**An insight into the bio-fortification in pulses towards global nutritional security**

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**Abstract**

A sustainable method to enhance the nutritional profile of food crops is biofortification through plant breeding. The majority of the world's population relies on basic food crops, however the majority are deficient in important micronutrients. In several breeding projects during the past ten years, biofortification has gained relevance as a means of enhancing the nutritional profile of pulse crops. Pulses and legumes, particularly chickpeas, mungbeans, soybeans, and peas, are members of the Fabaceae family and are high in nutrients. Many diets rely heavily on pulses and legumes as a source of plant protein. Additionally, they have a low GI (glycemic index) and are a great source of complex carbs and dietary fiber. Due to the presence of several bioactive compounds—among which phenolic acids, flavonoids, and tannins make up the majority—pulses play a crucial role in metabolic and physiological processes. Despite being a good source of 15 vital minerals and vitamins, pulses and legumes have a limited bioavailability due to the presence of antinutrient agents. Breeding-based biofortification is a technique for improving the nutritional content of pulses and legumes.Iron, zinc, selenium, iodine, carotenoids, and folates have been the main micronutrients targeted. With the support of international partners, HarvestPlus has recently introduced a number of biofortified pulse crops, such as common beans and lentils, in developing nations, which has assisted in alleviating micronutrient deficiencies in the target population. This analysis will concentrate on recent research advancements and future plans for the biofortification of pulses.

 Keywords: Biofortification, pulse, malnutrition, sustainable achievements, plant growth prmoting organisms, transgenic, plant breeding

1. **Introduction**

 Malnutrition is a critical issue that impacts a large number of people in the majority of African and Asian nations. Due to the significant health issues it is associated with, it has attracted interest from throughout the world. Twenty million newborns are born every year with a low body weight. [1] Population increase is putting more strain on natural resources such as arable land, and this, combined with the environmental pressures brought on by climate change, is adversely affecting plant growth, productivity, and nutritional value. Additionally, the overuse of pesticides and fertilisers has a negative influence on the ecosystem, leading soil and water quality to decline [2 -5]. All of these challenges are reflected in human health, with malnutrition problems becoming more severe, not only in emerging economies but even in rich countries where food is plentiful but has low nutritional value.[6] The chemical residues in products are becoming a growing source of worry for consumers in industrialised nations, who are calling for more sustainable agriculture practises that prioritise safer products with no chemical residues. A quick response is required. In order to ensure the production of foods with a high nutritional value and a small environmental impact, society must find a sustainable way to address today's difficulties.

 A promising approach to meet the requirements previously outlined is biofortification. The World Health Organisation (WHO) defines it as "the process by which food crops' nutritional value is improved through agricultural practices, conventional plant breeding, or modern biotechnology."[7] In this context, agronomical practices try to increase the solubilization and mobilization of nutrients in the soil by applying fertilizer, hence promoting mineral absorption [8]. In underdeveloped nations where cereals and legumes are the primary food sources, legumes are an excellent target for improving the nutritional value of diets. The combination of legumes and their naturally colonized bacteria appears to be a potent one for a sustainable and eco-friendly strategy to combat the effects of climate change on crops and enhance plant nutrition. Legumes have a high nutritional value in addition to being crucial for the environment [9].

1. **Pulses and Legumes**

The Fabaceae family, which includes pulses and legumes, is rich in nutrients, particularly chickpeas, mungbeans, soybeans, and peas. In many diets, pulses and legumes provide a significant source of plant protein. They have a low GI (glycemic index) and are a great source of dietary fiber and complex carbs. Due to the presence of numerous bioactive chemicals, primarily phenolic acids, flavonoids, and tannins, pulses play a significant role in metabolic and physiological processes [14]. The word “pulse” is derived from a Latin word “puls or pultis” meaning thick slurry. Pulses belong to the Legume family, which has been consumed for thousands of years as a part of traditional diet throughout the world. Every pulse is a legume but not vice versa like dry peas, lentils, chickpeas, dry beans, etc are pulses whereas legumes include soybeans, peanuts, fresh peas etc. [10]. Legume is a type of plant that is described as a "pod or fruit" that contains seeds or dry grains and has the propensity to nitrogen in soil is fixed. In 2021, approximately 88.97 million metric tonnes of pulses were produced in world wide [11]. The estimated production of pulses in 2021-22 as per the Department of Agriculture and Farmers’ Welfare is 26.96 million tonnes [12]. In India 2021-22, 30.37 Mha area is under pulses with an average yield of 888 kg/ha [13].

Only 20 of the more than 1000 recognized varieties of legumes that are grown for human use, including chickpeas, are known to exist. Pigeon pea (*Cajanus cajan* L.), cowpea (*Vigna unguiculata*), mung bean (*Vigna radiata* L.), urdbean (*Vigna mungo* L.), lentil, and *Cicer arietinum* L. Field pea (*Pisum sativum* L.), moth bean (*Vigna aconitifolia*), horse gram (*Macrotyloma uniflorum*), French bean (*Phaseolus vulgaris*), soybeans (*Glycine max*), lathyrus (*Lathyrus sativus* L.), etc. are grown [15].

1. **The Contribution of Legumes to the Achievement of the Sustainable Development Goals**

Due to the well-known climate change phenomena, which includes global warming and a rise in the frequency of extreme weather events (heavy rains and drought), the world is currently experiencing significant agricultural losses in terms of productivity and nutritious crop value. As a result, over two billion individuals experience malnutrition issues as a result of food shortages (acute hunger) or inadequate intake of crucial micronutrients (hidden hunger) [16].

 Even though they are crucial for human nutrition and health, the production of food legumes has only seen a slight annual increase of 0.95%, fluctuating only between 40.78 and 70 million tonnes. Pulses per capita availability has been under extreme strain due to the slow expansion in world pulses output, expanding population, diversifying end-uses, and people's improved purchasing power [17]. In this setting, there has been an increase in the demand for sustainable agricultural methods to achieve food security, and so-called "climate-smart agriculture," which strives to use agricultural methods that are "resistant" to the effects of climate change and are environmentally benign, has arisen [19]. In order to increase the nutritious content of meals, fortification of foods during processing looks to be a promising method; nevertheless, not all populations can use this tactic [20].

