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ADVANCEMENTS IN VEGETABLE BREEDING TECHNIQUES FOR QUALITY ENHANCEMENT

Ajeet Kumar[1], S.P. Kanaujia[1*], Rajat Rajput[1], Raj Kumar[2] Abdul Rahman M[1], Ravi Shankar[1], Rishabh Kumar Singh[1] and Sentirenja Jamir[1]
[1]Department of Horticulture, S.A.S., Medziphema Campus Nagaland University Nagaland 797106, India
[2]Department of Vegetable and Spice Crops, Uttar Banga Krishi Vishwavidyalaya Cooch Behar 736165, India
*Corresponding Author email: spkanaujia1868@gmail.com
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ABSTRACT

Vegetable breeding has witnessed significant advancements in recent years, leading to remarkable improvements in both the quality and yield of various vegetable crops. This paper reviews the latest techniques and methodologies employed in vegetable breeding, focusing on the enhancement of desirable traits such as nutritional content, shelf life, and sensory attributes. Modern approaches, including marker-assisted selection, genomic selection, and gene editing technologies like CRISPR/Cas9, have revolutionized traditional breeding methods, allowing for more precise and efficient development of superior vegetable varieties. These innovative techniques facilitate the identification and incorporation of specific genes associated with quality traits, expediting the breeding process and reducing the reliance on lengthy trial-and-error methods. The application of these advanced breeding techniques has not only enhanced the quality of vegetables but also contributed to sustainable agricultural practices by promoting the development of resilient and high-yielding crops. This review highlights the impact of these advancements on vegetable quality and discusses the future prospects and challenges in the field of vegetable breeding. By leveraging cutting-edge technologies, the vegetable breeding industry is poised to meet the growing demand for high-quality produce, ensuring food security and improving nutritional outcomes for populations worldwide.

***Keywords*:** CRISPR/Cas9, Genetic improvement, Genomic selection, Marker-assisted selection, Nutritional content, Quality enhancement, Vegetable breeding

Introduction

The growing global population, insufficient food and nutrition, lack of vital micronutrients and vitamins, and other issues plague most developing nations. Micronutrient deficiency is a serious health condition that causes hidden hunger in people who eat enough but not enough nutritious meals. Iron, zinc, iodine, selenium, and vitamin A insufficiency are common in malnourished people. Malnutrition during pregnancy and childhood causes morbidity, death, physical, and mental abnormalities. Due to chronic energy-protein malnutrition, one-third of all children worldwide are stunted and wasted in India (FAO, 2013; International Institute for Population Sciences, 2016). The National Health and Family Survey (2015-16) by the Indian Government found that 27% of rural women and 23% of men are malnourished. According to the Comprehensive National Nutrition Survey (2016-18), 34.7% of children under 5 are stunted and 33.4% are underweight (Rohatgi *et al.*, 2021). Micronutrient deficiency in India costs 2.4% of GDP, or $15–46 billion, in health and productivity losses. Global malnutrition status shows 2 billion people are malnourished and 795 million undernourished. According to malnutrition data, 155 million children under 5 are stunted, 52 million are wasted, and 17 million are severely wasted. International food policy research institute IFPRI (2013) reports malnutrition causes 11% GDP loss in Asia and Africa. Both fruits and vegetables are considered "protective foods" since they are high in CHO, fat, proteins, vitamins, and minerals, with veggies being the richest and cheapest source of vitamins, minerals, and crude fibre Vitamins and minerals are needed in smaller amounts than CHO, fat, and proteins because they require physiological and metabolic processes. Therefore, WHO-FAO nutritionists recommend vegetables for a balanced diet. VAD is a major public health issue in India. About 25% of the 15 million blind persons worldwide are from India. In impoverished countries like India, Africa, and Southeast Asia, vitamin-A shortage is severe. Vitamin-A deficiency kills 140 million pre-schoolers and 1.2-3 million children (UN-SCN 2004). Colourful veggies are nutritious, beautiful, and medicinal. Plant pigments include chlorophylls, carotenoids, flavonoids, and betalains. Besides being beautiful, these colours are necessary for reproduction, co-evolution, and ecosystem survival (Chen, 2015). Breeding for colour development improves vegetable crop hues and nutrient content, reducing the risk of cancer, obesity, and cardiovascular disease and allowing the development of novel cultivars with increased bioactive qualities. Traditional and current marker-assisted selection and genetic transformation breeding methods exist for colour development (Plazas *et al.*, 2014).

