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Advances in Biofortification of Vegetables to Combat Malnutrition

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Abstract

Vitamin and mineral deficiencies, known as "hidden hunger," present significant challenges to human health and economic development, affecting roughly one-third of the global population. These deficiencies, particularly in zinc, iron, provitamin A, and iodine, are major contributors to malnutrition. In 2020, undernutrition was linked to approximately 45% of deaths among children under five. Despite adequate caloric intake, many individuals lack essential nutrients. Bio-fortification, the process of enriching food crops with essential vitamins and minerals through plant breeding or agronomic practices, offers a viable solution to this problem. This approach aims to improve nutritional intake, particularly in rural areas, and enhance nutrient security. Plant breeders and biotechnologists are overheading a new Green Revolution to produce more nutrient-dense crops. There are three primary methods of bio-fortification: agronomic, conventional, and transgenic. Recent advancements have led to the development of transgenic crops with enhanced provitamin A levels, including varieties such as Pusa Betakesari, Bhu Sona, Bhu Krishna, Kufri Neelkanth, and Pant Lobia-1 & Pant Lobia-2. The ultimate goal of bio-fortification is to mitigate nutrient deficiencies and improve overall health and nutrition.

1. Introduction

Vegetables are a vital component of a healthy diet, providing essential vitamins, minerals, dietary fibre, and a wealth of health-promoting phytochemicals. Beyond their nutritional value, vegetables add vibrancy and visual interest to our plates. The captivating colours of vegetables are a testament to the presence of a diverse range of pigments, each with its own unique contribution to plant physiology and human health. As world is currently facing food crisis making it more threatening, the human body is exposed to malnutrition. Malnutrition can be defined as any health condition that develops when the human body does not get all its needed nutrients, in other words malnutrition starts to develop when the body is deprived of all the important nutrients such as vitamins, minerals and other essential

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nutrients that it needs to maintain healthy tissues and proper organ function (Luo et al., 2015)

This chapter delves into the fascinating world of breeding for colour development in vegetable crops. We will explore the significance of pigments in plants, their connection to human health, and the various breeding methodologies employed to achieve vibrant colours in vegetables. The chapter will provide a comprehensive overview of current approaches and achievements in this field, examining both traditional and advanced breeding techniques. Finally, we will discuss the future directions of breeding for colour development, highlighting the potential for integrating cutting-edge technologies and addressing consumer preferences.

1.1. Significance of Pigments in Vegetables

The captivating colours of vegetables arise from a diverse range of pigments naturally synthesized within the plant. These pigments not only enhance visual appeal, attracting consumers and pollinators, but also play crucial roles in plant protection and stress response. Here's a closer look at the major classes of pigments responsible for the vibrant hues of vegetables:

1.0.1. **Carotenoids:** Carotenoids are lipophilic, organic pigments. (Cazzonelli et al., 2010). They are a class of more than 750 naturally occurring pigments. These richly colored molecules are the sources of the yellow, orange, and red colors of many plants. Fruit and vegetables provide most of the 40% to 50% carotenoids found in the human diet. (Cazzonelli et al., 2010).

Importance

1. Significance of Provitamin A and Carotenoids in Developing Countries
2. Fertility and Reproductive Success
3. Prevention of Oxidative Stress and Inflammation
4. Vision and Diseases of the Eye
5. Cognitive Decline and Alzheimer's Disease
6. Cancer Prevention and Treatment
7. Metabolic Syndrome, Obesity, Cardiovascular Disease, and Diabetes

1.0.2. **Anthocyanins:** Anthocyanidin backbones: Cyanidin, malvidin, delphinidin, peonidin, petunidin, and pelargonidin. Antioxidants are scavenging free radicals. Natural colorants in foods and beverages. Protection from cardiovascular disease and cancer. Unstable and sensitive to temperature and radiation. (Krinsky et al., 2005).

