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**Shubham Singh**
Ph.D., Research Scholar, Division of Vegetable Science, ICAR-Indian Agricultural Research Institute, Pusa, New Delhi, India

**Sandeep Yadav**
Ph.D., Research Scholar, Department of Vegetable Science, GB Pant University of Agriculture and Technology, Pant Nagar, Uttarakhand, India

**Abhilash Singh**
Ph.D., Research Scholar, Department of Vegetable Science, GB Pant University of Agriculture and Technology, Pant Nagar, Uttarakhand, India

**Corresponding Author:**
**Shubham Singh**
Ph.D., Research Scholar, Division of Vegetable Science, ICAR-Indian Agricultural Research Institute, Pusa, New Delhi, India

# Recent approaches for breeding vegetable crops for quality, rich in nutrients and nutraceuticals

**Shubham Singh, Sandeep Yadav and Abhilash Singh**

## Abstract
Flavonoids, a group of secondary metabolites belonging to the class of phenyl-propanoids, have the widest colours range from pale-yellow to blue. Anthocyanin, naturally occurring pigment with high antioxidant responsible for red, blue and purple color in vegetables like eggplant, onion, red cabbage, purple cabbage (Zhang *et al*., 2013) [5, 20, 28, 29]. In the recent year certain improved anthocyanin concentration in several vegetable crops have been achieved like purple carrot (200-350mg), purple potato (17-20mg), red flesh potato (20-38mg), red onion (25-40mg), red cabbage (200-3320mg), purple tomato (20-60mg) respectively. In research purpose various mutants and transgenic gene responsible for color development and improved nutritional quality have been identified like *or* (cauliflower) and *MYB* (red cabbage and purple cauliflower), *Aft, Abg, atv* (Purple tomato) respectively. Tomato (*Solanum lycopersicum* L.) is an important solanaceous vegetable crop grown throughout the world for its versatile uses. It is one of the important 'Protective Foods' as it possesses appreciable quantities of vitamins and minerals and sometimes rightly referred to as "Poor Man's Orange". Traditional genetic studies had identified several genes controlling fruit shape in tomato such as *pr* (pyriform), *o* (ovate), *bk* (beaked tomato), *n* (nipple-tip tomato), *f* (fasciated) and *lc* (for locule number), *fs8.1* exerts its effect by changing the length of carpels during pre-anthesis resulting in longer and larger mature fruit. Similarly, another major fruit-shape QTL term *ovate*, controlling the transition from round to pear-shaped fruit, was mapped, cloned and characterized at the molecular level. CRISPR/Cas-9 was used to select specific sgRNAs targeting *SGR1, LCY-E, Blc, LCY-B1 and LCY-B2* used for significantly improving lycopene content in tomato fruit with advantages such as high efficiency, rare off-targeted mutation, and stable heredity. Genome editing technologies, transgenic, RNA interference, Transcriptomics and CRISPR/Cas-9 have great potential in vegetables for enriching health beneficial constituents.

**Keywords:** Vegetable crops, quality improvement, anthocyanin, carotenoid, transgenic approaches, molecular marker

## Introduction
The burgeoning world population, insufficient food and nutrition, malnutrition of essential micronutrients and vitamins etc. are the insidious challenges to most of the developing nations across the world. Micronutrient malnutrition is an alarming health issue, leads to hidden hunger, where it strikes peoples who, may appear to be consuming an adequate quantity of food with inadequate nutritional quality. Among the malnourished population, micronutrients like iron, zinc, iodine, selenium and vitamin A deficiency are predominant. Malnutrition during pregnancy and in the growing age of a child leads to a range of severe implications including increased morbidity, mortality, physical, and mental defects. Childhood stunting and wasting rate is highest in India due to chronicity of energy-protein malnutrition, occurring in approximately one-third of all children of the world (FAO, 2013 [9]; International Institute for Population Sciences, 2016). According to the latest data available on National Health and Family Survey (2015-16) by the Indian Government, in rural India about 27% women and 23% men are malnourished (Verma and Kumar, 2019) [15, 25, 26]. Comprehensive National Nutrition Survey (2016-18) data showed 34.7% of children under 5 year of age are still low height for age (stunted growth) and 33.4% are low weight for age (underweight) (Kumar and Kumar, 2020) [15, 25, 26]. The subsequent health and productivity costs of hidden hunger in the adult population also result in severe economic losses; the economic cost of micronutrient deficiency in India is ~2.4% of GDP which is equivalent to $15–46 billion. Status date for mal-nutritional at global level indicate total 2 billion people are malnourished worldwide while about 795 million people are undernourished worldwide. The viewing the mal-nutritional date among children indicate that the Children (<5 Year) 155 million stunted, while 52 million wasted, and 17 million severely wasted respectively. Mal-nutritional contribute to loss in 11% GDP in Asia and Africa reported by International food policy research institute (IFPRI, 2013).

Both fruits & vegetable are combinely called as "protective foods" as they are rich in CHO, fat, proteins, vitamins and minerals, particularly vegetables richest and cheapest source of vitamins, minerals and crude fibre. CHO, fat and proteins are required in larger quantity as they supply energy to our body but vitamins and minerals are required in smaller quantity as they do not supply energy to our body but they required physiological process and metabolic activities hence, Nutritionists of WHO-FAO suggested that vegetables are essential for balanced diet. Vitamin-A deficiency (VAD) is recognized as a serious public health problem in India. It is estimated that 25% of the 15 million blind people globally are from India. Vitamin-A deficiency is major acute problems in developing countries, like India, African countries and south east-Asia. According (UN-SCN 2004) reported that generally 140 million pre-schooled aged children and 1.2-3 million children dies due to Vitamin-A deficiency. Colourful vegetables have enormous amount of nutritional, aesthetic and medicinal value.

Pigments in plants have four major classes: chlorophylls, carotenoids, flavonoids, and betalains. These colours are not just visually decorative and attractive, but biologically essential in reproduction, co-evolution and ecosystem sustenance (Chen, 2015) [6, 19, 22]. Breeding for colour development enhances not only colours but also enriches the nutrient content of the vegetable crops, which helps to minimise the risk of cancer, obesity, cardiovascular diseases and allows the development of a new generation of cultivars with improved bioactive properties. There are several breeding strategies for colour development including conventional methods, as well as modern strategies that rely on marker assisted selection or genetic transformation (Plazas *et al*., 2014) [23].