Table 1 : Vegetable varieties rich in nutraceuticals compound

Crop	Varieties	Nutraceuticals
Brinjal	Pusa Safed Baingan, 31.21 mg/100 g fw	Phenolics
	Pusa Hara Baingan 1, 33.5 mg/100 g fw	Phenolics
	Pusa Shyamla, 48.2 mg/100g fw	Anthocyanin
Bottle gourd	Pusa Aushadhi, 6.51 mg/100 g fw	Phenolics
	Pusa Rasdhar, 4.3 mg/100 g fw	Phenolics
Onion	Pusa Madhvi, 101.2 mg/100g fw	Quercetin
	Pusa Ridhi, 107.42 mg/100g fw	Quercetin
	Pusa Soumya, 74.6 mg/100g fw	Quercetin
Carrot	Pusa Yamdagini, 7.55 mg/100g fw	Beta-carotene
	Pusa Rudhira, 386 mg/100g fw	Lycopene
	Pusa Asita, 339 mg/100gfw	Anthocyanin
	Pusa Rudhira, 45.15 mg/100g	Phenols
Beetroot	Crosby, 17.15 mg/g dm	Anthocyanin
Tomato	Pusa Rohini 4.5 mg/100g fw	Lycopene
Red cabbage	Primero 109 mg/100g fw	Anthocyanin
Cauliflower	Pusa Sharad, 23.94 µ mol/ 100 g fw	Sinigrin
	Pusa Beta Kesari 1, 8-10ppm	Beta carotene
Bathua	Pusa Green, 7.6 mg/100gdw	Iron
Palak	All Green, 16.2 mg/100g dw	Iron
Amaranth	Pusa Kirti, 38.5 mg/100g dw	Iron
Methi	Pusa Early Bunching, 17.2 mg/100g dw	Iron
Sag sarson	PusaSag -1, 16.3 mg/100g dw	Iron
Sweet Potato	Bhu Sona, 14.0 mg /100g	β-carotene
	Sree kanaka, 90.0 mg /100g	β-carotene
	Bhu Krishna, dry matter (24-25.5%), starch (19.5%), total sugar (1.9-2.2%)	Anthocyanin
Tapioca	Sree Visakham, 466 IU/100g	Carotene
Yam	Sree Neelima	Anthocyanin rich (Purple flesh)

(Singh *et al.*, 2021)

Improve vegetable crop quality features through breeding

Traditional and advanced breeding approaches combine one or more micronutrients from germplasm to boost vegetable crop nutrition. Many vegetable crops use molecular markers to identify and transmit gene(s) and QTLs for nutrients and biofortification. Breeding crop plants to improve nutrient levels is possible because they are genetically controlled. The accuracy of genetic architecture and breeding method determine its success. Gene action and additive genetic variance, dominance variance, and epistasis in a breeding population would determine the breeding method. Effective breeding involves manipulating and selecting desirable gene combinations, additive genetic variation, dominance variance, and achieving a close link between expected genetic gain and realised progress by selection. However, understanding of genetic variation, trait heritability, gene activity, trait associations, screening methods, and diagnostic tools is crucial. Good breeding requires multiple genetic sources, powerful genomic technologies, and biochemical diagnostics. Advanced breeding methods are as given below some of the