 With a focus on the most underprivileged people, biofortification looks to be a workable strategy to increase the nutritional value of meals [18]. Legumes in particular are superior to or on level with cereals in terms of their supplies of vitamins, minerals, and amino acids [21]. They have a wide range of possible health advantages when added to the human diet. Some specific chemicals in legumes, like soluble fibers, antioxidants, flavonoids, etc., appear to be linked to a decreased risk of diabetes, cardiovascular disease, and some types of cancer, among other conditions.[22] Legumes contribute to the symbiotic N fixation process in the environment, which lowers the requirement for nitrogen fertilizer, lowers agricultural CO2 emissions, and improves soil quality [23]. In addition to providing the subsequent crops with pest and disease resistance [24], the introduction of legumes in crop rotation systems can cut greenhouse gas emissions by up to 25% [25].

**4. Nutrient content of Mungbean, peas and soyabean**

**4.1 Mungbean**

One of the most important pulse crops, mungbeans (Vigna radiata), are produced 90% of the world's supply in Asia. Indian is the greatest producer of mungbeans, accounting for 50% of global production, followed by China and Myanmar [26].

**4.1.1 Protein**

About 20 to 25 percent of the total dry weight of mungbean is made up of this high-protein food. Numerous storage proteins, including globulin (60%) and albumin (25%) are found in mungbean [27]. It contains significant amounts of the important amino acids phenylalanine (1.44%), leucine (1.85%), isoleucine (1.008%), valine (1.24%), tryptophan (0.26%), arginine (1.672%), methionine (0.286%), and lysine (1.66%). Histidine (0.7%) and threonine (0.78%) [28,29]. Mung bean protein has more in vitro digestibility than soybean protein (70%). (65%) [30].

**4.1.2 Carbohydrates**

It has a high carbohydrate content of 55–65%. Because mungbeans contain starch, which is a significant carbohydrate, they can be used to make starchy noodles [31]. The starch granule is oval, circular, or bean-shaped, and it ranges in diameter from 7 to 26 m [32]. It has significant cross-linkage properties. Less common sugars including raffinose, stachyose, and verbascose are to blame for flatulence in the diet. The oligosaccharides in mungbean, however, can be easily reduced by soaking, germination, and fermentation because they are soluble in water. Compared to other legumes, mung bean carbohydrates are more easily digested and less likely to induce flatulence [33].

**4.1.3 Lipids**

Mungbean contains just 2.1–2.5% oil, the majority of which is composed of linoleic acid (3.3–4.7 g/kg), palmitic acid (2.9–4 g/kg), oleic acid (2.2–2.9 g/kg), stearic acid (1.4–1.9 g/kg), and arachidic acid (0.23-0.3 g/kg) [34].

**4.1.4 Vitamins**

The amount of vitamins in one hundred grams of raw mungbean is ascorbic acid (4.9 mg), thiamin (0.62 mg), riboflavin (0.23 mg), niacin (2.25 mg), pantothenic acid (1.91 mg), vitamin B-6 (0.38 mg), folate (616 mg), choline (98 mg), beta-carotene (69 mg), vitamin A (115 IU), vitamin E (alpha-tocopherol) [35].

**4.1.5 Minerals**

The fact that it contains between 3.1 and 4 percent of ash [36,37 ,38] indicates that mungbean has a sizable amount of minerals. 3.5-4.5 mg/100g, 129-169 mg/100g, 8.9-13.2 mg/100g, 363-415 mg/100g, 80-114 mg/100g, and 1.3-2.1 mg/100g are the ranges for the minerals iron, magnesium, sodium, potassium, calcium, and zinc, respectively [39].

**5. Peas**

The pea (*Pisum sativum*), which is grown for both human and animal consumption, is one of the most significant crops ever produced. 17.5 million tonnes are produced worldwide each year of dry and green peas.. In 2021/22 China produced 11,459,352 tonnes of Green Peas. India is the world's second-largest Green Peas producer, with 5,846,000 tonnes, and first in acreage (1,426,896) [40-41].

**5.1 Protein:**

A range of environmental and genetic factors influence the protein content of peas, which ranges from 21.4% to 33.1%. it contains the majority of the protein. The nutritional value of storage proteins or globulins is determined by the amino acid composition. Higher levels of arginine are present in it. Compared to soybeans and lupin, the lupin has less glutamic acid, cysteine, valine, and methionine. Despite having a lower in vitro digestibility than soybean and other pulses due to the presence of protease inhibitors, raw pea nevertheless has a better in vitro digestibility [42].

**5.2 Carbohydrates**

In whole peas, the range of carbohydrate content is 56.7–74.0%, and in the kernel, it is 62.7–78.7%. The two kinds of glucose that make up starch are amylose and amylopectin. Rough-cut peas compared to smooth peas' (44–46%) reduced level of starch (28–38%) the wrinkle peas, however, have a greater (60–75.2%) amylose concentration. comparative to smooth peas (22–38%). Additionally, discovered is that sucrose Content is greater in wrinkled peas than in smooth peas [43]. A desire- The ratio of amylose to glucose directly affects a starch's ability. the reaction of postprandial hyper glycemia to amylopectin [44].

**5.3 Lipids**

Peas have a low lipid content of 0.8–6.2%. The percentages of total lipids in wrinkle seeds and round seeds are 4.5–5.3% and 2.8–3.2%, respectively. Re-evaluated fatty acid composition of peas (g/100g total fatty acids) reported to be made up of oleic acid, stearic acid, and palmitic acid (8.5–19.5g). (6.5-13.5g), linolenic acid (14.3-23.3g), and linoleic acid (38-70g) [45].

**5.4 Vitamins**

Ascorbic acid is one of the vitamins contained per kg in raw peas. Niacin (22-297 mg), thiamin (4.2–6.2 mg), and riboflavin (0.7–6.5 mg) Vitamin B-6 (1.2 mg), pyridoxine (10.5 mg), pantothenic acid (21.2 mg), and (1.1mg), beta-carotene (7.0g), biotin (8.3mg), and folic acid (0.7mg), among others. Inositol (1.6 mg), tocopherol (22–72 mg), and vitamin K (1.7 mg) [46].