**Pigments in vegetable crops**

Pigments make nature colourful and likable. Plant pigments usually refer to four major well-known classes: chlorophylls, carotenoids, flavonoids, and betalains. Each class may contain various numbers of chemical compounds that can be structurally categorized into distinct subgroups. Most pigments are coloured. These coloured pigments not only visually attract animals for flower pollination and seed dispersal but also function in critical biological processes for plants and play essential co-evolutionary roles in ecosystems. The biological, ecological, and evolutionary importance of plant pigments and the derived colours cannot be overstated. On the other hand, many pigment-rich fruits and vegetables are critical in the human and animal diet. Some pigments are essential nutrients, and others may serve as nutraceuticals with additional medical benefits, including the prevention and treatment of certain diseases (Chen, 2015) [6, 19, 22]. Breeding for colour improvement enhance not only colours also enrichment with nutrition which helps minimise the risk of cancer, obesity, cardiovascular disease diabetics etc.

**Table 1:** Four major groups of plant pigment

| Pigment | Main sub groups | Typical colors | Examples |
|---|---|---|---|
| Carotenoids | Carotenes, lycopene and Xanthophylls | Orange, red, yellow | Carrot, Tomato, Water melon, Pepper, Leafy Vegetables |
| Flavonoids | Anthocyanins; flavonols | Purple, blue, red | Eggplant, red Cabbage, Onion |
| Betalains | β-cyanins and β-xanthins | Red, orange, yellow | Beet, Swiss Chard |
| Chlorophylls | a and b | Green | Any green plants |

**Anthocyanin**

Anthocyanin are naturally occurring plant pigment belonging to flavonoids family, responsible for red, purple, blue and orange pigmentation in vegetable crops. The phytomedicinal value of anthocyanin are well recognised, reported that both anthocyanin and cyanidin aglycone extract from cherries inhibit the growth of colon cancer in human. Anthocyanin has many health properties improving eyesight including night vision, anti-microbial properties, cardiovascular disease, anti-tumour, antimutagenic and also has demonstrated quite effective for inhibition the process of aflatoxin biosynthesis. Anthocyanin are belonging to aglycone cyanidin and having potent for inhibit cycloxygenase enzymes, hence it can be used as a marker for initial stage for Carcinogenesis. (Kang, 2003) reported that both anthocyanin and cyanidin. The extensive research has been made to increase the level of anthocyanin concentration in different vegetable crops illustrated table: 1.

**Table 2:** Improved anthocyanin concentration in vegetable crops

| Vegetables | Genes | Concentration enhanced level of anthocyanin |
|---|---|---|
| Purple carrot | - | 200-350 mg |
| Purple potato | - | 17-20 mg |
| Red fleshed potato | - | 20-38mg |
| Red onion | - | 25-40 mg |
| Red cabbage | *MYB* gene | 200-320 mg |
| Red radish | - | 100-154 mg |
| Purple fleshed sweet potato | *MYB* gene | - |
| Purple tomato | *Aft, Abg, atv* | 20-60 mg |
| Purple cauliflower | *MYB, Pr* (single dominant gene) | - |

**Carotenoids**

Carotenoids are most abundant in several fruits and vegetable responsible for orange, yellowish pigmentation, and reported more than 700 different kinds of carotenoid are existing. Among all types β-carotene are most widely recognised for health, phytomedicinal as well as preventive against certain degenerative disease like cervical, esophageal, pancreatic, lung, prostate, colorectal and stomach disorder. Therefore β-carotene are found in leaf, fruit and root vegetables and play important role against eye vision (Night blindness), macular degeneration and cataract. Carotenoids are lipid soluble tetraterpenoids and consisting more than 700 different kind pigment include α-carotene, β-carotene, and lycopene are carotenes; lutein, zeaxanthin, and violaxanthin are xanthophylls contribute major part of food colorant (Tanaka, 2008). Among these pigments some like α-carotene, β-cryptoxanthin, and zeaxanthin are mainly responsible for yellow while β-carotene, and lutein for orange colour and lycopene are widely distributed wide range of fruit and vegetables responsible for production of Red colour respectively. Recently extensive research have been carried out to enhanced level of β-carotene in certain vegetables, which lead to developed enormous amount of β-carotene diversity in several vegetable crops which are illustrated table no.2:

**Table 3:** Enhanced level of β-carotene in vegetable crops

| Vegetables | Enhanced level of β-carotene (μg/100 gm) |
|---|---|
| Orange carrot | 3000-6000 |
| Orange cauliflower | 80-320 |
| Pusa Beta-kesari (First biofortified cauliflower cultivar), IARI | 8-10 ppm (Orange curd colour), *Or* gene, |
| Orange cucumber | 22-48, *Ore* gene |
| Yellow potato | 200-500 |
| Orange tomato | 3000-45000 |
| Kale and collard | 30000-100000 |

## Chlorophyll

Chlorophyll are widely distributed pants pigmentation called "Real life force" that responsible for green colorant. The chlorophyll are consisting large amount of different kind pigment but, α-chlrophyll and β-chlorophyll are most widely distributed in green leafy vegetables like Palak, spinach, Amaranthus, Basella, fenugreek etc. These plant pigments have certain phytomedicinal properties like anti-mutagenic, acts as chemo-preventive compound in human are well recognized.

## Betalins

Betalin are a class of water-soluble indole-derived glycoside pigments that are found only in the order Caryophyllales (e.g., beets, cacti, and amaranths) and never co-exist in plants with anthocyanins. In other words, betalins substitute for anthocyanins that are completely absent in the Caryophyllales plants. Betalins differ from anthocyanins in the chemical structures and some properties, but share similarities to anthocyanins in the colour spectra, biological functions, and other properties. Betalins can be structurally divided into betacyanins and betaxanthins that colour flowers, fruits, and sometimes vegetative organs primarily into yellow, red or violet. Betanin, with the molecular formula $C_{24}H_{27}N_2O_{13}$, is an important food colorant produced by beetroot. Compared to those many with carotenoids and flavonoids, betalains-containing fruits and vegetables are rather limited and less well known, but include beet (*Beta vulgaris*), Swiss chard (*Beta vulgaris* spp. *cicla*).