1) Germplasm Evaluation

2) Mutation Breeding

3) Polyploidy Breeding

4) Molecular Breeding

5) Transgenic Method

6. Targeted Genome Editing

7) RNA interference

1) Germplasm Evaluation

Production of phytonutrient-rich vegetable cultivars begins with germplasm characterisation. Agricultural cultivars, obsolete varieties, landraces, and wild relatives are nutritious. All genetic resources, including wild cousins, must be tested for phytonutrients such micro and macronutrients, vitamins, pigments, and antioxidants. Genetic diversity will be analysed using nutritional and phytonutrient data. It will identify high-nutrient lines for creating nutritionally better crop cultivars. Carotene-rich tomato varieties are produced due to genetic variability in beta carotene content. Small-fruited wild relative *S. pimpinellifolium* possesses the most lycopene, vitamin C, phenolics, and solids. Genetically wild *S.cheesmaniae*, *S. pimpinellifolium*, *S. habrochaites* have great potential to boost standard tomato cultivars' mineral content. Fernaindez-Ruiz *et al.* (2011). Guil Guerrero and Rebolloso-Fuentes (2009) categorised eight tomato cultivars by moisture, crude protein, carbohydrates, total lipids, dietary fibre, ash, energy, vitamin C, fatty acids, carotenoid profiles, mineral elements, nitrate, and oxalic acid. Skin hardness, pericarp thickness, TSS, K, P, Fe, Zn, Cu, Mn, titratable acidity, a-carotene, lycopene, and ascorbic acid were measured in 30 tomato varieties. Unpungent *C. annuum* genotypes have more -cryptoxanthin, ascorbic acid, total phenolics, and -tocopherol than pungent genotypes. Ascorbic acid, capsanthin, zeaxanthin, a-cryptoxanthin, a-carotene, a-tocopherol, and lutein are found in brown fruit varieties. Ascorbic acid, carotenoids, and tocopherol were highest in red-fruited 'Verdano Poblano' and 'Guajillo Ancho' (Hanson *et al.* 2004). *Cucumis melo* exhibits genetic variety for sugar, carotenoid, and ascorbic acid content. Burger *et al.* (2006) found genetic variability in Cucumis melo for sugar content, carotenoid and ascorbic acid concentrations, and ridge gourd minerals.

Table 2 : Wild relative of respective crops having quality traits

Vegetable Crop	***Wild Relative***	**Doner Character**
Tomato	*Solanum pimpinellifolium*	High carotene content
	S. peruvianum	High ascorbic acid content
	S. chmielewskii	High TSS content
Carrot	*Daucus carota* subsp. *carota*	Root size, pest resistance, increased nutrient content
Tomato	*S. pureja*	Protein
	S. vernei	Starch
	S. microduntom	Calcium
Cucumber	*C. sativus var xishuargbannaensis*	Rich in b carotene
Water melon	*C. lantus var. lantus*	High water content
Round melon	*C. colocynthin*	High bitter test
Buffalo gourd	*C. foetidissime*	Rich in protein and oil content
Spine gourd	*Momordica dioca*	Protein
Lettuce	*Lactuca serriola*	Disease resistance, heat tolerance, improved shelf life
Pepper	*Capsicum annuum var. glabriusculum*	Rich in capsaicin content

(Swarup,V. 2016.)

2) Mutation Breeding

In 1900, Hugo de Vries defined a mutation as a sudden heritable DNA sequence change that changes an organism's properties. A sudden heritable change in genetic personality. Mutagenesis is crucial in vegetable breeding. Mutagenesis involves treating biological materials with a mutagen to cause mutations. Mutation breeding involves inducing and isolating mutants for crop improvement. Genomic sequences from many plant species and a variety of molecular-genetic technologies have greatly improved our ability to detect or engineer genetic variation at specific loci (reverse genetics), allowing us to probe gene function and genetic engineering. There are various quality mutant genes in vegetables for creating Neutra-rich hybrids. Using hp-1 and lip-2 mutant genes for carotenoid production, tomato hybrids with high carotene levels have been developed. Mutans are physical or chemical agents that increase mutation frequency. Physical mutagens include ionising radiations (a, β, γ, X-rays) and non-ionizing (UV, infrared) rays. Singh *et al.*, 2021, list chemical mutagens as ethyl methyl sulphonate, dimethyl sulphonate, and bromouracil.

McCallum *et al.* (2000) devised a reverse genetic method that uses chemical mutagenesis' high point mutation density and rapid mutational screening to identify generated lesions. TILLING (Targeting Induced Local Lesions IN Genomes) uses chemical mutagenesis and a sensitive mutation detection technique. TILLING's versatility makes it useful for vegetable crop genetic modification. The quality mutant-rich genes in diverse vegetable crops will be used to create multi-coloured, nutrient-rich vegetables.

