters

8. Nanotoxicology

Nanotoxicology is the study of the potential health and environmental risks associated with nanoparticles, particularly in the context of biofortification. These nanomaterials can enhance the bioavailability and absorption of nutrients due to their distinct physicochemical properties, including a high surface area, tailored surface functionalities, and enhanced reactivity. However, these features also raise critical questions about how they interact with biomolecules in the human body, thereby determining their biological behaviour and the safety of the nanoparticles. Research indicates that nanoparticles may penetrate biological barriers, enter the bloodstream, and accumulate in various tissues. The consequences include potential oxidative stress, inflammation, and even genetic damage, which could result in serious long-term health effects. For example, metallic nanoparticles, such as silver and zinc oxide, are effective antimicrobial agents or nutrient carriers; however, they have been shown to induce cytotoxicity and DNA damage under certain conditions. These unintended consequences highlight the importance of comprehensive risk assessments, particularly since chronic exposure can lead to long-term health effects, even at low doses.

Also, there are growing concerns about the behaviour of these materials in the environment. These nanoparticles may disrupt the essential microbial ecosystems in the soil by interacting with them. This can alter the nutrient cycles, affect plant-microbe interactions, and potentially affect agricultural productivity. Recent studies also suggest that nanoparticles can influence plant metabolism and gene expression, sometimes leading to unpredictable physiological responses. Due to these multifaceted concerns, it is essential to conduct robust toxicological evaluations during the development of nanomaterials. This includes both in vitro and in vivo studies, as well as in silico tools such as quantitative structure-activity relationship (QSAR) models, physiologically based pharmacokinetic (PBPK) modelling, and molecular dynamics (MD) simulations. These computational methods enable researchers to predict and visualize nanoparticle interactions at the molecular level, reducing the need for extensive animal testing and accelerating the design of safer nanomaterials [116]; [121].

Beyond its application in food, nanotoxicology has become increasingly important in various fields, including medicine, cosmetics, and environmental remediation. In targeted drug delivery, nanoparticles enhance specificity but may elicit immune responses or accumulate in unintended tissues [117]. Similarly, in water treatment, engineered nanomaterials such as carbon nanotubes and nano-silver are highly effective but can harm aquatic ecosystems if not controlled [121]. These risks underscore the need for responsible innovation, supported by evolving regulatory frameworks. Strategies such as Safe-by-Design (SbD) and the precautionary principle are crucial for ensuring that nanomaterials are developed and used sustainably [119]; [120]. By conducting a thorough safety evaluation from the design phase and taking into account the full life cycle of nanotechnology, its advantages can be enhanced without adversely affecting human or environmental health.

9. Safety and Regulatory Aspects

Biofortification is a solution for the micronutrient deficiency challenge, but its implementation must be accompanied by severe safety testing and regulation. It must ensure consumer health, environmental sustainability, and regulatory compliance to work effectively. One of the primary safety concerns is the nutritional impact of biofortified crops. While increasing the amount of nutrients is better for health, excessive intake of certain micronutrients, such as iron and vitamin A, will lead to toxicity. Clinical trials and research provide safe nutrient levels and ensure absorption without adverse effects. The potential for allergenicity and genetic hazards must be thoroughly evaluated, especially when biofortification involves genetic modification. Regulatory agencies conduct strict assessments, including compositional analysis and allergenicity testing, to ensure food safety. Post-market monitoring is designed to assess the long-term health risks associated with biofortified food. Another crucial factor is environmental effects. The large-scale production of biofortified crops should be investigated for their potential to conserve biodiversity and improve soil health.