**Table 4:** Colour rich vegetables and pigments

| Colour | Pigments/bioactive compound | Vegetables |
|---|---|---|
| Red | Lycopene | Tomatoes, Carrot and Watermelon |
| Red/crimson | Betacyanin | Beet root |
| Orange | Beta-carotene | Carrot, Cantaloupe, Pumpkin, Sweet potato, Cassava |
| Yellow | Lutein | Baby corn, Yellow corn, carrot, watermelon |
| Green | Chlorophyll | Broccoli, Kale, Spinach, Cabbage and Asparagus |
| Black/purple | Anthocyanins | Carrot, Brinjal, Broccoli, Cauliflower, Lettuce, Okra |

## Significance of colours in vegetable

### 1. Photosynthesis

In the presence of CO2 and water green plants absorb the sunlight for their food production and release $O_2$ and produced CHO are stored in starch as a reserved
food material for future use.

### 2. Pollination

Due to attractive colours in flowers pollinating agents attracts towards them and their bye encourages the pollination.

### 3. Nutritional value

Antioxidants which are enhanced by nutrition which neutralise the free radicals and other organic acids which are produced during heavy food metabolism.

### 4. Consumer preference

Colour is the most important visual component to consumer preference as it fetches higher price and thereby increase in demand for export value.

Schematic colour spectrum of carotenoids, anthocyanins, betalains and chlorophylls, with examples of pigment-rich fruits and vegetables.

Each pigment class was marked in the zones with the approximately corresponding wavelengths (400-700 nm) of visible light and the examples given in the columns below with the symbolic colours. In colour spectrum from 400-490 nm violet, indigo, blue colours are produced with anthocyanin and betalains pigments are developed.

- From 490-575 nm green colour with chlorophyll pigment is produced
- From 575-700 nm yellow, orange and red colours are produce with again anthocyanin, carotenoids and betalin pigments are developed.

## Carotenoids

Carotenoids are a large family of lipid-soluble tetraterpenoids with a basic 40-carbon polyene hydrocarbon chain structure. This family of over 600 members can be generally divided into two subgroups, carotenes ($C_{40}H_{56}$) and xanthophylls ($C_{40}H_{56}O_2$ or $C_{40}H_{56}O$, the oxygenated derivatives of carotenes), which differ in the terminal rings and oxygenation. In plants, certain carotenoids function as complementary light-harvesting pigments to precisely absorb wavelengths of light not gathered by chlorophylls, the primary photosynthesis pigment. They also provide photo-protection against excess light damage to the photosynthetic reaction centre by quenching excited species such as singlet oxygen and free radicals or by other carotenoid enabled mechanisms.

## Role of carotenoids in human nutrition

Pro-vitamin-A carotenoids in developing countries

A carotenoid availability is of particular importance in developing countries where vitamin-A deficiency (VAD) is a significant public health concern. The main underlying cause of VAD in low-income countries is a poor diet that consistently insufficient in vitamin-A, eventually leading to depleted stores that fail to achieve physiological needs. Persistent, severe deficiency can lead to xerophthalmia, a form of preventable, but irreversible, blindness in young children, and facilitates infectious diseases such as measles diarrhoea, and intestinal parasites, which increase infant mortality risks. In such low-income populations, due to the poor availability of animal sources of preformed vitamin A, dietary carotenoids from plant sources, which need to be converted to vitamin A in the intestine, contribute to ~ 80 % of daily vitamin A intake and become highly necessary.

## Prevention of oxidative stress and inflammation

However, the idea that carotenoids are important classical antioxidants, which play a role in major diseases where oxidative stress and inflammation are causative factors, is less well established. Small amounts of carotenoids can be found in almost all the lipid membranes of the body. In the animal kingdom, they are often associated with specific proteins. The xanthophylls, such as lutein and zeaxanthin, having hydrophilic hydroxyl groups, orient themselves across membranes. The carotenes, such as β-carotene and lycopene, are oriented within the bilipid layers and can disturb the phospholipid structure to a small extent, allowing for greater penetration of small molecules. Their location and orientation may play a role in their ability to act as classical antioxidants.

Sunlight is an environmental hazard over the life of human skin. Not only UV-A and UV-B but also visible and infrared light are responsible for singlet oxygen and radical production, especially in the presence of natural photosensitizers such a porphyrins and riboflavin. This can result in photoaging (roughened or patchy skin, wrinkles), UV-induced erythema (sunburn), and skin cancer.

**Table 5:** Genetic inheritance of colour in vegetable crops

| Crop | Gene | Pigments |
|---|---|---|
| Tomato | *B* | β-Carotene |
| Tomato | *Aft* | Anthocyanin |
| Tomato | *(Psy-1)* | Carotenoids |
| Carrot | *A* | α-Carotene Synthesis |
| Carrot | *K* | Lycopene Synthesis |
| Carrot | *O* | Orange Xylem |
| Carrot | *Y* | Yellow Xylem |
| Carrot | *P-1, P-2* | Purple Root |
| Chilli | *A* | Anthocyanin |
| Chilli | *B* | Beta Carotene |
| Chilli | *t* | High Beta Carotene |

## Important quality traits in some selected vegetable crops

In vegetable crops, the quality attributes are grouped as (i) intrinsic quality attributes which are inherent to the product itself and provide stimuli to consumers such as sensory attributes (flavour, taste, appearance, colour, texture and smell) and health attributes which are concerned with nutritional and health-promoting values, and (ii) extrinsic quality attributes are linked to the production method but not a property of the food itself like pesticides, eco-and animal friendliness, packaging materials, processing technology which can influence the purchasing policy of some consumers. The extrinsic quality traits are not directly related to the product performance or core benefit of the product but contribute in general benefit of product and add value. Further, extrinsic quality can be grouped in narrower sense (characteristics that are perceptible through the product itself *i.e.*, packaging, colour, brand name, price, country of origin) and broader sense (characteristics which are conveyed through marketing instruments such as distribution, communication policies, advertising and pricing). The intrinsic quality traits are generally complex in nature; hence, modern high-throughput biochemical and molecular analytical tools and techniques have great potential to handle complex traits with shorten breeding cycles.