In 1970, Bradforsh identified the cauliflower “Or” mutant that produces beta-carotene-rich orange curd in Ontario, Canada. Introducing the ‘Or’ gene to Indian cauliflower will produce -carotene-rich cauliflower. Kalia *et al.* (2018) created Pusa Kesari VitA-1 and probable introgression lines in Indian cauliflower with 8-20 ppm beta-carotene. Zhang found SCAR markers linked to the “Or” gene, which enhances beta-carotene in Chinese cabbage. Zou *et al.* (2016) fine-mapped the or gene and found a BrPro1 molecular marker in the promoter region of Bra031539, which is projected to create CRTISO, a carotenoid isomerase essential for carotenoid biosynthesis. This marker may be used to identify orange head materials early A single dominant mutant gene “Pr” produces purple curd. The Pr mutant gene encodes MYB-1, a transcriptional regulator that regulates curd anthocyanin accumulation. Singh *et al.* (2021) are using tomato hp-1 and hp-2 mutant genes for carotenoid production to create carotene-rich hybrids. LaBonte and Don (2012) found a 30–100 ppm -carotene orange flesh sweet potato mutant of tuber crops. Swathy *et al.* (2021) examined how Helium-Neon (He-Ne) laser irradiation affected *Solanum melongena* L. chlorogenic acid. var. Field-ready Mattu Gulla. The study indicated that the maximum chlorogenic acid content was observed in the 20 J/cm^2 laser dose compared to other treatments.

Table 3 : Mutant cultivar with improve quality parameters

Crop	Variety	Special feature
Tomato	Pusa Lal Meeruti	Uniform ripening
	Thar Anant	High lycopene content
	S-12	High juice and acidity
	Co-3	High vit. C
	PKM	GREEN FLESH TYPE
Chilli	MDU 1	High Capsaicin Content
Cucumber	Swarna Ageti	Green colour without placenta hollowness
Bitter gourd	Mdu-1	High sex ratio (20:1) and white colour fruit
Garden pea	Stral-Art	High generative capacity
Dolichos bean	Co-10	White colour pod
Fenugreek	R 303	Bold seeded

(Kanaujia, *et al.*, 2020

3) Polyploidy Breeding

Abnormal cell division can cause polyploidy. This can happen in mitosis and meiosis. Nutraceuticals and colours can be improved in vegetable breeding using this strategy. Radish, pumpkin, muskmelon, and watermelon tetraploids produce and improve. (Zhang *et al.*, 2010) created tetraploid muskmelon with higher soluble solid, soluble sugar, and vitamin C content than diploid fruit. (Liu *et al.*, 2010) found that diploid watermelon fruit had 33.2 to 54.8 mg/kg lycopene and triploid 41.2 to 61.8 mg/kg. Tetraploids had 38.1–59.8 mg/kg lycopene. They found that triploid and tetraploid have more lycopene than diploid. (Marzougui *et al.*, 2009) polyploidized *Trigonella foenum-graecum* L. utilising 0.5% colchicine and found that the autotetraploid cultivar has more seed, pod, and branch numbers than diploids and bigger leaf area. Its leaves include potassium, sodium, calcium, and phosphorus. (Choudhary and Rajendra, 1980) found that tetraploid palak Pusa Jyoti had 50 to 120 mg/100 g more ascorbic acid than diploid. (Sreekumari *et al.*, 2004) examined cassava triploid and diploid tuber yield. Triploid cultivars yielded more tubers than diploids. Triploids have more dry matter and starch than diploids. The study found that ploidy level positively affects lycopene expression in watermelon (*Citrullus lanatus*) fruit growth and ripening (Dou *et al.*, 2017).

Table 4 : Polyploid cultivar with improve quality parameters

Crop	Variety	Special feature
Water melon	Pusa Bedana	1st seedless variety TSS 12-13%
	Arka Madhura	Seedless TSS 14%
	Shonima	Seedless, red flesh and high TSS content
	Swrana	1st yellow flash seedless High citrulline content
Cassava	Sree Athulya	High starch content (30.2%)
Palak	Pusa Jyoti	High vit. C

(Fageria *et al.*, 2020)