**5.5 Minerals :**

Peas are good source of many important minerals such as calcium (0.3–1.5 g/kg DM), potassium (7.5–12.5 g/kg DM), phosphorus (2.2–5.3 g/kg DM), Magnesium (0.9–2.5 g/kg DM), iron (22–495 mg/kg DM), zinc (20.5–63.5 mg/kg DM), sodium (29.5–1500 mg/kg DM), copper (5–10.1 mg/kg DM) and manganese (8.7–140 mg/kg DM). Most of these minerals are present between the testa and the kernel; the latter is richer in calcium and potassium [47]. Because of high phytate content in peas, the bioavailability of Zn, Fe, and Ca are very low [48].

**6. Soyabean**

Soybean is recognized as one of the most important prospective protein sources for human nutrition under conditions of climate change and population growth. In 2021-22 the total world soyabean production goes to 385.524 million tonnes. With an annual production volume of 117,208,380 tonnes, the United States of America is the greatest soybean producer in the world. With 96,296,714 tonnes produced annually, Brazil comes in second. More than 60% of global production is produced jointly by the United States of America and Brazil. India is rated at #4 with 14,008,000 points [49-50].

**6.1 Protein**

Soybean has a protein level of between (36 and 56%) [5]. big in soybean Numerous proteins, including globulin and other storage proteins, the primary globulin constituents are glycinin (11S) and -conglycinins (7S). Although soy protein has few amino acids that include sulfur, contains all the amino acids necessary for proper human body operation, including Leucine, lysine, methionine, cysteine, phenylalanine, isoleucine, tyrosine, threonine, tryptophan, valine, and histidine [51].

**6.2 Lipids and Carbohydrates**

About 17–20% of the dry weight of the soybeans are made up of oil. Linoleic and linolenic acids, which are found in abundance in soybean oil, make it appropriate for human consumption [5]. According to estimates, the fatty acid composition of soybean oil consists of the following: palmitic acid (10.5–25%), stearic acid (2.7–30%), oleic acid (20.8-85%), linoleic acid (58.6%), and linolenic acid (5–17%). About 33% of the carbs in soybeans are fiber, which makes up the majority of those carbs. The two main soluble carbohydrates, accounting for (4-6%)of the total, are raffinose and stachyose. Dry matter base ranges from 1.4% to 4.1%. The four main types of insoluble carbohydrates are starch, cellulose, hemicellulose, and pectin are present in the soybean [52].

**6.3 Vitamins**

Both fat-soluble and water-soluble vitamins can be found in soybeans. The water-soluble vitamins are thiamin, riboflavin, niacin, pantothenic acid, and folic acid, while the four fat-soluble vitamins are A, D, E and K. Vitamin E (Tocopherol) content primarily relies on the kind of soybean. The quantity and the range of tocopherols in soybean is 10.9–28.5, 150–190, and on a dry matter basis, they are, respectively, 24.5-72.5 g/g [53].

**6.4 Minerals**

It has 5% ash in it. Significant minerals found in soybeans include potassium, the elements sodium, chloride, phosphorus, magnesium, calcium, and sulfur are present in from 0.2% and 2.1%, while the levels of silicon, iron, zinc, magnesium, copper, cobalt, cadmium, lead, arsenic, and chromium are between 0.01–145 ppm [54- 55].

**7. Approaches for Improvement of Nutritional Profile**

To improve the nutritional profile of crops and combat micronutrient insufficiency, many methods including dietary diversification, food supplements, food fortification, and biofortification are applied.

**7.1 Dietary diversification**

Dietary diversification is a food-based strategy that entails consuming a wide variety of foods, particularly varied plant-based meals including fruits, vegetables, and healthy grains. In order to increase the amount and bioavailability of micronutrients in food, dietary diversification also employs household-level tactics including food preparation that involves soaking, fermentation, and germination [56]. It is best to consume less foods high in anti-nutrients like phytic acid and polyphenols, which prevent the absorption of minerals, while consuming more fruits and vegetables high in promoter compounds like ascorbate and beta-carotene. Foods high in ascorbic acid (a promoter of iron absorption) should be ingested, for instance, to boost iron absorption [57-58].

**7.2 Food Fortification**

Fortification is the process of enhancing food's nutritional value by including vital micronutrients, such as vitamins and minerals. The World Food Programme (WFP) has several food aid programmes in place that use grains and pulses that have been partially pre-cooked, milled, and fortified with micronutrients overcome nutrient deficits and offer health advantages with little danger. To strengthen food in conjunction with iron, ferrous sulphate, ferrous fumarate, ferric pyrophosphate, and electrolytic iron powder Compounds are frequently employed [59].

**7.3 Biofortification**

The method of "biofortification" involves agronomic adjustments, genetic engineering, and traditional plant breeding to enhance the nutritional profile of plant-based foods (Fig 1).



Fig 1. Different approaches of biofortification for improvement of nutritional profile

**8. Agronomic Approaches**

Through the use of agronomic methods, biofortification can be accomplished through foliar fertilisation [60], the application of mineral fertilisers to the soil, and the introduction of beneficial microbes into the soil.

**8.1 Foliar fertilisation:**

Application of fertilisers directly to leaves is known as foliar fertilisation. It might be successful when mineral elements are not instantly available in the soil or are not easily transported to edible tissues [60,61]. Micronutrients Fe, Zn, and Se were added as foliar fertilisers to pulse crops to biofortify them. Application in multiple trials led to enhanced levels of these micronutrients in the harvested Fe concentration in cowpea seeds was found to be higher (29–33%). [63]. After applying Fe to the leaves of mungbeans, [64] scientists have found that the Fe concentration had increased (46%). In a similar manner, foliar application of Fe and Zn considerably raised the content of these minerals along with protein in seeds of cowpeas and chickpeas [65,66].