## Tomato

### Appearance

Fruit shape (oblong or square round), size or weight (80-90 g), smoothness (without ribs), stylar or blossom end rot smooth without any depression or scar, fruit colour, uniform, red and deep red, fruit firmness, a desirable trait for good transportability and long shelf life, pericarp tissues has more sugars than the locular tissue, acid content in locular tissue has predominant influence on fruit flavour and if we want to breed for processing quality attributes some important objective such as High TSS (5.5 °Brix or higher) because high TSS gives mores cases of finished products per tons of raw fruit and thus require less energy in concentrates), fruit acidity or pH (below 4.35; because longer time required if pH increases) and low titrable acidity (as percentage of citric acid) and 0.4-0.5 % and Solid/acid ratio of 15, sugar /acid ratio of 8.5 must be maintained for obtaining high quality finished product.

## Potato

High TSS (5.5 °Brix or higher) because high TSS gives mores cases of finished products per tons of raw fruit and thus require less energy in concentrates), Acidity: pH (below 4.35; because longer time required if pH increases) and low titrable acidity (as percentage of citric acid) and 0.4-0.5%.

## Cole crops

High TSS (5.5 °Brix or higher) because high TSS gives mores cases of finished products per tonnes of raw fruit and thus require less energy in concentrates), Acidity: pH (below 4.35; because longer time required if pH increases) and low titrable acidity (as percentage of citric acid) and 0.4-0.5% and Solid/acid ratio of 15, sugar /acid ratio of 8.5 are desired for cabbage. Cauliflower; Curd colour, curd compactness, curd depth, curd shape, self-blanching is major breeding objective while, in Knol-khol: Round, medium-sized knobs of desired colour-green or purple, low fibres and creamy white to greenish white flesh in tubers are important quality attributes.

## Cucurbits

Cucumber; fruit shape and size, free from bitterness, white spine colour and development of orange flesh colour, rich in Beta-carotene rich rare major objective while in watermelon for developing orange fleshed colour, Fruit quality- shape, size, epicarp/skin colour and surface (smooth, sutures, netted), flesh thick and attractive colour, seed cavity, preferably small and sweet taste, musky flavour, juiciness and high TSS (Not less than 10%; 11-13%) are important quality attributes considered during breeding programme. Watermelon; Intermediate fruit shape advantageous because long fruits are prone to gourd neck fruits and round fruits to ‘hollow-heart’, flesh: firm, colour – attractive red, pink or yellow, seeds: smaller and fewer and TSS (more than 10%), Transportability and shelf-life are desired traits.

**Bulb crops**
**Onion for dehydration**
Snow white colour, globe shaped bulbs, thick neck, free from greening and moulds, high pungency and high TSS (>18%) and low reducing sugar that reduces caking and deterioration of the colour during storage. Garlic: Bulb size index, bulb colour, clove colour, clove shape (sickle), number of cloves, clove diameter, TSS, dry matter are important quality traits.

**Root crops**
Generally non-branching and non-forking, smooth surface without fibrous in roots, thick flesh, thin self-coloured core, high carotene and high sugar and dry matter in roots, good flavour and taste are important quality traits in Carrot. Beetroot: uniform size, shape and colour, uniformly coloured roots without roots without internal zoning or white rings. Turnip: root shape, size, colour of skin and flesh and their uniformity. Radish: Root length, shape and colour, pungency, taste and edible quality, late pithiness, non-forking roots.

**Table 6:** Genetic for important quality traits in selected 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) | |

| | | | |
|---|---|---|---|
| Radish | Root colour (3 complementary gene) | *R2 r2/R3 r3/cc* | Red root colour |
| | | *r 2 r2 /r3 r3 /CC* | White root colour |
| | | *r 2 r2 / R3 r3 /cc, R2 r2 /r3 r3 /Cc* | Purple root colour |
| Beet root | Root colour (Two loci *R-Y)* | *R* (Red): *R, Rt, r, Rp,* and *Rh. Y* (Yellow): *Y, Yr,* and *y* | |
| Cucumber | Fruit colour (Digenic *R-C*) | *R-C* | Red |
| | | *R-cc* | Orange |
| | | *rrC* | Yellow |
| | Flesh colour (*V-W*) | *V-W* | Diggy White |
| | | *V-ww* | Intense White |
| Muskmelon | Flesh colour (Digenic) | *gf* | Green flesh colour |
| | | *wf* | White flesh |
| | Carotenoids | *CmOr* | Orange flesh colour |
| Onion | Bulb colour (Five gene I,*C,G,L,R*) | *II* | Inhibitory gene – white colour |
| | | *iicc* | White bulb colour |
| | | *iiC-R* | Red colour |
| | | *iiC-rr* | Yellow colour |

**Breeding of vegetable crops for nutrients and nutraceuticals**

The chief immediate and long-term objective of plant breeding is to enhance productivity to meet the ever-increasing food requirement of people. New varieties with improved desirable traits are major contributor in increasing production. Increasing the awareness among the People for their health conscious and use to take colored vegetables rich in vitamins, minerals, antioxidants etc., current breeding program have been shifted to developed Nutra-rich and multi-colored vegetable varieties suitable to meet their requirement. Most of the quality characters are complex and determined by both genetical and environmental factors. Conventional breeding in conjunction with molecular biology will make it possible to get vegetable varieties enriched with nutraceuticals and edible colors suitable for fresh market as well as industry. There are some following Pre-requisite objective must be considered before initiating any breeding programmed such as Identify suitable donors in crop germplasm to develop genetic stocks for specific nutrients and nutraceuticals, Identify genetics of the target compounds in donors and develop appropriate breeding strategy, Identify robust and tightly linked molecular markers for target traits as well as for background selection and ultimately to developed advance tools and techniques of genomic and biochemical tools for rapid detection of compounds in advance stage breeding materials. Conventional breeding plays a significant role for developing various Nutra-rich and multi-colored vegetables cultivars which as follow Table no 7.