1.0.3. **Betalains:** Betalains are a class of water-soluble pigments that are found only in the order Caryophyllales. Betalains differ from anthocyanins in the chemical structures but share similarities to anthocyanins in the color spectra, biological functions. For example, betalains contain nitrogen but anthocyanins do not. Similarly, they are also localized in vacuoles. (Chen, 2005).

1.0.4. **Flavonoids:** Flavonoids are a class of polyphenolic secondary metabolites found in plants, and thus commonly consumed in the diets of humans. They are phytonutrients and have beneficial anti-inflammatory effects.

The specific pigments present in a vegetable and their relative abundance determine its overall colour. For example, the vibrant orange colour of a carrot is primarily due to the presence of beta-carotene, a specific type of carotenoid. Conversely, the deep purple colour of an eggplant arises from a high concentration of anthocyanins. Interestingly, the interplay between these pigments can create a diverse range of hues. For instance, the combination of carotenoids and chlorophylls in green leafy vegetables like spinach and kale contributes to their characteristic dark green colour.

1.1. **The Link Between Colour and Nutrition**

The link between colour development in vegetables and their nutritional value is a significant factor driving breeding programs. While not always a perfect correlation, breeding for colour often leads to increased levels of health-promoting compounds associated with specific pigments. Here's a closer look at this connection:

1.2.1. Coloured vegetables

Coloured vegetables are receiving enough attention as these contain different health promoting phytochemicals and phytonutrients. Carotenoids, anthocyanins, polyphenols, bioflavonoids, ellagic acids, iron, sulphur etc. are present have positive health benefits (AICR 2007, Mulabagal et al., 2010). Pigmentation prevents disease and a pest attack (Polturak et al., 2017). It increases tolerance to abiotic stresses. '**Eat a Rainbow**' is now the slogan in which the importance of daily consumption of coloured vegetables has been emphasized.

1.2.2. Rainbow vegetable

The rainbow diet is not a new idea, but it's newly popular. The idea behind it is that colourful vegetables and fruit contain specific micronutrients that support your health and combat biological stress with antioxidants and anti-inflammatory molecules. This type of biological stress affects your body at a cellular level, you probably know it as "oxidative stress", which is caused by free radicals (Schreinemachers et al., 2015). Fortunately, the antioxidants in rainbow diet foods help the body to neutralise free radicals and stop them from damaging your cells. Free radicals are generated by your metabolism (the sum of life-giving chemical reactions inside your cells) and your environment. The rainbow diet nutrients don't act directly on free radicals. Instead, they prompt your body's natural antioxidant mechanisms, which increase your natural ability to reduce oxidative stress. It also has a few other great benefits too. Plant foods are full of fibre, and fibre is what keeps your digestive system running optimally. Plus, fibre and other plant nutrients are prebiotics: food molecules for your gut bacteria that also help keep you healthy.

1.2.3. Red vegetables:

Red fruits and vegetables are coloured by a natural plant pigment called lycopene. Lycopene is a powerful antioxidant that can help reduce the risk of cancer and keep our heart healthy. It maintains healthy heart, memory function, urinary tract health and also lower the risk of cancer.

1.2.4. Orange/Yellow vegetables:

Carotenoids give this group their vibrant colour. A well-known carotenoid called Betacarotene is found in sweet potatoes, pumpkins and carrots. It is converted to vitamin A, which helps maintain healthy mucous membranes and healthy eyes. Another carotenoid called lutein is stored in the eye and has been found to prevent cataracts and age-related macular degeneration, which can lead to blindness.

1.2.5. Green vegetables:

Green vegetables contain a range of phytochemicals including carotenoids, indoles and saponins, all of which have anti-cancer properties. Leafy greens such as spinach and broccoli are also excellent sources of folate.

1.2.6. White vegetables:

White fruits and vegetables contain a range of health-promoting phytochemicals such as allicin (found in garlic) which is known for its antiviral and antibacterial properties. Some members of the white group, such as bananas and potatoes, are also a good source of potassium.