**8.2 Mineral Fertiliser :**

Mineral fertilisers are inorganic substances that contain necessary minerals and can be applied to the soil to increase the soil's micronutrient status and, consequently, the quality of the plants. The phytoavailability of minerals in the soil is frequently poor, thus to increase the concentration of minerals in the edible plant tissues, Mineral fertilisers must be applied with increased mineral solubility and mobility [67]. Plants can be strengthened with minerals using this technique, but not with organic substances like vitamins, which the plant itself makes. For Se, this approach was successfully used. I, and Zn, as these elements had good mobility in the soil as well as in the plant [68].

Regular fertilisation is usually necessary for biofortification, although this could be bad for the environment's health and could reduce the amount of other minerals that are available. Furthermore, the soil type in a particular region, variations in mineral mobility, and the Successful mineral chelation may also be hampered by the potential for antinutrient molecules to restrict mineral bioavailability putting this strategy into practise [69, 70 ].

**8.3 Plant growth promoting Organisms**

Beneficial soil microorganisms like rhizobia, mycorrhizal fungi, actinomycetes, and diazotrophic bacteria are symbiotically linked to plant roots and provide plants with a variety of benefits, including the availability and production of plant growth hormones and the promotion of nutrient mineralization. Although they occur naturally in the soil, inoculation or agricultural management methods can increase their numbers. To boost the phytoavailability of micronutrients, a variety of plant growth-promoting (PGP) soil microorganisms, such as Enterobacter, Bacillus, and Pseudomonas, can be utilised. These are mostly utilised as seed inoculants and promote plant growth by producing growth hormones, antibiotics, chitinases, siderophores, and inducing systemic resistance and mineralization [71].

 Increased amounts of Fe, Se, and Zn have been seen in numerous experiments using microorganism inoculants via mycorrhizal connections [72,73]. Additionally, it has been noted that the colonisation of legumes' roots and nodules with Pseudomonas sp., Brevibacterium sp., Bacillus sp., Enterobacter sp., and Acinetobacter sp. increases nitrogen fixation, plant growth, and grain yield.

In comparison to control (uninoculated) plants, chickpeas with PGP actinobacteria inoculation had higher seed mineral concentrations, including Fe (10–39%) and Zn (14–30%) [74]. Similar to this, the nutritional profile of chickpea grains was enhanced by arbuscular mycorrhizal fungi field inoculation by raising Fe and Zn concentration as well as yield and protein content [75]. Application of PGP rhizobacteria with Fe compound (FeSO4) in soil enhanced iron concentration in chickpeas (up to 80%) when compared to a control, according to Khalid et al. [76]. They also speculated that microbes may play a part in the extra uptake of Fe from soil when supplemented with Fe.

**9. Transgenic methods**

This technique has been used to generate biofortified soybean (*Glycine max*), common beans (*Phaseolus vulgaris*), lupines (*Lupinus angustifolius*), and many other pulse crops. This approach could need a significant initial investment, but if a transgenic variety is created and commercialized, it becomes a more dependable and sustainable method. Regarding important micronutrients like iron, zinc, and folate, this approach is still in development for pulses. Several limitations in genomic technologies have been overcome by the introduction of modern technologies like CRISPR/CAS9, ZFN (zinc finger nucleases), and To improve the nutritional profile of pulses, TALENs (transcription activator like effector nucleases) have been used to circumvent a number of genomic technology limitations. Additionally, desired bacterial pathways can be added using these gene editing tools to benefit metabolic engineering.

The capacity to apply this approach for multigene transfer, micronutrient enrichment, and the removal of antinutrients from edible tissues is made possible by the availability of fully sequenced genomes of pulse crops. Despite the fact that single gene modifications have no impact of target micronutrients in the plants [77].

**10. Genetic Engineering :**

When a particular micronutrient does not naturally exist in crops, variation in the desired traits is not available in the available germplasm, and/or modifications cannot be achieved by conventional breeding, biofortification through genetic engineering is an alternative strategy [78].

The recent availability of fully sequenced genomes in numerous crops encouraged this strategy. This strategy can simultaneously target the removal of antinutrients or the addition of promoters that can increase the bioavailability of micronutrients, in addition to boosting the concentration of micronutrients [12]. This method used genes from bacteria and other species in addition to those connected to several metabolic pathways used by plants [45].

Transgenic crop development demands a significant initial expenditure, however this could require a substantial investment during the initial stage and this is the best method to target the large populations especially in developing countries. To the best of our knowledge, there are no examples of transgenic pulse crops that have been biofortified with Fe, Zn, Se, I, carotenoids, or folates in the literature. However, a genetic engineering strategy has been used in pulse crops to improve another nutritional profile. Examples include the expression of a methionine-rich storage albumin from the Brazil nut in transgenic common bean plants (up to 22%) and the expression of a sunflower seed albumin gene in transgenic lupins (up to 95%). Similar to this, cowpeas were successfully modified using CRISPR/Cas9-mediated genome editing technology to stop the activation of the SNF gene. These results pave the path for the use of gene editing technologies for numerous features of interest in legumes.

**11. Plant breeding**

Limitations in the long-term effectiveness and sustainability of fertilizer approaches necessitates the development of economical and longstanding strategies for increasing micronutrient density in plants. Genetic engineering technology to produce genetically modified plants with desirable traits has been used in corn, rice, wheat, and soybeans. This can be an effective approach for crop improvement; however, political opposition to GMOs in many countries, a complex legal framework for the acceptance and commercialization of transgenic crops, along with expensive and time-consuming regulatory processes are the major limitations of this method [45].

HarvestPlus decided to take the lead in addressing micronutrient deficiencies through traditional plant breeding because of limitations on the use of genetically modified crops in several nations [42]. The health status of low-income individuals worldwide can be improved through biofortification through plant breeding [10]. Carotenoids, Fe, and Zn deficiency have all been addressed using this strategy [11].