**Advanced breeding approaches for quality and colour improvement in vegetable crops**

Breeding method exploited for crop improvement are depend upon genetic architecture or pattern of inheritance of trait, and nature of inheritance. The choice of breeding programme depends on gene action (Additive gene action, dominance, and epistasis) in breeding population, donor source and genetic underlying for trait expression, hence efficient breeding procedure should be effectively utilized for crop improvement. Breeding method utilized in self- and cross-pollinated vegetables differ among themselves.

**Pre-breeding**

Using crop wild relatives (CWR) in crop improvement is much more difficult than breeding with domesticated varieties. Pre-breeding aims to isolate desired genetic traits (e.g. disease resistance) from unadopted material like CWR and introduce them into breeding lines that are more readily crossable with modern, elite varieties. Broccoli Two QTLs such as QTL1 and QTL2 have been identified in *Brassica villosa* (Rich source Gluraphone and introgressed in cultivated types for developing glucoraphanin rich content in Brassica vegetable crops. Similarly, in tomato one wild germplasm such as *S. pimpinellifolium* has been reported rich source to tomato- β carotene content (3.81- 6.55mg/ 100 g FW) and some other novel trait such as purple pigmentation has been reported in *S. chilense – Aft* gene (Anthocyanin fruit) and *S. lycopersicoides- Abg* gene are responsible for purple fruit color in tomato.

**Table 7:** Natural colour rich vegetable varieties through conventional breeding

| Crop | Varieties | Nutraceuticals |
|---|---|---|
| Brinjal | Pusa Safed Baingan, 31.21 mg/100 g fw<br>Pusa Hara Baingan 1, 33.5 mg/100 g fw<br>Pusa Shyamla, 48.2 mg/100g fw | Phenolics<br>Phenolics<br>Anthocyanin |
| Bitter gourd | Pusa Aushadhi, 6.51 mg/100 g fw<br>Pusa Rasdhar, 4.3 mg/100 g fw | Phenolics |
| Onion | Pusa Madhvi, 101.2 mg/100g fw<br>Pusa Ridhi, 107.42 mg/100g fw<br>Pusa Soumya, 74.6 mg/100g fw | Quercetin |
| Carrot | Pusa Yamdagini, 7.55 mg/100g fw<br>Pusa Rudhira, 386 mg/100g fw<br>Pusa Asita, 339 mg/100g fw<br>Pusa Rudhira, 45.15 mg/100g | Beta-carotene<br>Lycopene<br>Anthocyanin<br>Phenols |
| Beetroot | Crosby, 17.15 mg/g dm | Anthocyanin (Sawicki *et al.*, 2016) |
| Tomato | Pusa Rohini 4.5 mg/100g fw | Lycopene |
| Red cabbage | Primero 109 mg/100g fw | Anthocyanin (Ahmadian *et al.*, 2014) |
| Cauliflower | Pusa Sharad, 23.94 µ mol/ 100 g fw<br>Pusa Beta Kesari 1, 8-10ppm | Sinigrin<br>Beta carotene |

| | | |
|---|---|---|
| Purple Cauliflower | Graffiti, 375 mg/100g fw | Anthocyanin (Chiu *et al*., 2010) |
| Broccoli | Green broccoli, 15.2-59.3 µ mol/100 g fw<br>Purple broccoli, 26.3 µ mol/100 g fw | Glucosinolates |
| Bathua | Pusa Green, 7.6 mg/100g dw | 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<br>Sree kanaka, 90.0 mg /100g<br>Bhu Krishna, dry matter (24-25.5%), starch (19.5%), total sugar (1.9-2.2%) | β-carotene<br>Anthocyanin |
| Tapioca | Sree Visakham, 466 IU/100g | Carotene |
| Yam | Sree Neelima | Anthocyanin rich (Purple flesh) |

**Mutation breeding**

Mutation is a sudden heritable change in DNA sequence which leads to change in characteristics of an organism as classically defined by Hugo de Vries in 1900. It is the phenomenon of a sudden intermittent change in the hereditary character. In vegetable, breeding Mutagenesis has a great importance. Mutagenesis is all about treating a biological material with a mutagen in order to induce mutations. The entire operation of the mutation induction & isolation of mutants for crop improvement is called as 'Mutation breeding. In recent years, the availability of genomic sequences from many plant species and the development of a wide array of molecular-genetic technologies have enhanced our ability to detect or engineer such variation at specific genetic loci (reverse genetics), greatly expanding our capacity for both probing gene function and genetic engineering. McCallum *et al.* (2000) have introduced a new reverse genetic strategy that combines the high density of point mutations provided by traditional chemical mutagenesis with rapid mutational screening to discover induced lesions. TILLING (Targeting Induced Local Lesions IN Genomes) combines chemical mutagenesis with a sensitive mutation detection instrument. The general applicability of TILLING makes it appropriate for genetic modification of vegetable crops. There are several quality mutant rich mutant gene have been identified in several vegetable crops, used for developing multi-colour and Nutra-rich vegetable varieties. Cauliflower "Or' mutant responsible for orange curd colour (Rich in Beta-carotene) was first identified by Bradforsh in 1970 at Ontario, Canada. The introgression of this '*Or*' gene into Indian cauliflower will lead to development of β-carotene rich cauliflower. Kalia *et al*. (2018) [15, 25, 26] developed Pusa Kesari VitA-1 and promising introgression lines in cauliflower rich in beta-carotene content (8-20 ppm) in Indian cauliflower. Zhang *et al*. (263) [5, 20, 28, 29] found SCAR markers linked to "*Or*" gene inducing beta-carotene accumulation in Chinese cabbage. Zou *et al*. (2016) performed fine mapping of or gene and identified BrPro1 molecular marker in the promoter region of Bra031539 (predicted to encode CRTISO, a carotenoid isomerase specifically required for carotenoid biosynthesis) that can be used for early identification of orange head materials. Similarly, a Single dominant mutant gene "*Pr"* has been identified responsible for purple curd formation. The *Pr* mutant gene encodes MYB-1 a transcriptional factor responsible for anthocyanin accumulation in curd. Tomato two mutant gene *hp-1* and *hp-2* have been reported for carotenoid biosynthesized and used for developing carotene rich tomato hybrid. Among tuber crops one orange flesh sweet potato mutant has been recognised having rich source β-carotene (30-100 ppm) reported by La Bonte and Don (2012).