4) Molecular Breeding for Quality Traits

Molecular breeding involves using DNA markers linked to phenotypes to aid in selecting for specific breeding goals. A marker, often a DNA sequence near the gene of interest, can identify individuals or species. Molecular markers such as RAPD, AFLP, SSR, CAPS, and SNP are commonly utilised in breeding programmes. SNPs are popular markers due of their abundance, stability, and affordability. SNPs are single base changes in DNA sequences that occur in a considerable proportion (1%) of a large population. SNPs are single nucleotide base changes induced by transitions (C/T or G/A) or transversions (C/G, C/A, or T/A, T/G) (Singh *et al.*, 2021). Molecular markers like RAPD, ISSR, SSR, SCAR, CAPS, STSs, ESTs, SNPs, and DART are used to study gene linkage with high nutraceuticals and edible colours. MAS uses morphological, biochemical, or DNA/RNA markers to indirectly select a characteristic. To find gene-linked molecular markers, mapping populations like NILs and RILs are used. Zhang *et al.* (2008) discovered SCAR markers connected to the “or” gene, causing β-carotene accumulation in Chinese cabbage. (Ripley and Roslinsky, 2005) found a Brassica ISSR Marker for 2-propenyl glucosinolate. (Kalia *et al.*, 2018) at IARI designed and oriented biofortification of Indian cauliflower, which is deficient in carotene. The orange cauliflower, first discovered in Bradford Marsh, Ontario, Canada in 1970, results from a spontaneous mutation of a single dominant gene called ‘Or’ for orange gene (Dickson *et al.*, 1988). Tomato fruit antioxidant QTL analysis using S. pennellii introgression lines found 15 QTL, including six for ascorbic acid and nine for total phenolics (Rousseaux *et al.*, 2005). in broad bean 2 gene-specific SCAR marker increased protein content and reduced fibre content in seeds, which could help identify tannin-free broad bean varieties. A gene controlling β-carotene content was added to the *Cucumis sativus* var. cucumber background. xishuangbannanesis. Endocarp b-carotene inheritance shows a single recessive gene. Seven endocarp carotene-specific SSR producers were found on linkage group three and matched to cucumber chromosome with the candidate gene. (Sinclair *et al.*, 2004) found two vitamin C QTL in melon that caused 14% and 12% phenotypic variance. Four of nine molecular markers consistently associated to fruit sweetness. Seven SSR producers can be used in marker-assisted breeding to develop cucumber germplasm with high beta carotene content. (Staub, 2010) found higher carotenoid levels in Xishuangbanna gourd (700 µg/100 g on flesh weight basis) compared to typical cucumbers (22-48 ig/100 g). Thus, Xishuangbanna gourd is a unique germplasm for improving cucumber nutrition.

Molecular breeding of a novel orange-brown tomato fruit with increased beta-carotene and chlorophyll accumulation by (Manoharan *et al.*, 2017) found that two SNPs in CYC-B and SGR gene sequences segregated ‘orange-brown’ fruit colour in F2 generation. Across phenotypes, fleshy fruit carotenoid and chlorophyll concentration correlated strongly with carotenoid biosynthesis gene expression and SGR function. The orange-brown fruit is rich in β-carotene and chlorophyll. Our findings help breeders create new-colored tomato fruit using molecular markers. Jeong *et al.* (2015) developed capsinoids-containing Capsicum annuum pepper cultivars using marker-assisted backcrossing (MABC). ‘SNU11-001’ had high capsinoid levels (9649.92 lg/gDW). Our goal was to introduce C's defective pAMT allele. C from 'SNU11-001' chinense. annuum commercial cultivar, 'Shinhong.' (Kwabena et al., 2022) used marker-assisted backcrossing (MABC) to transfer shelf-life gene (alc) into two peak Ghanaian tomato breeding lines. The MABC-derived BC2F3 lines were tested for selections using molecular markers. Except for Alc-LA3134, the alc donor parent, which was not significantly different from one of the backcrosses (BC2F2.3-E-80-19-26), all MABC-derived lines had much longer shelf life than the checks. This proves that the alc gene controls shelf-life genetics.