There is an issue of gene flow from genetically modified biofortified crop varieties to non-modified crops. Strict biosafety regulations, such as confined field trials and buffer zones, must be applied to minimize risks. Regulatory regimes differ between countries. The guidelines are provided by global organisations such as the Codex Alimentarius Commission, the WHO, and the FAO, while national authorities, including the FDA, EFSA, and FSSAI, conduct safety tests. Label regulations provide transparency, enabling consumers to make informed choices about biofortified foods. Public education and awareness are essential in promoting acceptance of these foods. Labelling and information campaigns help break down myths, such as those surrounding genetically modified biofortification. Secondary protection checks and monitors after the product enters the market to ensure continued safety and regulatory compliance [122]. It is essential to strike a practical balance between encouraging new developments, ensuring products are affordable, and ensuring that safety rules are followed. Safety and accessibility shouldn’t be sacrificed for advancements in nutrition and health. At the same time, strict regulations should not stop useful ideas from reaching the people who need them the most.

Particularly in low-income communities, where the benefits can be transformative, clear and fair guidelines can help ensure that innovative technologies are both safe and accessible to all. This balance is crucial in ensuring that progress in health and nutrition reaches all levels of society. To reach consumers, biofortified crops must pass through a clear process. Crop development begins with techniques such as gene editing, which produces Golden Rice with added vitamin A. The crops are then cultivated, harvested, and processed in factories to produce nutrient-rich products. These products are packaged and sent to stores for consumers. Examples include rice with vitamin A for better vision, wheat flour with iron to prevent anaemia, and milk with vitamin D for stronger bones. Other examples include cereals with zinc for immune health, iodised salt for thyroid support, and eggs or bread enriched with calcium or folic acid. This process ensures that biofortified foods provide real health benefits to people everywhere [122]. The complete pathway of biofortified crops from development and cultivation to processing, packaging, and distribution to consumers is systematically illustrated in Figure 5. This flowchart illustrates the integration of biotechnology, regulatory checkpoints, and consumer delivery to ensure that nutrient-rich food reaches target populations effectively.

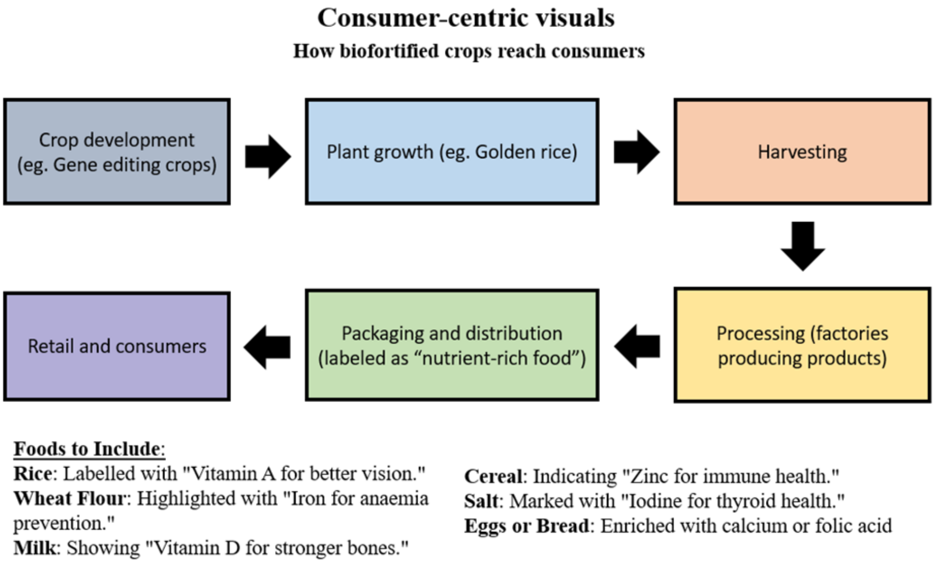

Figure 5: Process flowchart of the biofortified crops, from their growth to reaching consumers

10. Future Trends and Innovation

Future technologies and data-driven innovations are transforming the future of nutrient delivery, leading to the development of bioengineered nanoparticles designed to provide efficient solutions for enhancing nutrient bioavailability and ensuring targeted release. The use of artificial intelligence and big data marks a turning point in personal nutrition, enabling the development and tailoring of nutritional formulations to specific health profiles or individual requirements. Using machine learning, natural language processing, and tools for real-time health monitoring, these technologies consider individual health metrics, such as genetic predispositions, microbiome composition, dietary habits, and activity levels, to suggest highly specific therapeutic or nutritional interventions. For example, AI technology in maternal and pediatric health has enabled real-time monitoring of variables such as blood pressure, weight gain, and nutritional health, allowing patients to make informed decisions about their health and assisting doctors with early detection and preventive care planning.