**RNAi for quality improvement**

RNAi is a promising gene regulatory approach that has significant impact on crop improvement; it permits down regulation in gene expression in a more precise manner without affecting the expression of other genes. RNAi-mediated technology has been used in the metabolic engineering of plants with respect to improvement of various traits and to target genes linked to different undesired characters. In several plants, RNAi has been used to improve their nutritional value, flavour, genetic modification of fatty acid composition, and reduce toxicity/allergenicity. Tomatoes are susceptible to changes in climate, they have autocatalytic activities of ethylene during their ripening period. Therefore, increase in shelf-life or delay in the ripening process can be achieved by the introduction of 1-aminocyclopropane-1-carboxylate (ACC) oxidase dsRNA in tomatoes, which suppresses the expression of ethylene genes. Transgenic tomato plants targeting more than one homolog than a single unit of ACC oxidase by using RNAi technology during the ripening stage would be more effective than a single homolog. The chimeric RNAi-ACS (1-aminocyclopropane-1-carboxylate synthase) construct designed to target ACS homologs effectively repressed ethylene production in tomato fruits and enhanced tomato shelf-life by 45 days. Tomato a single gene *DET1* was effectively degraded in transgenic tomato with suppressed *DET1*, with an increase in the level of flavonoids and carotenoid content and a single gene *SlNCED1* gene in tomato that encodes 9-cis-epoxycarotenoid dioxygenase, which is an important enzyme in the ABA biosynthesis, was suppressed by RNAi. The fruits showed increased accumulation of upstream compounds, chiefly lycopene and β-carotene, from these RNAi lines. RNAi technology has also been utilized to increase the carotenoid content of rapeseed (*B. napus*) by downregulating the expression of the lycopene epsilon cyclase gene (ε-CYC). Yu *et al*. (2007) highlighted the seed quality of transgenic Brassica having a higher content of β-carotene, violaxanthin, zeaxanthin, and lutein. Generally, Parthenocarpic fruits were also observed in tomato, in which genes of the *AUCSIA* family coding for 53-amino-acid-long (protein or peptide) were functionally suppressed by RNAi technology for development of parthenocarpy fruit in tomato. Some other remarkable achievement using RNAi for targeting modification tin several vegetable crops such as sweet potato (*IbSBEII* gene) for amylose content in tuber, potato (β -carotene hydroxylase gene) for increased β -carotene content and in tomato (*SIRF7* gene) for production of seedless fruit respectively. RNA silencing is a novel gene regulatory mechanism that limits transcript level by either suppressing transcription (TGS) or by activating a sequence-specific RNA degradation process (PTGS/RNA interference). RNAi technology has great promise in improvement of vegetable crops for specific traits like β carotene in tropical carrot and late bolting in Palak, radish and cauliflower etc. Post-transcriptional gene silencing (PTGS) is defined as the silencing of an endogenous gene caused by the introduction of a homologous double-stranded RNA (dsRNA), transgene, or virus. In PTGS, the transcript of the silenced gene is synthesized but does not accumulate because it is rapidly degraded. The few selected example is cited below using RNAi for quality improvement in several vegetable crops.

**Table 8:** Status of RNAi for quality improvement in vegetable crops

| Crops | Targeted genes silencing | Targeted traits |
|---|---|---|
| Tomato | Introgression<br>1-aminocyclopropane-1 carboxylate (ACC) oxidase dsRNA in tomatoes or RNAi ACS. | Suppressed Ethylene production and Improve shelf life |
| | α-Man and βHex 9 (Ethylene ripening inducer) | Enhanced shelf life. |
| | *MiR156* | Enhanced shelf life |
| | Colorless never ripe (*CNR)* gene | Enhanced shelf life |
| | *DET-1* | Enhanced flavonoids and carotenoid content. |
| | *SlNCED-1(*9-cis-epoxycarotenoid dioxygenase) | Enhanced lycopene and β-carotene content. |
| Strawberry | CHS gene | Delayed ripening in fruit, low anthocyanin production. |
| | lycopene epsilon cyclase gene (ε-CYC) | Enhanced β-carotene, violaxanthin, zeaxanthin, and lutein in seed. |
| Sweet potato | *IbSBEII* gene | Enhanced Amylose content in starch. |
| Potato | β-carotene hydroxylase gene | Enhanced β-carotene content. |
| Soyabean | *GmFNSII* gene | Enhanced flavone and isoflavone production. |
| Tomato | *SlARF7* (Auxin and gibberellic acid signaling gene), CHS gene, | Production of seedless fruit. |
| | Expressing the CP gene | TMV resistance |
| | *Lyc e 3* | Reduced allergenicity and toxicity in fruit. |

**Transgenic achievement for improvement and biofortification in vegetable crops**

The *Flavr Savr* tomato (also known as CGN-89564-2) was the first commercially grown, GE vegetable for human consumption granted a license by the US Food and Drug Administration (FDA) in 1994. These tomatoes were taken off the market by 1997 because of their nonacceptable taste and aroma. In 1995, an insect-resistant Bt-potato crop was approved by the FDA. Just after commercialization of Bt-potato a few more transgenic vegetables such as virus-resistant squash, "AMFLORA potato," etc. were developed by different Agri-based companies after receiving approvals for commercial cultivation. In China, the GM tomato "Huafan No. 1" (from Huzahong Agricultural University), which had long shelf-life characteristics, was approved for commercialization in 1996. The transgenic vegetable "AMFLORA potato" was accepted by the European Commission for production in the European Union and the registration period was 12 years. The company BASF succeeded in developing this GM plant by suppressing genes for the production of amylase; the EH92-527-1 potatoes produce over 98% of amylopectin. Some remarkable and recent achievement such as introgression of three genes, encoding phytoene synthase (*CrtB*), phytoene desaturase (*CrtI*) and lycopene beta-cyclase (*CrtY*) from *Erwinia* in potato to produce β-carotene and Introgression of AmA1 GENE FROM *Amaranthus hypochondriacus* tend to increased amino acid composition, finally lead to increased protein content. This transgenic potato shows 35–60% increase in total protein content in moderately expressed transgenic events of all varieties. The balance of amino acid is a key determinant for nutritive proteins because plant proteins typically do not provide the optimum amino acid ratios required for efficient protein synthesis in animals. Transgenic modification of plant nutritional value can be achieved by adopting the following methods: (1) improving the quality, composition, and levels of nutrients such as protein, starch, and fatty acid in different crops and (2) increasing the levels of antioxidants (e.g., carotenoids and flavonoids). A major example of transgenic plants for improved nutrition value is golden rice developed for vitamin-A deficiency. Details of crop wise development are presented in following table.