1.2.7. Purple vegetables:

The plant pigment anthocyanin is what gives blue/purple fruits and vegetables their distinctive colour. Anthocyanin also has antioxidant properties that protect cells from damage and can help reduce the risk of cancer, stroke and heart disease.

1.2. **Consumer Preferences and Market Demands**

Consumer preferences for colour play a significant role in the success of breeding programs for vegetables. Vibrant colours often attract consumers, influencing their purchasing decisions. Studies have shown that consumers are more likely to choose orange-fleshed sweet potatoes over paler varieties, even if they are unaware of the increased vitamin A content in the orange variety. This highlights the importance of understanding consumer preferences and incorporating them into breeding goals.

The market demand for specific colours can also be influenced by cultural traditions and regional preferences. In Asia, for instance, red and yellow vegetables are often associated with good luck and prosperity, leading to a higher demand for these colours in certain markets.

2. Breeding Methodologies for Colour Development

2.1. Traditional Breeding Methods

Traditional breeding methods have been the cornerstone of vegetable improvement for centuries. These methods rely on the principles of Mendelian genetics and natural variation within plant populations. Here are some key techniques employed:

• **Selection:** This fundamental approach involves identifying and selecting plants with desirable colour traits for use as parents in the next generation. There are two primary methods of selection:

o **Mass Selection:** This method involves selecting individual plants with the desired colour phenotype (physical appearance) and pooling their seeds for the next generation. This approach can be effective for simply accumulating desirable genes over time but lacks the precision of other methods. For instance, mass selection might be employed to gradually increase the overall anthocyanin content in a population of red cabbage.

o **Pedigree Selection:** This method tracks the lineage of individual plants through generations. Breeders select parents with the desired colour traits and cross them to create offspring with a predictable combination of genes. This approach is more targeted than mass selection but can be time-consuming. Pedigree selection might be used to develop a new variety of orange carrots with a specific shade of orange by carefully selecting parents with the desired colour characteristics and tracking the inheritance patterns in subsequent generations [Allard, 1960]. The two famous example of the variety bred through selection of carrot the characteristic cream colour variety Pusa Kulfi rich in lutein and Pusa Ashita rich in anthocyanin.

• **Hybridization:** Crossing between genetically dissimilar parents. It is the best method for crop improvement in cross pollinated crops (Chen et al., 2005). High-Carotene Cucumber Germplasm developed by crosses between U.S. pickling cucumber lines (*Cucumis sativus* L. var. *sativus*) and the orange-fruited Xishuangbannan cucumber (*C. sativus* L. var. *xishuangbannanesis* Qi et Yuan). Variety developed by this method are Pusa Nayanajyoti and Pusa Vasudha in carrot. However, maintaining hybrid seed production can be a challenge for some vegetable crops, as it often requires specialized techniques and facilities.

o Interspecific Hybridization: Crosses were made between two different species of the same genus known as Interspecific hybridization. Caro-Red, Provitamin- A rich tomato variety developed by cross between common tomatoes, *Lycopersicon esculentum* Mill., and the wild species, *L. hirsutum* Humb.

• **Backcrossing:** This method involves repeatedly crossing a plant with desirable colour traits (donor parent) to a recurrent parent of a different variety. This allows the breeder to introduce the desired colour gene(s) while maintaining the genetic background of the recurrent parent. Backcrossing is a valuable tool for incorporating specific colour traits into established varieties. For instance, a breeder might use backcrossing to introduce genes for intense red colour from a wild tomato species into a cultivated tomato variety known for its flavour and disease resistance (Tanksley et al., 1982).

2.2. Advanced Breeding Techniques

Modern advancements in biotechnology have opened doors to more precise and efficient breeding methods. These advanced techniques complement traditional breeding and offer new avenues for achieving desired colours in vegetables:

2.2.1. Mutagenesis- Mutagenesis referred to as "variation breeding", is the process of exposing seeds to chemicals, radiation, or enzymes in order to generate mutants with desirable traits. Plants created using mutagenesis are sometimes called mutagenic plants or mutagenic seeds.