5) Genetic Engineering/Transgenic Approach

Genetic engineering in vegetable breeding has improved nutrient value by integrating chosen transgenes into well-established varieties. This method offers unique chances to improve vegetable nutrition, taste, and bitterness. Genetic engineering in vegetables aims to boost carotenoids, calcium, zinc, and folic acid to alleviate nutritional deficits and improve health. Transgenic carrots with more calcium (Ca) may improve calcium uptake. By introducing calcium metabolism genes, carrot plants can collect more calcium in their edible sections, making them a healthier food source. Some places suffer from zinc (Zn) shortage, however genetically engineered lettuce can assist. By integrating zinc absorption and accumulation genes, genetically modified lettuce can accumulate more zinc in its tissues, making it a better zinc source. Folic acid deficiency, especially in pregnant women, is a global health issue. Genetically modified tomatoes with more folic acid are a solution. A vegetable supply naturally rich in folic acid can help alleviate folic acid insufficiency with biofortification. Carrots modified with the bacterial crtB gene have increased carotenoid content. Carrots contain antioxidant-rich carotenoids, which can improve health. Agrobacterium-mediated lettuce genetic transformation increased content to 400mg/gondry weight. By modifying plant genetics to obtain desired nutritional features, genetic engineering helps vegetable breeders increase crop nutrition, correct nutritional deficiencies, and improve human health. Direct or artificial manipulation of one or more genes, usually insertion of a foreign gene to achieve a desired phenotype.

Potatoes synthesise beta carotene by introducing Erwinia genes for phytoene synthase (CrtB), phytoene desaturase (CrtI), and lycopene betacyclase (CrtY). A bacterial carotenoid gene (crtI) expressing phytoene desaturase was used to create transgenic tomato lines with higher carotenoid content. (Gerjets and Sandmann, 2006) created a keto-carotenoids-containing potato. Transgenic cauliflower with Or transgenesis may differentiate proplastids or other non-colored plastids into chromoplasts for carotenoids, according to Lu *et al.* (2006). They showed that the Or gene can cause carotenoid accumulation in a major staple food crop in a better way. Post-transcriptional gene silencing changes nutrient biosynthesis pathways to optimise nutritional content. Tomatoes have been modified to boost flavour and nutrients. Tomato anthocyanin was increased using snapdragon (Antirrhinum) genes (Tohge *et al.*, 2015). RNAi-mediated DET1 repression under fruit-specific promoters has been demonstrated to increase tomato carotenoid and flavonoid levels without affecting plant development (Williams *et al.*, 2004). The public suffers from folate deficiency, (Diaz *et al.*, 2007) engineered tomatoes by fruit-specific over-expression of GTP cyclohydrolase I (catalyses the first step of pteridine synthesis) and amino deoxychorismate synthase (catalyses the first step of PABA synthesis) to produce ripe tomatoes with 25-fold more folate than Similar methods can boost folate in other plants. transgenic sweet potato plants by down regulating CHY-b via RNAi. In transgenic sweet potato storage roots and leaves, b-carotene levels increased and storage roots changed colour. RC plants also tolerated MV-mediated oxidative stress and salt stress better (Lee *et al.*, 2017).

Table 5 : Gene responsible for different quality traits on vegetable crops

Crops	**Traits**	**Gene**	**Features**
Tomato	Fruit weight	*Fw2.2*	
	Fruit shape	*fas* (fasciated), SUN	
	Sugar content	*Lin5*	
	Vitamin C	*Vtc9.1* (Higher vitamin C)	
	Shelf life	*Rin, nor, Nr, Cnr.*	
	Fruit colour/Carotenoids	*B (Beta)*	Yellow fruits
	higher lycopene content	Og^c	
	Orange fruits	*Del (Delta)*	
	higher lycopene content	*hp-2* (high pigment)	
	Anthocyanins	*Aft, atv, Abg*	Purple fruit color