**Table 9:** Status of transgenic achievement in vegetable crops

| Crops | Genes | Biofortified traits |
|---|---|---|
| Tomato | Psy-1, *Dxs, CrtB, CrtI, CrtY, CRY-2, CYC-B, LCY-B, LCY-B, CHY-B* | Carotenoids-rich tomato |
| | *Ros1 and Del.* | Anthocyanin-rich tomato (Increased 70-100 folds higher than normal tomato). |
| | Petunia *chi-a* gene, *CHI, CHS, CHI, F3H, FLS, MYB12, STS, CHR, FNS-II, Del, Ros1.* | Flavonols-rich tomato |
| | S-adenosylmethionine decarboxylase gene (*ySAMdc; Spe2*) | Folate-rich tomato |
| Tomato | *GaLDH, GME, GCHA* and/or *ADCS* | Ascorbic acid content |
| | *HMT* and *SAMT.* | Enhanced Iodine content. |
| | beta-Lcy gene | High β-carotene content. |
| Potato | *AmA1 (Amaranthus hypochondriacus)*<br>*tar1* (tarin) gene from Colocasia esculenta | Protein rich potato |
| | *SBE I* antisense | High amylose starch |
| | *SUSI* (sucrose synthase) | Carbohydrate engineering |
| | *xylA* (glucose isomerase) | High tuber fructose |
| | *Phosphofructokinase* | Low sugar content in tuber |
| | *ZEP* gene | Enhanced zeaxanthin content. |

| | | |
|---|---|---|
| | Phytoene synthase (*CrtB*), phytoene desaturase *(CrtI)*, and lycopene betacyclase *(CrtY)* from Erwinia spp. | Enhanced β-carotene content in tuber. |
| | *PSY*, phytoene desaturase, and lycopene β-cyclase | Enhanced β-carotene content. |
| | *Bch* gene (Silencing using RNAi Technology) | Enhanced β-carotene content |
| | *Or* gene | Enhanced β-carotene and phytoene, phytofluene, and z-carotene. |
| | Strawberry *GalUR* gene | Enhanced Ascorbic acid content. |
| | *StMGL1* gene silencing using RNAi technology, (*CgS)* gene | Enhanced the level of methionine content. |
| | *CHS* and *CHI* genes | Enhanced phenolic acid, and anthocyanin, fibre and fructan and inulin content. |
| Carrot | *SCAX1* | Enhanced Ca content |
| Sweet potato | *IbOr*-Insunder. | Enhanced lutein and total carotenoids content in tuber. |
| | *IbMYB1* gene | Anthocyanin accumulation in root. |
| Cassava | *FEA1* gene (*Chlamydomonas reinhardtii*) | Enhanced Fe accumulation. |
| | *ZIP (ZRT, IRT*-protein) and *CDF* | Enhanced Zn accumulati0on. |
| | *cis-β-carotene* | Enhanced β-carotene content. |
| | *Crt-B* | Enhanced β-carotene content |
| Lettuce | Soybean ferritin gene | Enhanced Fe uptake. |

**Genome editing**

**Tools for biofortification in vegetable crops**

TGE facilitates targeted and stable editing of DNA using engineered nucleases including mega nucleases, ZFNs, TALENs and CRISPR/Cas9 nucleases. Among others CRISPR/Cas9 is simple and cheap design of a single guide RNA (sgRNA) that is complementary to the target sequence. Genome editing with site-specific nucleases allows reverse genetics genome engineering and targeted transgene integration experiments to be carried out at specific locations in the genome of both model and crop plants, as well as in a variety of other organisms. The four steps necessary for modifying a plant gene through genome engineering include: (1) designing and developing an engineered nuclease construct, (2) delivering the construct and perhaps donor molecule into the plant (typically by genetic transformation), (3) inducing nuclease expression, and (4) screening the plants for the desired DNA sequence change. Genome editing by CRISPR/Cas (Clustered regularly interspersed short palindromic repeats/CRISPR—associated proteins) is one such revolutionary technology in diverse plant species because of its easy manipulation, high efficiency and its wide application in functional studies of genes and genetic crop improvement. CRISPR/Cas9 tool contains a non-specific Cas-9 nuclease and a single guide RNA which is specific to target gene directs Cas9 to the specific genomic location creating double-strand breaks and subsequent repair process creates insertion or deletion mutations. The first CRISPR/Cas9-mediated genome editing in vegetable crops was reported in tomato in 2014 where *ARGONAUTE7 (SlAGO7)* gene involved in leaf development was targeted. Later on, genes for anthocyanin biosynthetic pathways in tomato such as Anthocyanin 1 (*ANT1*), Phytoene desaturase (*SlPDS)*, Phytochrome interacting factor (*SlPIF4*), and Phytoene synthase (*PSY1*) functioning in carotenoid biosynthesis were mutated by CRISPR/Cas9. Potato starch quality is an important factor for food as well as many commercial product developments, in this direction, the waxy genotypes of potato, producing only amylopectin containing starch was developed by mutating granule bound starch synthase (GBSS) gene using CRSIPR/Cas9 and another successfully achievement made by introduced mutations in *SlIAA9- a* key gene controlling parthenocarpy and their results showed that, regenerated mutants exhibited seedless fruit-a characteristic of parthenocarpic tomato. The status of genome editing for biofortification in vegetable crops are cited below.