Table 1: List of mutants in tomato and pepper

Species	Mutant name	Phenotype	References
Tomato	r (yellow flesh)	Yellow fruit color	Fray and Grierson, 1993
	tangerine	Orange fruit color	Isaacson et al., 2002
Pepper	c2	Yellow fruit color	Thorup et al., 2000

1.1.2. Marker-Assisted Selection (MAS): This method utilizes DNA markers linked to genes controlling colour development. By identifying these markers, breeders can select plants with the desired colour traits even at the seedling stage. This significantly accelerates the breeding process compared to traditional selection methods based solely on phenotype (physical appearance). MAS relies on the development of reliable DNA markers that are closely associated with the genes of interest. Once these markers are identified, breeders can screen large numbers of seedlings quickly and efficiently to identify those possessing the desired colour genes.

1.1.3. Quantitative Trait Loci (QTL) Mapping: This advanced technique identifies specific regions of the genome associated with colour traits. Colour development is often a polygenic trait, meaning it is controlled by multiple genes. QTL mapping helps breeders pinpoint these regions on the chromosomes, allowing them to gain a deeper understanding of the genetic architecture of colour development and use this information to develop more targeted breeding strategies. QTL mapping involves analyzing the inheritance patterns of colour traits across large populations of plants. By statistically associating specific regions of the genome with variations in colour, breeders can identify potential QTLs for further investigation.

1.1.4. Association Mapping: This approach identifies associations between specific genetic markers and colour variation within a population. This method is particularly useful for identifying genes with minor effects on colour traits that may be difficult to isolate using traditional breeding methods. Association mapping is similar to QTL mapping but utilizes a different statistical approach. It analyzes existing genetic variation within a population to identify markers associated with colour differences. This method can be particularly helpful for identifying genes with subtle effects on colour that might be overlooked by traditional breeding techniques.

3.Biofortification: Biofortification is an effective and economical method to improve the micronutrient content of crops, particularly staples that sustain human populations in developing countries. Whereas conventional fortification requires artificial additives, biofortification involves the synthesis or accumulation of nutrients by plants at source.

3.1. Genetic Engineering: While still in its early stages for vegetable breeding, genetic engineering holds promise for targeted manipulation of colour pathways. This technique involves introducing genes responsible for specific pigment production or modifying existing regulatory genes to achieve desired colour modifications. For example, researchers at the John Innes Centre in the United Kingdom successfully used genetic engineering to develop tomato varieties with significantly elevated anthocyanin levels by introducing an anthocyanin regulatory gene from snapdragon. However, ethical considerations and regulatory hurdles remain surrounding genetically modified organisms (GMOs), and consumer acceptance of such products needs to be carefully evaluated.

1.1.1. Biotechnological Approach: Transgenic plants are plants into which one or more genes from another species have been introduced into the genome, using genetic engineering processes. Anthocyanin in Tomato- it is increased by over expressing either the regulatory or structural genes involved in the biosynthetic pathway by transgenic approaches. Anthocyanin and beta-Carotene in Cauliflower-Purple cauliflower (*B. oleracea* var. *botrytis*), Anthocyanin synthesis is regulated by transcriptional regulation of structural genes. Anthocyanin in Sweet Potato-Mano et al., (2001) found the IbMYB1, R2R3-type MYB gene, which was new, from a purple-fleshed sweet potato.