Conventional plant breeding techniques can help both big populations and those who live in rural locations with little access to commercially available fortified foods [14]. Farmers can expand and multiply this strategy over years at essentially zero marginal cost after a one-time investment in plant breeding. Because recurrent expenses are cheap and germplasms can be obtained globally without harming production or health, biofortification is well-liked by the general population [17]. For the plant breeding strategy to biofortification to be successful, there needs to be genetic variety in the gene pool. Several studies have shown substantial variation in the concentration

of minerals and vitamins in various crops [34]. Screening a wide variety of germplasms can identify parental genotypes with high micronutrient concentration, and they can be used in crosses, genetic research, and the creation of molecular markers to aid marker-assisted selection in breeding. To find the genotype X environment interaction (G X E), promising lines might be examined at several sites [106]. After thorough regional testing over numerous places and several seasons, they can be submitted to national government authorities for testing for agronomic performance and released [45].

**12. Challenges in biofortification of pulses**

Due to the adoption of all the aforementioned solutions, the process of pulse biofortification has been effective in many regions, but there are still certain obstacles to overcome. Plants include antinutrients that are naturally occurring, which decreases the ability of micronutrients to be absorbed. Phytic acid was found, according to Biel et al. This lowers the iron and zinc bioavailability in pulses and legumes. Other Saponins, lathyrogens, protease inhibitors, and alpha-blockers are the inhibitors that are present. The presence of polyphenols as well as amylase inhibitors in legumes black bean seed coat's ability to absorb iron is hampered.

Another obstacle to bio-fortification is the limited genetic diversity of the plant gene pool and the lengthier period needed for research and cultivation. Due to poor marketing tactics and the low acceptance of transgenic pulses, the production of pulses has been viewed as a cashless crop, and many farmers have migrated to other crops. Yield and the nutritional composition of harvested pulses are also impacted by biotic and abiotic stress.

The nutritional profile is further impacted by post-harvesting procedures since many micronutrients are removed during milling and polishing. Collaboration between plant breeders, nutritionists, genetic engineers, and marketing specialists is crucial to solving these issues.Both conventional and genetic engineering methods call for the expansion of germplasm conservation for better characterisation and screening of pulses varieties. A single locus multinutrient characteristic aids in the qualitative enhancement of pulses [45].

**13. Current status of biofortification in pulses**

Prof. M.S. Swaminathan, a great agriculturalist and visionary, has previously stated that nutritional security should be discussed instead of food security. He has also emphasised the importance of including all essential micronutrients in every person's diet as opposed to diets that are high in calories and protein. This can only be accomplished by partnering with agriculture, health, and nutrition all at once (Yadava et al., 2018). As a result, the idea of biofortification supported the need for a tool to combat malnutrition in the current situation.

Breeding-led biofortified crops have, among other methods of biofortification, improved crop nutrition profiles over time. With regard to biofortified crops developed through breeding, cereals account for 58.1% of the total, followed by legumes at13.3%, vegetables at 19.8%, and fruits at 9%. In addition to this, an agronomic approach was used to distribute cereals at a rate of 69.5%, legumes at a rate of 14.5%, oilseeds at a rate of 3.2%, vegetables at a rate of 11.3%, and fruits at a rate of 1.6%. The transgenic method of biofortification produced 44% of the biofortified crops in cereals, 10.3% in legumes, 12.6% in oil seeds, 17.7% in vegetables, 9% in fruits, and 2.3% in fodder [42].

By 2030, it is intended to provide more than one billion people with access to biofortified crops and the varieties of those crops so they can benefit from them in terms of nutrition and agronomy. Among the 12 biofortified crops, cowpea (Fe and Zn), lentils (Fe and Zn), and common beans (Fe) are frequently sold and consumed as biofortified crops under the category of pulses [1]. In India, 17 biofortified varieties in 8 crops, namely, 2 each for rice, wheat, pearl millet (*Pennisetum glaucum*), mustard (*Brassica* spp.), and sweet potato (Ipomoea batatas); 4 in maize (*Zea mays*) and one each for lentil, cauliflower (*Brassica oleracea* var. *botrytis*), and pomegranate (*Punica granatum*) have been released for its cultivation. Out of eight biofortified crops, one variety of lentil called "Pusa Ageti Masoor" was developed that is high in Fe [44]. On October 16, 2020, in honour of the Food and Agriculture Organization's (FAO) 75th anniversary, the Indian Prime Minister dedicated these varieties. In the world, 466 varieties with different traits were developed through mutational breeding in the legume and pulse categories. 54 mutant varieties were made available for the cultivation of pulse crops in India, including 16 in mung beans, 10 in cowpeas, 8 in pigeon peas, 8 in urd beans, 6 in chickpeas, 2 in lentils, and one each in peas, common beans, moth beans, and hyacinth beans [45].

**14. Future prospects**

Government policies governing food security vary from one nation to the next in order to secure the lives of those in need by giving them access to food grains. Despite this, nutritional security is now being discussed more than food security because it promotes a healthy lifestyle among the populace and serves as a weapon against malnutrition. In order to address that, the following strategies should be implemented in the future:

* The development of technologies and products ought to be economical and have long-term effects, given that the majority of populations suffer from Fe and Zn deficiency.
* The implementation and strengthening of a fast-track breeding programme for the creation of nutritionally dense varieties in accordance with the agro-climatic conditions and dietary preferences of residents of particular areas and or regions.
* Investigate and pinpoint potential genome editing technologies as well as molecularly-based nontransgenic reverse technology for the creation of trait-specific (biofortified) pulse crop varieties.
* Due to the presence of phytic acid, research should concentrate more on the bioavailability of micronutrients in pulses, particularly Fe and Zn.
* Consumption of biofortified crops would be encouraged by the advancement of technological infrastructures and market chains from growers to consumers.
* It would be very helpful to the breeder to identify such landraces or wild/primitive cultivars that have good micronutrient contents in order to create new varieties.
* The creation of novel plant species that are incredibly effective at absorbing, transporting, and accumulating micronutrients in edible plant parts. Identification of target genes or pathways that promote the synthesis of micronutrients and improve the efficiency of their conversion in pulse crops is also important
* Distribution of these developed varieties to farmers for commercialization.
* A separate MSP should be implemented by the governments for the bio-fortified crops. By taking this action, farmers would be more likely to grow biofortified crops and make a greater profit from them

**15. Conclusion**

One in three people worldwide lacks micronutrients, which are crucial for human growth and development. The most affordable and long-lasting method of addressing micronutrient deficiencies is thought to be biofortification through plant breeding. deficiencies. This strategy is widely accepted and has the potential to reach individuals living in Remote rural areas with little access to commercially marketed fortified foods. Further, Farmers can multiply seeds over years for practically nothing with a one-time investment. Marginal price. With the introduction of numerous biofortified products, significant advancements have been made recently. Crop varieties that are assisting the target populations in overcoming micronutrient deficiencies. A better nutritional profile for pulse crops will lead to a significant increase in consumption because they are a significant source of protein and energy. The last ten years have seen a rise in biofortification efforts to enhance the nutritional profile of pulse crops. But if the use of biofortified foods is to be effectively maximised, there are a number of issues that must be resolved.