Chilli	capsaicin	*Pun-1, Cap, cap3.1, cap4.2, cap7.1, cap7.2.*	
	Fruit Colour	*Y*	Yellow fruit colour (*yy cl+cl+)*
		C2	Orange fruit colour
		cl	Brown fruits (*y+y+clcl*)
		A	Purple fruit colour
		y+y+cl+cl	Red fruit colour
		yy clcl	Green fruit colour
	Capsanthin content	*C* (Single dominant gene)	
Turnip	Flesh colour	Monogenic	
	Skin colour	Two independent gene	
Beet root	Skin colour (Digenic)	*R_Y*	Red roots, hypocotyls and petioles
		RrY	Yellow roots, petioles and hypocotyl
		R-yy	White roots with red hypocotyls
Pea	Pod colour	*Gp*	Yellow colour
		Dp	Blue Green
		Pu, Pur	Purple fruit.
Brinjal	Fruit weight	*fw2.1, fw9.1, fw11.1*	
	Fruit colour	3 genes (*P, X, Puc*)	
	Fruit shape	*Ofa, Ofb1, Ofb2 and Ofb3*	
	Anthocyanin	*fap10.1*	
	Parthenocarpy	*Cop3.1, Cop8.1*	
Cauliflower	Curd colour	*Or* gene	β-carotene accumulation
		Pr (Single dominant gene)	Purple curd color
Cabbage	Head shape	*Htd 3.1, Htd 8.1*	
Carrot	Carotenoids	*PSY*	
	Root colour (Digenic)	*iiPPYYEE*	Deep purple
		iiPPYYee	Purple
		IippYYee	Yellow
		iiPPyyEE	Red
		iippyyee	Orange
	Root shape (*D, N, P*)	*D-N-P*	Long or Desi type
		dd, nn, p	Cylindrical
		dd, N-P	Chantenay type
		dd, N-, Pp	Round shape
Watermelon	Lycopene	*LCYB*	Red flesh color
	Watermelon (Monogenic)	*Wf – Y*	White flesh
		Wfwfy	Red flesh
		C	Canary yellow flesh
		B	Yellow fles
		yO	orange flesh
		y	salmon yellow
Potato	Skin colour	Digenic (*D-R-)*	Red: *D-R*, White: *D_rr, dd R, dd rr.*
	Yellow Flesh colour	*Chy2* (Single dominant gene)	

(Ram, 2006)

6. Targeted genome editing

Recently, people are willing to pay more for high-quality vegetables for their health benefits. According to consumers, vegetables with uniform fruit colour, size, flavour, longer shelf life, and many nutritional elements are high-quality. Organic gardening produces quality crops but is less popular owing to low yield and long process. Using biotechnology like CRISPR/Cas9 can improve vegetable quality. After targeting MYB12, ANT1, and PSY1, CRISPR/Cas9 effectively grow pink, purple, and yellow tomatoes. Rodriguez-Leal *et al.*, (2017) used CRISPR/Cas9 to mutate SlCLV3 promoter to grow huge tomato fruits on WL plants. Using CRISPR/Cas9 to replace 317T of the

ALC gene with 317A in tomato plants produced high output, shelf life, and uniform fruit size (Yu *et al.*, 2018). In sweet potato and rapeseed, CRISPR/Cas9 was used to increase seed oil and total starch by targeting IbGBSSI or IbSBEII, BnSFAR4 and BnSFAR5. Eliminating three polyphenol oxidase (PPO) genes-SmelPPO4, SmelPPO5, and SmelPPO6 with CRISPRCas9-based mutagenesis stops brinjal fruit browning (Maioli *et al.*, 2020). Because Cswip1 mutations impede cucumber carpel formation, Hu *et al.*, 2017) used CRISPR-Cas9 to produce gynoecy (plants bear only pistillate flowers) in cucumber. Using SLIAA9 mutations, (Ueta *et al.*, 2017) produced parthenocarpic (seedless) tomato fruit using CRISPR/Cas9. Under high temperatures, CRISPR/Cas9-induced tomato SlAGL6 mutants produced seedless fruit. Since induced mutations do not affect fruit shape, weight, or pollen viability, SlAGL6 mutant can be employed to grow (cucumber and watermelon. Colour is determined by the buildup of carotenoids in the pericarp and flavonoids in the peel, as well as chlorophyll breakdown during ripening. Fruit colour is a multigenic trait, hence traditional crossbreeding takes years to introgress all colour-related genes in a single genetic background. Avoiding linkage drag is difficult. We presented a quick breeding technique to develop tomato lines with different-colored fruits from red-fruited materials using CRISPR/Cas9-mediated multiplex gene editing of three fruit-color-related genes (PSY1, MYB12, and SGR1). This method has been used to engineer the red-fruited cultivar 'Ailsa Craig' into tomato genotypes with yellow, brown, pink, light-yellow, pink-brown, yellow-green, and light green fruits (Yang *et al.*, 2023).