**Table 10:** Status of genome editing in vegetable crops

| Crops | Traits | Targeted genes | Method | References |
|---|---|---|---|---|
| Citrus | Carotenoids | | CRISPR/Cas-9 | Jia and Wang 2014 [8, 19, 20, 22, 29]. |
| Tomato | Anthocyanin | *ANT-1* | CRISPR/Cas-9 | Cemak *et al*., 2015 |
| Tomato | Carotenoids | *SIPDS* | CRISPR/Cas-9 | Pan *et al*., 2016 [22] |
| Tomato | Carotenoids | *SIPIF4* | CRISPR/Cas-9 | Hayut *et al*., 2017 [10] |
| Tomato | Fruit color | *PSY* | CRISPR/Cas-9 | Filler Hayut *et al*., (2017) [10] |
| Tomato | Pink tomato fruit color | *SlMYB12* | CRISPR/Cas-9 | Deng *et al*., (2018) [8] |
| Tomato | Fruit ripening | *rin* | CRISPR/Cas-9 | Ito *et al*. (2015) [14] |
| Grapes | Carotenoids | | CRISPR/Cas-9 | Nakajma *et al*., 2017 |
| Watermelon | Carotenoids | | CRISPR/Cas-9 | Hayut *et al*., 2017 [10] |
| Tomato | Parthenocarpy | *S1IAA9, S1AGL6* | CRISPR/Cas-9 | Ueta *et al*., 2017 |
| Potato | Starch quality | *GBSS* | - | Andersson *et al*. (2017) [1] |
| Potato | Starch biosynthesis | *StALS1, StALS2* | - | Kusano *et al*., (2018) |
| Watermelon | Carotenoid biosynthesis | *Phytoene desaturase (ClPDS)* | CRISPR/Cas-9 | Tian *et al*. (2016) [19, 28] |
| Carrot | Anthocyanin biosynthesis | *F3H* | | Klimek-Chodacka *et al*., (2018) |

**Molecular marker and genome sequencing and transcriptome analysis**

Genomics by sequencing (GBS) is a simple, affordable and robust procedure for SNP discovery and mapping. This approach is suitable for population studies, germplasm characterization, breeding, and trait mapping in diverse organisms. The advent of molecular breeding approaches has helped to explore the variation in gene pools and combine it with conventional breeding methods. Study of genome wide single-nucleotide polymorphism (SNP) opens the field of comparative genetic mapping. Detection of quantitative trait loci (QTL) in populations helps to locate the linkage groups between genomes. Potential molecular markers can be used for selection and cloning of important genes for desired trait(s). In Brassica, expressed sequence tags (ESTs) are being used for deployment of microarray platforms for high-throughput transcriptome analysis. Genome sequencing used to study complete genome and gene functions and an efficient tool for revealing molecular mechanisms of plant growth, development and differentiation. The discovery of molecular marker and NGS platform provide a flexible platform for understanding the genetic of complex trait or biochemical mechanism lying under various quality trait and biosynthesis pathway for various carotenoid and flavonoid. Transcriptome sequencing has been a cheaper alternative comparatively. Transcriptomic analysis understanding the quality related traits and some remarkable achievement have been made such as in sponge gourd a cultivars '*Luffa* cultivar 'Fusi-3' analyzed transcriptomic changes occurring during the browning of fresh-cut fruits and identified 11 genes from five gene families (*i.e*., PPO, PAL, POD, CAT and SOD)-associated with enzymatic browning of flesh, similarly in Pea three Mutant alleles at three loci *r* (referred to ADP-glucose pyro-phosphorylase), *rb* (starch branching enzyme) and *bsg* (phosphor-glucomutase) affect starch and sugar synthesis and 795 novel genes and large number of DEGs related to carotenoid biosynthesis, plant hormone pathways, and sugar and cell wall metabolism during fruit ripening have been identified in watermelon respectively. Molecular Marker such as RAPD, RFLP, SSR, CAPS and SNP are widely used for tagging and used to study linkage with gene responsible for high nutraceuticals and edible colors using mapping population. In India Kalia *et al*. (2018) [15, 25, 26] at IARI designed and orient bio-fortification of Indian cauliflower with a carotene enhancing native '*Or*' gene following MAS. Limited number of molecular markers have been identified linked to quality traits in some selected vegetable crops such as SCAR marker linked 'Or' gene responsible for Beta-carotene accumulation and a ISSR marker has been identified found to be linked with 2-propenyl glucosinolate content in *Brassica* vegetables.

**Molecular breeding for improvement of quality traits in tomato**

Using crop wild relatives (CWR) in crop improvement is much more difficult than breeding with domesticated varieties. Pre-breeding aims to isolate desired genetic traits (e.g. disease resistance) from unadopted material like CWR and introduce them into breeding lines that are more readily crossable with modern, elite varieties. Broccoli Two QTLs such as QTL1 and QTL2 have been identified in *Brassica villosa* (Rich source Gluraphone) and introgressed in cultivated types for developing glucoraphanin rich content in Brassica vegetable crops. The tomato rank high in nutritional value; one medium fresh tomato (135 g) provides 47% RDA of vitamin-C, 22% RDA vitamin-A, and 25 calories. However, by virtue of volume consumed, it contributes significantly to the dietary intake of vitamins A and C as well as essential minerals and other nutrients. Many QTL have been mapped for various quality traits in tomato such as for fruit size *fw1.1* (explaining 17% of the variation), *fw1.2* (13%), *fw2.1* (12%; previously known as locule number, lc), *fw2.2 (23%), fw3.1 (12%)* and *fw11.3* (37%; previously known as *fasciated, f)*. Of these QTLs, *fw2.1 (lc)* and *fw11.3 (f)* are associated with an increase in locule number have been mapped; fruit shape such as *pr* (pyriform), *o* (ovate), *bk* (beaked tomato), *n* (nipple-tip tomato), *f* (fasciated), and *lc* (for locule number). During the past two decades, a few of these genes and several other genes and QTLs affecting fruit shape in tomato have been located on tomato molecular linkage map and/or cloned and characterized at the molecular level. Tomato fruit colour has recently increased as the health benefits of lycopene, the major carotenoid in tomato that is responsible for the red fruit colour, has become more obvious. As indicated earlier, several major genes with significant contribution to high contents of fruit lycopene (e.g., *hp-1, hp-2, dg* and *Ogc*) and other carotenoids (e.g., β-carotene, B) were previously identified and mapped onto the classical linkage map of tomato.

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