**Reference**

[1] WHO (World Health Organisation), Global Nutrition Report, 2018 (2020, February

9). https://globalnutritionreport.org/reports/global-nutrition-report-2018/

executive-summary/.

[2-5] J.S. Kaushik, M. Narang, A. Parakh, Fast food consumption in children, Indian

Pediatr. 48 (2011) 95–101.

[3] T. Farzana, S. Mohajan, T. Saha, M. Hossain, M. Haque, Formulation and nutritional

evaluation of a healthy vegetable soup powder supplemented with soy flour,

mushroom, and moringa leaf, Food Sci. Nutr. 5 (4) (2017) 911–920.

[4] FAO (Food and Agriculture Organisation of the United Nation), FAOSTAT(2016),

2020. February 9, http://www.fao.org/faostat/en/#data/QC/visualize.

[5] S.F. O'Keefe, L. Bianchi, J. Sharman, Soybean Nutrition, 2015.

[6] K. Liu, Soybeans as a powerhouse of nutrients and phytochemicals, Soyabeans

Funct. Foods Ingred. (2004) 1–22

[7] G.J. Guzman, P.A. Murphy, Tocopherols of soybean seeds and soybean curd (tofu),

J. Agric. Food Chem. 34 (5) (1986) 791–795.

[8] K.S. Liu, Soybeans: Chemistry, Technology, and Utilization, Klewer Academic

Publishers, 1999.

[9] Soares, J.C.; Santos, C.S.; Carvalho SM, P.; Pintado, M.M.; Vasconcelos, M.W. Preserving the Nutritional

Quality of Crop Plants under a Changing Climate: Importance and Strategies. Plant Soil 2019, 443, 1–26.

[CrossRef]

[10] M. Asif, L.W. Rooney, R. Ali, M.N. Riaz, Application and opportunities of pulses in

food system: a review, Crit. Rev. Food Sci. Nutr. 53 (11) (2013) 1168–1179.

[11https://www.statista.com/statistics/721945/pulses-production-volume-worldwide/#:~:text=In%202021%2C%20approximately%2088.97%20million,million%20metric%20tons%20from%202020.

[12]https://pib.gov.in/PressReleseDetailm.aspx?PRID=1808671#:~:text=The%20estimated%20production%20of%20pulses,DA%26FW)%20is%2026.96%20million%20tonnes

[13] Biofortification of pulses and legumes to enhance nutrition

Shishir Kumar, Geetanjali Pandey

Heliyon 6 (3), 2020

[15] B.N. Rao, Pulses and legumes as functional foods, NFI Bull. 23 (1) (2002) 1–4.

[16] Campbell, B.M.; Thornton, P.; Zougmoré, R.; van Asten, P.; Lipper, L. Sustainable Intensification: What Is Its Role in Climate Smart Agriculture? Curr. Opin. Environ. Sustain. 2014, 8, 39–43. [CrossRef]

[17] https://link.springer.com/chapter/10.1007/978-81-322-2716-8\_4

[18] –Carvalho, S.M.P.; Vasconcelos, M.W. Producing More with Less: Strategies and Novel Technologies for

Plant-Based Food Biofortification. Food Res. Int. 2013, 54, 961–971. [CrossRef]

Jha, A.B.; Warkentin, T.D. Biofortification of Pulse Crops: Status and Future Perspectives. Plants 2020, 9, 73.

[CrossRef]

[19] –Campbell, B.M.; Thornton, P.; Zougmoré, R.; van Asten, P.; Lipper, L. Sustainable Intensification: What Is Its

Role in Climate Smart Agriculture? Curr. Opin. Environ. Sustain. 2014, 8, 39–43. [CrossRef]

[20]

Rehman, H.M.; Cooper, J.W.; Lam, H.-M.; Yang, S.H. Legume Biofortification Is an Underexploited Strategy

for Combatting Hidden Hunger. Plant. Cell Environ. 2019, 42, 52–70. [CrossRef]

[21]

Rehman, H.M.; Cooper, J.W.; Lam, H.-M.; Yang, S.H. Legume Biofortification Is an Underexploited Strategy

For Combatting Hidden Hunger. Plant. Cell Environ. 2019, 42, 52–70. [CrossRef]

[22]

Martín-Cabrejas, M. Legumes: Nutritional Quality, Processing and Potential Health Benefits. In Legumes and

Their Associated Health Benefits; Royal Society of Chemistry: London, UK, 2019.

Ferreira, H.; Vasconcelos, M.; Gil, A.M.; Pinto, E. Benefits of Pulse Consumption on Metabolism and Health:

A Systematic Review of Randomized Controlled Trials. Crit. Rev. Food Sci. Nutr. 2020, 1–12. [CrossRef]

[23]

Karkanis, A.; Ntatsi, G.; Lepse, L.; Fernández, J.A.; Vågen, I.M.; Rewald, B.; Alsin, a, I.; Kronberga, A.;

Balliu, A.; Olle, M.; et al. Faba Bean Cultivation—Revealing Novel Managing Practices for More Sustainable

And Competitive European Cropping Systems. Front. Plant Sci. 2018, 9, 1115. [CrossRef]

[24] Atienza, S.G.; Rubiales, D. Legumes in Sustainable Agriculture. Crop Pasture Sci. 2017, 68, i–ii. [CrossRef]

[25]

Ma, Y.; Schwenke, G.; Sun, L.; Liu, D.L.; Wang, B.; Yang, B. Modeling the Impact of Crop Rotation with

Legume on Nitrous Oxide Emissions from Rain-Fed Agricultural Systems in Australia under Alternative

Future Climate Scenarios. Sci. Total Environ. 2018, 630, 1544–1552. [CrossRef]

[26]

 C.J. Lambrides, I.D. Godwin, Mungbean. In Pulses, Sugar and Tuber Crops, 2007,

pp. 69–90.