7) RNA interference in vegetable quality attributes

Double-stranded RNA (dsRNA) drives RNA interference. DsRNA is normally not found in differentiated somatic cells, therefore its presence signifies a problem and activates a cellular defence mechanism. In the first step, Dicer, a conserved cellular RNAse, breaks down dsRNA into tiny oligoribonucleotides with 22 base pairs and 3′ overhangs. Known as siRNA, this tiny dsRNA is interfering. Second, siRNA binds to RISC, a multimolecular protein complex. Unwinding the double-stranded siRNA molecule creates a ribonucleoprotein particle with RISC proteins and one siRNA strand. When this complex encounters an mRNA with a sequence corresponding to the siRNA moiety, Slicer cleaves it and renders it inactive. If complementarity is imperfect, RISC may only attach to mRNA, blocking translation and expression. Crop improvement has expanded with the discovery of RNA interference (RNAi) and its regulatory potential (Jagtap *et al.*, 2011). Antisense technology is less precise, efficient, reliable, and publically accepted than RNAi technology.

The novel gene regulatory mechanism RNA silencing limits transcript levels by suppressing transcription (TGS) or activating a sequence-specific RNA degradation process (PTGS/RNA interference) (Agrawal *et al.*, 2003). This technology has been used to increase antioxidants in tomatoes (Niggeweg *et al.*, 2004) or suppress overexpression of negative traits like sinapate. RNAi technology, which inhibits the expression of ACC oxidase gene and suppresses α-mannosidase and β-D-N-acytlhexosaminidase (β-Hex), has been shown to extend tomato shelf life (Xiong *et al.*, 2005; Meli *et al.*, 2010). Carrot allergy sufferers found Dau c 1.01 and Dau c 1.02-silenced transgenic carrot plants less allergic using RNAi technology by (Peters *et al.*, 2011). McCormick *et al.* (2004) created CarVY-resistant carrot genotypes using RNAi. Further, (Moreno *et al.*, 2013) found that DcLcyb1 transgenic silenced lines reduced carrot storage root thickness and colour. India made few attempts to use this revolutionary technique to increase vegetable crops, particularly carrot, tomato, watermelon, and sweet pepper. However, RNAi technology has significant potential in improving vegetable crops for specific qualities including beta-carotene in tropical carrot, late bolting in palak, radish, and cauliflower, illnesses, insect pests, and male sterility for hybrid seed production. (Xiong *et al.*, 2005) found that the short linker was more effective in RNAi of transgenic tomato plants, with 13.0%, 18.0%, and 69.0% effects on ACC oxidase gene silencing. We developed transgenic tomato plants with fruit that emitted ethylene and had a shelf life of over 120 days by quickly shutting down the ACC oxidase gene. RNA and protein analysis showed non-RNA, semi-RNA, and full-RNA interference of ACC oxidase in transgenic tomato plants.

Future prospects

The historical successes of plant breeding show its future potential. Therefore, agricultural genetic modifications may be lucrative in the future. As the variety in cultivated species is exhausted, related species genes will be prioritised. Genetic engineering is growing, and we grow genetically altered crops. Genes from many creatures may improve crop performance, notably biotic and abiotic stress resistance and quality. Crop plants may also be grown to recover valuable chemicals like medicines from genetically engineered genes.

Conclusion

The nutritional value and health advantages of vegetable crops are be coming more important in consumer diets. Vegetables contain health benefits, however some crops have poor levels of public health nutrients (β-carotene, ascorbic acid, iron, calcium, and iodine). Improving these nutrients can increase public intake. Because bio fortification is sustainable and scalable, it could help combat micronutrient deficiencies. In distant rural locations where traditional foods dominate local diets and food shortages occur in the off-season, vegetable cultivars with improved nutritional content and storage-cum-transport life can be more useful. Breeders are prioritising vegetable breeding initiatives to increase nutrient content and shelf life. To improve such traits are discussed below

1. Identify suitable donors in crop germplasm to develop genetic stocks for specific nutrients and nutraceuticals,
2. Identify genetics of target compounds in donors and develop appropriate breeding strategy,
3. Identify robust and tightly linked molecular markers for target traits and background selection to track introgression level and efficiency, and
4. Deploy advanced biochemical tools and techniques.

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