Table 2: Genes involved in colour development in vegetables

Vegetable	Coloured pigment	Genes
Tomato	Beta carotene crimson fruit colour yellow fruit colour	*B* *(og^c),* *(r)*
Tomato	Anthocyanin	*Aft, Abg , Atv*
Brinjal	Anthocyanin (purple)	*Pfa, Pfb*
Chilli	Beta carotene Anthocyanin (Immature fruit)	*B;bc* *A/F*
Potato	Beta carotene	*Or* *Crt-B*
Sweet Potato	Anthocyanin	*ibMYB1*
Cauliflower	Beta carotene Anthocyanin	*Or* *Pr*
Cabbage	Anthocyanin	*MYB*

3.1.1 Achievements in Breeding for Colour vegetable

Significant progress has been made in breeding vegetable varieties with improved colour:

3.1.1.1. Cauliflower *Or* gene

- Semi-dominant, single-locus mutation
- Function- regulating the differentiation of some non-photosynthetic plastids into chromoplasts, which provide the deposition "sink" for carotenoid accumulation
- The *Or* gene encodes a plastid membrane protein
- Beta carotene range from 3 to 320 µg/ 100 gram fresh tissue

3.1.1.2. Cauliflower (*Pr* gene)

- Anthocyanins are responsible for purple color.
- Purple (*Pr*) gene mutation is a spontaneous mutation.
- The cauliflower purple mutation controlled by a single, semi dominant gene
- Phenotype - Intense purple color in curds and a few other tissues
- A commercial purple cauliflower cultivar Graffiti (Harris Seeds) cultivated in USA.
- The curds accumulated approximately 3.75 mg cyanidin diglucoside equivalent per g fresh weight

3.1.1.3. Black Carrot: Rich source of Anthocyanins

- Anthocyanins contains sinapic acid, coumaric acid or cinnamic acid gives rise to the black or purple carrots. (45.45mg/ kg FW)
- The natural pigment exhibits significant antioxidant activity and promote immune system.
- Recent black carrot variety develop is Pusa Asita having anthocyanin content of about 39.29mg/100g (ICAR, 2021).

3.1.1.4. Orange Tomato: High ß-Carotene

- β-carotene content of the orange-fruited high beta types ranges from 3.81-6.55 mg/100 g FW compared to 0.60-0.9 mg/100 g for normal red-fruited tomato.
- The highest levels of lycopene, vitamin C, phenolics and solids contents found in wild relative *S. pimpinellifolium* (Hanson et al., 2004).

Table 3: Gene responsible for different yellow/orange colour development in tomato

Genes	Colour development
Apricot (*at*)	Yellowish pink
Beta carotene (*Beta*)	Orange

Genes	Colour development
Delta (*Del*)	Reddish orange
Greenflesh (*gf*)	Reddish brown
Sherry (*sh*)	Yellow with reddish tinge
Yellow (*r*)	Yellow

3.1.1.5. Purple Tomato: High Anthocyanin conetnt

▪ Dominant gene Anthocyanin fruit (Aft), which induces limited pigmentation upon stimulation by high light intensity, was introgressed from S. *chilense*.

▪ Aubergine (Abg), introgressed from *Solanum lycopersicoides* Dunal, can induce a strong and variegated pigmentation in the peel of tomatoes (Jones et al., 2003 and Mes et al., 2008)

▪ Gene responsible for purple colour in tomato are *Aft*, *Abg*, *Atv*

3.1.1.6. Red Okra: Anthocyanin rich

▪ Two major anthocyanins (delphinidin and cyanidin) are responsible for colour development in okra (Zang et al., 2019)

▪ It acts as antioxidant and helps in reducing levels of sugar. Therefore, known as anti-diabetic fruit (Alsuhaibani et al., 2017)

▪ Kashi Lalima develop by IIVR, Varanasi is well known red Okra variety.

▪ The anthocyanin content in okra fruit is (0.004 mg/100g) (Yora et al., 2018)

3.1.1.7. Yellow watermelon

▪ Colour development in watermelon is govern by monogenic inhertiance

▪ Gene regulating yellow flesh colour is (*y*) which is recessive in nature

▪ Durgapur Kesar is a Yellow fleshed watermelon developed by Rajasthan Agricultural Research Institute, Durgapura

▪ Varieties developed by private companies are Yellowgold 48 (Bayer's), Yellow Crimson, Yellow Doll, Buttercup Yellow Melon

3.1.1.8. Purple or Black Hot pepper

▪ The gene regulating purple or black colour in pepper is (*A*) which has high anthocyanin content in it.