[27] -13 [13] K. Ganesan, B. Xu, A Critical Review on Phytochemical Profile and Health

Promoting Effects of Mung Bean (Vigna Radiata), Food Science and Human

Wellness, 2017.

[28] – A.E. Mubarak, Nutritional composition and antinutritional factors of mung bean

seeds (Phaseolus aureus) as affected by some home traditional processes, Food

Chem. 89 (4) (2005) 489–495.

[29] United States Department of Agriculture (USDA), Agriculturalresearch Service,

National Nutrient Database for Standard Reference Release, 2016 (2020, February

9). nutrient database laboratory home page:https://ndb.nal.usda.gov/ndb/search

/list.

[30]- R.M. Nair, R.Y. Yang, W.J. Easdown, D. Thavarajah, P. Thavarajah, J.D.A. Hughes,

J.D.H. Keatinge, Biofortification of mungbean (Vigna radiata) as a whole food to

enhance human health, J. Sci. Food Agric. 93 (8) (2013) 1805–1813.

[31] – K. Ganesan, B. Xu, A Critical Review on Phytochemical Profile and Health

Promoting Effects of Mung Bean (Vigna Radiata), Food Science and Human

Wellness, 2017.

[32]- C.P. Marinangeli, A.N. Kassis, P.J. Jones, Glycemic responses and sensory

Characteristics of whole yellow pea flour added to novel functional foods, J. Food

Sci. 74 (2009) S385–S389.

[33]- R.M. Nair, R.Y. Yang, W.J. Easdown, D. Thavarajah, P. Thavarajah, J.D.A. Hughes,

J.D.H. Keatinge, Biofortification of mungbean (Vigna radiata) as a whole food to

enhance human health, J. Sci. Food Agric. 93 (8) (2013) 1805–1813.

 [34]- K. Ganesan, B. Xu, A Critical Review on Phytochemical Profile and Health

Promoting Effects of Mung Bean (Vigna Radiata), Food Science and Human

[35]- K. Ganesan, B. Xu, A Critical Review on Phytochemical Profile and Health

Promoting Effects of Mung Bean (Vigna Radiata), Food Science and Human

[36] -United States Department of Agriculture (USDA), Agriculturalresearch Service,

National Nutrient Database for Standard Reference Release, 2016 (2020, February

9). nutrient database laboratory home page:https://ndb.nal.usda.gov/ndb/search

/list.

[37]- M. Kurniadi, C.D. Poeloengasih, A. Frediansyah, A. Susanto, Folate content of mung

bean flour prepared by various heat-treatments, Procedia Food Sci. 3 (2015) 69–73. [38]- 18

[38]- P.K. Dahiya, A.R. Linnemann, M.J.R. Nout, M.A.J.S. Van Boekel, R.B. Grewal,

Nutrient composition of selected newly bred and established mung bean varieties,

LWT - Food Sci. Technol. 54 (1) (2013) 249–256.

[39] -P.K. Dahiya, A.R. Linnemann, M.J.R. Nout, M.A.J.S. Van Boekel, R.B. Grewal,

Nutrient composition of selected newly bred and established mung bean varieties,

LWT - Food Sci. Technol. 54 (1) (2013) 249–256.

[40 41]

FAO (Food and Agriculture Organisation of the United Nation), FAOSTAT(2016),

2020. February 9, http://www.fao.org/faostat/en/#data/QC/visualize.

https://divercitytimes.com/agriculture/crops-and-livestock/green-peas.php#:~:text=China%20is%20the%20top%20country,first%20in%20acreage(1%2C426%2C896)

[42] W.J. Dahl, L.M. Foster, R.T. Tyler, Review of the health benefits of peas (Pisum

sativum L.), Br. J. Nutr. 108 (S1) (2012) S3–S10.

[43]

G.P. Savage, S. Deo, The nutritional value of peas (Pisum sativum). A literature

Review, Nutr. Abstr. Rev. 59 (No. 2) (1989) 66–88.

[44]

W.J. Dahl, L.M. Foster, R.T. Tyler, Review of the health benefits of peas (Pisum

sativum L.), Br. J. Nutr. 108 (S1) (2012) S3–S10.

[45]

 G.P. Savage, S. Deo, The nutritional value of peas (Pisum sativum). A literature

review, Nutr. Abstr. Rev. 59 (No. 2) (1989) 66–88.

[46]

G.P. Savage, S. Deo, The nutritional value of peas (Pisum sativum). A literature

review, Nutr. Abstr. Rev. 59 (No. 2) (1989) 66–88.

[47]

G.P. Savage, S. Deo, The nutritional value of peas (Pisum sativum). A literature

review, Nutr. Abstr. Rev. 59 (No. 2) (1989) 66–88.

[48]

W.J. Dahl, L.M. Foster, R.T. Tyler, Review of the health benefits of peas (Pisum

sativum L.), Br. J. Nutr. 108 (S1) (2012) S3–S10.

[49 50]

https://www.sopa.org/statistics/world-soybean-production/

https://www.atlasbig.com/en-in/countries-by-soybean-production#:~:text=United%20States%20of%20America%20is,14%2C008%2C000%20is%20ranked%20at%204

[51]

 S.F. O’Keefe, L. Bianchi, J. Sharman, Soybean Nutrition, 2015.

[52] K. Liu, Soybeans as a powerhouse of nutrients and phytochemicals, Soyabeans

Funct. Foods Ingred. (2004) 1–22.

[53] G.J. Guzman, P.A. Murphy, Tocopherols of soybean seeds and soybean curd (tofu),

J. Agric. Food Chem. 34 (5) (1986) 791–795.

[54] K.S. Liu, Soybeans: Chemistry, Technology, and Utilization, Klewer Academic

Publishers, 1999.