Improved treatment outcomes, greater patient engagement, and reduced healthcare costs are among the numerous benefits of initial treatment. However, the risks cannot be ignored. AI systems may make inaccurate recommendations if they are trained on incomplete data. Additionally, the reliance on technology has raised concerns about data security, privacy, and the potential for unequal access, which could lead to health disparities. Therefore, AI has the potential to improve the accuracy and compassion of medicine. Its application needs to be careful, ethical, and inclusive. With these technological advancements, there is a growing focus on making nano-biofortified food production more sustainable, affordable, and environmentally friendly. All these trends together have the potential to change the way nutritional health is approached worldwide.

11. Conclusion

Micronutrient deficiencies pose a significant challenge, affecting billions of people worldwide, particularly in low- and middle-income countries, where access to diverse and nutrient-rich foods is often limited. These deficiencies can lead to numerous health issues like stunted growth in children, weakened immune systems, cognitive problems, and greater vulnerability to diseases. Fixing these nutritional gaps is a moral responsibility. Good nutrition is vital for everyone's well-being and development. One way to fight this hidden hunger is biofortification. This process involves adding crucial micronutrients to staple crops during their growth. This ensures that nutritious food reaches people who need it the most. Various methods, including traditional plant breeding and genetic engineering, have been proven to increase the number of vitamins and minerals in crops without altering their taste or appearance. This provides a practical pathway to eradicate malnutrition, especially in areas where fortified foods are not available.

In addition to biofortification, nanotechnology offers exciting opportunities in improving nutrient delivery. Nutrient delivery systems can be encapsulated to prevent vitamins and minerals from becoming toxic or degraded during food processing and storage. Nanocarriers, such as zinc oxide and iron nanoparticles, can also enhance nutrient bioavailability in the body, increasing the rate of absorption and targeting specific deficiencies. Nanotechnology can also aid in agricultural advancements by enhancing crop health, reducing dependency on chemical fertilisers, and promoting more sustainable farming practices. These technologies show great promise, but their implementation must be done carefully to ensure safety and fairness. The use of nanomaterials offers significant benefits but also poses potential risks, including toxicity, environmental impacts, and long-term health effects. Therefore, robust regulations are necessary to strike a balance between innovation and safety. It is also important to make these advances accessible to everyone, especially vulnerable groups like women and children who are more at risk of malnutrition. Without careful planning, there is a risk of creating deeper problems rather than bridging them.

The future of eating habits is evolving rapidly, driven by tools such as artificial intelligence and big data. Imagine being able to develop personalised diets based on someone's unique genetics, lifestyle, and health needs. This could lead us to better solutions that are much more effective. Improving nutrient delivery via bioengineered nanoparticles and advanced food technologies is already making it easier to deliver nutrients more efficiently. Governments, scientists, and industries must collaborate to reduce costs and make these solutions accessible to those who need them most. The combination of biofortification and nanotechnology is effectively fighting against malnutrition. This approach enables the provision of healthier food that is accessible, practical, and sustainable. By continuing to invest in current ideas, encouraging teamwork, and ensuring these solutions are equitable for everyone, the challenges of bringing these technologies to those in need can be addressed. A future where everyone has access to proper nutrition is essential for improving health and well-being. In addition to enhancing individual health, these advancements can help reduce poverty, improve education outcomes, and boost economic growth, creating a more equitable and sustainable world.

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