▪ For beta carotene gene responsible is (*B*)

▪ Incomplete Dominant gene Anthocyanin fruit (*A*), which induces limited pigmentation upon stimulation by high light intensity, was introgressed from *C. frutescens* and *C. chinense*

▪ Contain vitamins A, B6, and C, manganese, potassium, folate, fiber, and anthocyanins, which are antioxidants that can help boost overall health.

Table 4: Coloured rich varieties of different Vegetable

Vegetable	Varieties	Phytochemeicals	Availability
Carrot	Pusa Asita Pusa Rudhira Pusa Nayanjyoti Pusa Vrishti Pusa Yamdagni Pusa Meghali	Anthocyanin Lycopene Beta carotene Beta carotene	39.29mg/100 g 10.55mg/100g 7.55mg/100g 3.35 mg/100g (Tanmay et al., 2013) 4.60mg/100g (Tanmay et al., 2013) 4.77mg/100 g (Tanmay et al., 2013)
Red cabbage	Red Acre	Anthocyanin	
Bitter gourd	Pusa Aushadi	Beta carotene	

Table 5: Different vegetables with enriched pigments

Vegetable	Varieties	Pigment present
Watermelon (yellow)	Durgapur Kesar Yellow Doll	Beta carotene Beta carotene
Watermelon (pink)	Sweet Princess	Lycopene
Tomato	Pusa Rohini	Lycopene
Brinjal	Pusa Shyamla	Anthocyanin
Radish	Pusa Jamuni Pusa Gulabi	Lycopene Anthocyanins
Beetroot	Crossby	Anthocyanins (Sawicki et al., 2016)

Table 6: Improvements made in coloured vegetables through breeding approaches

S.No.	Characterstics	Modification	Targeted Crops
1.	Nutrition	Improved nutrition	Red okra, red spinach, Reddish purpleyard long bean, reddish Chenopodium.
2.	Yield	Increased yield	Reddish purple yard long bean, yellow seeded mungbean.
3.	Disease tolerance	Tolerance to leafspot, white blister and anthracnose disease	Red spinach, kangkongha vingred coloured stem.

S.No.	Characterstics	Modification	Targeted Crops
4.	Insect Infestations	Tolerant to shoot & fruit borer, resistance to leaf miner and tolerant to leaf curl complex.	Red okra, Reddish purple yardlong bean, purple type hot pepper.
5.	Plant architecture	Gigantisms	Reddish Chenopodium
6.	Seasonal sensitivity	Capability of growing in the off season	Red okra & red spinach
7.	Adaptability	Tolerant to high soil moisture	Purple & black hot pepper.

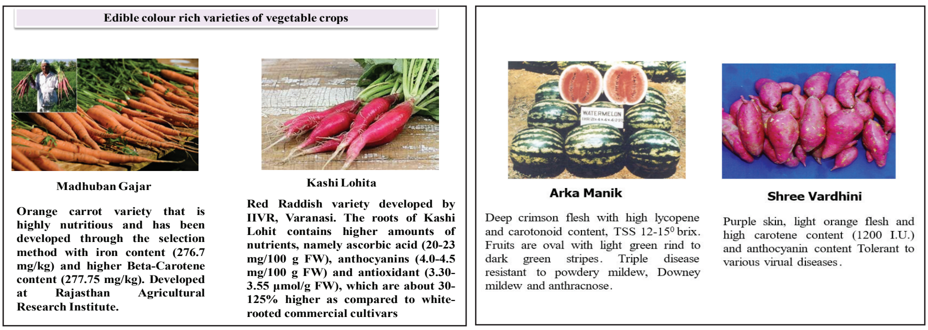

Figure 1: Coloured vegetables variety of carrot (Madhuban Gajar), red radish (Kashi Lohita), watermelon (Arka Manik), sweet potato (Shree Vardhini)