[55]-B.L. O’Dell, Effect of Soy Protein on Trace mineral Availability, 187, Academic

Press, New York, 1979.

[56] [57] [58]

. Gibson, R.S.; Hotz, C. Dietary diversification/modification strategies to enhance micronutrient content and

bioavailability of diets in developing countries. Br. J. Nutr. 2001, 85, S159–S166. [CrossRef] [PubMed]

. Hurrell, R. How to ensure adequate iron absorption from iron-fortified food. Nutr. Rev. 2002, 60, S7–S15.

[CrossRef]

. World Health Organization and Food and Agriculture Organization. Vitamin and Mineral Requirements in

Human Nutrition, 2nd ed.; WHO: Geneva, Switzerland, 2004.

[59]-World Health Organization and Food and Agriculture Organization of the United Nations. Guidelines on

Food Fortification with Micronutrients; Allen, L., de Benoist, B., Dary, O., Hurrell, R., Eds.; WHO: Geneva,

Switzerland, 2006.

[60]-

White, P.J.; Broadley, M.R. Biofortification of crops with seven mineral elements often lacking in human

diets-iron, zinc, copper, calcium, magnesium, selenium and iodine. New Phytol. 2009, 182, 49–84. [CrossRef]

[PubMed]

[61]-

White, P.J.; Broadley, M.R. Biofortification of crops with seven mineral elements often lacking in human

diets-iron, zinc, copper, calcium, magnesium, selenium and iodine. New Phytol. 2009, 182, 49–84. [CrossRef]

[PubMed]

[62]-

Garg, M.; Sharma, N.; Sharma, S.; Kapoor, P.; Kumar, A.; Chunduri, V.; Arora, P. Biofortified crops generated

by breeding, agronomy, and transgenic approaches are improving lives of millions of people around the

world. Front. Nutr. 2018, 5, 12. [CrossRef] [PubMed]

[63]-Márquez-Quiroz, C.; De-la-Cruz-Lázaro, E.; Osorio-Osorio, R.; Sánchez-Chávez, E. Biofortification of cowpea

beans with iron: Iron’s influence on mineral content and yield. J. Soil Sci. Plant Nutr. 2015, 15, 839–847.

[CrossRef]

[64]- Ali, B.; Ali, A.; Tahir, M.; Ali, S. Growth, Seed yield and quality of mungbean as influenced by foliar

application of iron sulfate. Pak. J. Life Soc. Sci. 2014, 12, 20–25.

[65,66]-1 Salih, H.O. Effect of foliar fertilization of Fe, B and Zn on nutrient concentration and seed protein of Cowpea

“Vigna unguiculata”. IOSR J. Aric. Vet. Sci. 2013, 6, 42–46. [CrossRef]

. Nandan, B.; Sharma, B.C.; Chand, G.; Bazgalia, K.; Kumar, R.; Banotra, M. Agronomic fortification of Zn and

Fe in chickpea an emerging tool for nutritional security—A global perspective. Acta Sci. Nutr. Health 2018

[67]-

White, P.J.; Broadley, M.R. Biofortification of crops with seven mineral elements often lacking in human

diets-iron, zinc, copper, calcium, magnesium, selenium and iodine. New Phytol. 2009, 182, 49–84. [CrossRef]

[PubMed]

[68]-

White, P.J.; Broadley, M.R. Biofortification of crops with seven mineral elements often lacking in human

diets-iron, zinc, copper, calcium, magnesium, selenium and iodine. New Phytol. 2009, 182, 49–84. [CrossRef]

[PubMed]

[69]-

Frossard, E.; Bucher, M.; Machler, F.; Mozafar, A.; Hurrell, R. Potential for increasing the content and

bioavailability of Fe, Zn and Ca in plants for human nutrition. J. Sci. Food Agric. 2000, 80, 861–879. [CrossRef]

[70]- Ismail, A.M.; Heuer, S.; Thomson, M.J.; Wissuwa, M. Genetic and genomic approaches to develop rice

germplasm for problem soils. Plant Mol. Biol. 2007, 65, 547–570. [CrossRef] [PubMed]

[71]-

. Mahaffee, W.F.; Kloepper, J.W. Applications of plant growth-promoting rhizobacteria in sustainable

Agriculture. In Soil Biota: Management in Sustainable Farming Systems; Pankhurst, C.E., Doube, B.M.,

Gupta, V.V.S.R., Grace, P.R., Eds.; CSIRO: Melbourne, Australia, 1994; pp. 23–31.

[72]-

 Rengel, Z.; Batten, G.D.; Crowley, D.D. Agronomic approaches for improving the micronutrient density in

Edible portions of field crops. Field Crop. Res. 1999, 60, 27–40. [CrossRef]

 Smith, S.E.; Read, D.J. Mycorrhizal Symbiosis, 3rd ed.; Elsevier: London, UK, 2007.

[73]- Cavagnaro, T.R. The role of arbuscular mycorrhizas in improving plant zinc nutrition under low soil zinc

Concentrations: A review. Plant Soil 2008, 304, 315–325. [CrossRef]

[74]-

133. Sathya, A.; Vijayabharati, R.; Srinivas, V.; Gopalakrishnan, S. Plant growth-promoting action-bacteria on

chickpea seed mineral density: An upcoming complementary tool for sustainable biofortification strategy.

3 Biotech 2013, 6, 138. [CrossRef]

[75]- Pellegrino, E.; Bedini, S. Enhancing ecosystem services in sustainable agriculture: Biofertilization and

biofortification of chickpea (Cicer arietinum L.) by arbuscular mycorrhizal fungi. Soil Biol. Biochem. 2014, 68,

429–439. [CrossRef]

[76]- Khalid, S.; Asghar, H.N.; Akhtar, M.J.; Aslam, A.; Zahir, Z.A. Biofortification of iron in chickpea by plant

growth promoting rhizobacteria. Pak. J. Bot. 2015, 47, 1191–1194.

[77]-

https://www.frontiersin.org/articles/10.3389/fsufs.2020.571402/full