3. Nutritional Targets for Biofortification

The application of fertilizers is employed to enhance the micronutrient levels in edible parts (Prasad et al., 2015). Zinc (administered through foliar applications of ZnSO4), Iodine (applied to soil as iodide or iodate), and Selenium (in the form of selenate) are among the most suitable micronutrients for agronomic biofortification. Foliar application stands out as a rapid and convenient method for fortifying plants with micronutrients (such as Fe, Zn, Cu, etc.). Numerous studies have demonstrated that mycorrhizal associations lead to increased concentrations of Fe, Se, Zn, and Cu in crop plants. Arbuscular mycorrhizal (AM) fungi enhance the uptake and efficiency of micronutrients like Zn, Cu, and Fe. Additionally, sulfur-oxidizing bacteria augment the sulfur content in onions.

4.1 Biofortification of crops with Iron

Tomato plants exhibit a remarkable ability to withstand elevated iodine levels, accumulating this element in both vegetative tissues and fruits at concentrations surpassing those required for human dietary needs. This suggests that tomatoes represent an excellent candidate for iodine-biofortification initiatives. In plants treated with 5 mM iodide, the iodine concentration in fruits was found to exceed the daily human intake requirement of 150 μg (Martina et al., 2011). Furthermore, research indicates that the iron levels in Amaranthus plants can be increased by utilizing *S. platensis* as a microbial inoculant compared to control conditions. It has been noted that Spirulina platensis serves as an effective biofortifying agent for enhancing the iron content in Amaranthus gangeticus plants (Kalpana et al., 2014).

4.2. Biofortification of crops with Zinc

The correlation between tuber zinc (Zn) concentration and foliar Zn application demonstrates a saturation curve, peaking at approximately 30 mg Zn per kg of dry matter (DM) with a foliar Zn application rate of 1.08 g per plant. Even with a 40-fold increase in shoot Zn concentration compared to unfertilized controls, White observed after foliar Zn fertilization with 2.16 g Zn per plant, saturation was still evident (White & Martin, 2009). The application of the fertilizer "Riverm" during the cultivation of sweet peppers, eggplants, and tomatoes contributes to their enrichment with zinc. Biofortified vegetables exhibit a Zn content 6.60-8.59% higher than that of the control group (Yudicheva, 2014).

4.3. Biofortification of crops with Selenium

Selenium-enriched *S. pinnata* proves to be beneficial as a soil amendment for enhancing the selenium content in broccoli and carrots, providing them with health-promoting organic forms of selenium. Onions and carrots were subjected to biofortification through foliar application of a solution containing 99.7% enriched 77Se (IV) (Banuelos et al., 2015). In brassica vegetables, selenium application did not demonstrate any significant impact on yield or oil content (Kapolna et al., 2012). Furthermore, a substantial accumulation of selenium was detected in the seeds and meal, with levels ranging from 1.92 to 1.96 μg per gram (Seppanen et al., 2010).

5. Challenges and Limitations

Despite the significant progress made in breeding for colour development, some challenges remain. Here are some key considerations:

- **Genetic Complexity:** The inheritance of colour traits in vegetables is often complex, involving multiple genes and interactions with environmental factors. This polygenic nature can make it challenging to predict the outcome of breeding crosses and significantly extend the breeding timeline. Breeders may need to conduct multiple generations of crosses and selection to achieve the desired colour characteristics (Tanksley, 2004).

- **Linkage Drag:** In some cases, desirable colour genes may be linked to undesirable traits, such as reduced yield or susceptibility to disease. This phenomenon, known as linkage drag, can hinder breeding progress. As breeders select for the desired colour gene, they may inadvertently introduce the linked undesirable trait along with it. Techniques like marker-assisted selection (MAS) can help mitigate linkage drag by allowing breeders to select for the desired colour gene while avoiding unwanted traits.

- **Consumer Acceptance:** While vibrant colours are generally appealing, consumers may be hesitant to try new or unfamiliar colours. Breeding programs need to strike a balance between colour development and maintaining consumer acceptance. Incorporating consumer research and involving them in the breeding process can help ensure the development of varieties with colours that are not only visually appealing but also meet consumer preferences.

- **Environmental Factors:** Colour development in vegetables can be significantly influenced by environmental factors like light intensity, temperature, and nutrient availability. For example, high light intensity can promote anthocyanin production, leading to more vibrant colours in vegetables like purple cabbage. This necessitates breeding for colour under controlled conditions, such as greenhouses, or selecting varieties that adapt well to different environmental variations.

These challenges highlight the need for a comprehensive approach to breeding for colour development. Combining traditional breeding methods with advanced techniques like MAS and incorporating consumer preferences can lead to the development of superior coloured vegetable varieties that are not only visually appealing but also meet market demands and thrive under diverse environmental conditions.

6. Conclusion

Malnutrition and hidden hunger are both present in developed and developing countries and have devastating effects globally. The recent implications of the global pandemic have shown that food systems need to be adapted to advance global changes that can limit deficiencies in our food supply. Through plant breeding, transgenics, and mineral fertilizer applications, micronutrient malnutrition can potentially be tackled. It is important to add that to successfully combat hidden hunger through biofortification, even after the development of biofortified varieties, it will be essential to address various socio-political and economic challenges to promote their cultivation and finally their consumption by customers. For future actions, an integrated approach is required, where politicians, farmers, food product developers, genetic engineers, dietitians, and educators need to be included in the developing efforts. One of the biggest challenges of biofortification aside from the methods to strengthen the nutritional value of crops is the public acceptance. Especially for the transgenic techniques more education and marketing should be invested for the success of biofortified products in the market as only few cultivars are finally released for costumers. Overall, biofortification represents a promising group of techniques that can improve the global nutritional wellbeing and lead us closer to minimize hunger and malnutrition.

• Novel plants with health and sensory advantage often attracts people and so their acceptance remains high

• Coloured vegetables are associated with nutraceutical benefits making it important criteria for their increase in consumption

• Some of the coloured varieties developed includes –Durgapur Kesar (watermelon), Sweet Princess (watermelon), Pusa Rohini (tomato), Pusa Rudhira(carrot), etc.

• Biotechnological approaches and transgenic have emerged as the two most potent technologies which have potential to attain quality improvement in coming years

7. Future Prospects

Breeding for colour development in vegetables is a dynamic field with immense potential. Here's a glimpse into the future of this exciting field:

• **Integration of Breeding Techniques:** Combining traditional breeding methods like selection and hybridization with advanced technologies like MAS, QTL mapping, and even genetic engineering can accelerate the development of superior coloured varieties. Utilizing a multi-pronged approach allows breeders to leverage the strengths of each technique for more efficient and targeted breeding efforts.

• **Focus on Underutilized Vegetables:** Many underutilized vegetables hold immense potential for diversification and colour improvement. These vegetables may contain unique pigments or offer opportunities to create novel colours not readily found in mainstream varieties. Breeding programs focusing on underutilized vegetables can increase their diversity and appeal to consumers, promoting a more balanced and nutritious diet.

• **Consumer Preferences:** Tailoring breeding programs to incorporate consumer preferences for specific colours and their associated health benefits can further drive the adoption of these improved varieties. Conducting consumer research and involving them in the breeding process can lead to the development of vegetable varieties with enhanced visual appeal, nutritional value, and consumer acceptance.

• **Nutritional Enhancement:** While enhancing colour is a key focus, future breeding programs should strive for a holistic approach. Breeders can utilize advanced techniques like metabolic engineering to not only improve colour development but also increase the levels of other health-promoting compounds within the vegetable.

• **Sustainability Considerations:** Breeding programs should incorporate sustainability principles. Selecting varieties that thrive under diverse environmental conditions or require minimal inputs like water and fertilizer can contribute to a more sustainable agricultural system.

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