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Breeding strategies in vegetable crops for protected cultivation for yield and bio-chemical traits

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Abstract
Protected cultivation of high-value vegetables has gained significant attention and potential worldwide. The primary reason for this is enhancement of fruit quality and protection from insects, winter, rains etc. These technologies not only benefit larger-scale farming but also offer advantages to growers with smaller landholdings, as they can achieve higher productivity levels that are economically relevant to agriculture (Sabir *et al*., 2013) [38]. Keeping in view the majority of Indian farmers occupy small land holding (< 1.0 ha), protected cultivation may prove an Amritkaal technology for them due to its great potential to enhance their income by off-season cultivation for fetching 3-4 times high prices, raising off-season nursery for early crop raising, hybrid seed production of vegetable crops, enhanced per squaremeter productivity, multilayerd cropping etc.

Not only tomato, parthenocarpic cucumber, capsicum under crop rotation may be good options for protected cultivation. But the major challenge is non-availability of hybrids of these crops, suitable for Indian agro-climatic conditions. No major breeding efforts have made for breeding under these agro-climatic conditions. The hybrids/varieties released so far have been developed through breeding under open field conditions. To realize the full potential of hybrid, it should be developed/bred in specific environment for which it is being developed. Serious efforts are needed for this field. Presently, Cherry tomato Varieties like "Sweet 100," "Sun Gold," and "Yellow Pear" are excellent choices for greenhouse cultivation due to their compact growth and high yield potential. Arka meghali, arka surbhi, avtar, FA 180, GS 600 are some other tomato varieties. Cucumber Varieties like "Marketmore 76" and "Diva" are long and slender, making them ideal for vertical trellising in nethouses. Bell peppers varieties like "Ace" and "King Arthur" which produce good-sized fruits and have a manageable growth habit. In the context of developing high-color chilli varieties for extracting paprika oleoresin, the focus has shifted towards natural colors for food coloring in general (Prasath *et al*., 2008) [34]. The development of parthenocarpic fruit in cucumbers has been determined to be regulated by an incompletely dominant gene. This conclusion is drawn from the observation of the gene's segregation pattern in the F2 population and through test crosses conducted with the gynoecious and parthenocarpic cucumber line PPC-2 (Jat *et al*., 2017) [16].

Keywords: Parthenocarpy, breeding, cherry tomato, gynoecious, nematode

Introduction
Greenhouse cultivation or protected cultivation has emerged as a modern and widely adopted approach to achieve high-quality and high-yield in horticultural crops. Over the past few decades, the Controlled Environment Agriculture (CEA) technique has gained global popularity. Protected cultivation offers several advantages including increased productivity, efficient water and land usage and environmental protection (Jensen, 2002). The method involves cultivating horticultural crops in a controlled environment where various factors such as temperature, humidity, light, soil, water and fertilizers are carefully regulated to optimize yields and enable continuous production even during off-seasons. By embracing protected cultivation technology, farmers can anticipate improved profits through the production of superior-quality crops. In recent years, protected cultivation has gained significance in vegetable cultivation leading to increased production, higher yield and quality, enhanced productivity per unit area and protection from pests and diseases. Although protected cultivation is popular among farmers, there has been no specific variety or hybrid developed exclusively for polyhouse conditions.

The major crops commonly cultivated in greenhouses include tomatoes, cucumbers, capsicums and bitter gourds among others.

Tomato

Tomato (*Solanum lycopersicum.* L) is among the most widely cultivated vegetables and ranks second in production after potatoes in India. India contributing 10% to the global production of 170.75 million tonnes per annum holds the second position in tomato production after China, producing 18.74 million tonnes annually (NHB, 2017) [14]. Due to its nutritional value, tomatoes are considered protective food containing essential nutrients like vitamin C, lycopene and β-carotene, offering various health benefits. They are grown for both fresh consumption and processing into various products. Indeterminate types of tomatoes have shown to be well-suited for polyhouse cultivation, ensuring higher yields and a prolonged growing season.

Tomato plants in net houses are trained to a single stem and supported by strings or wires, achieved by removing lateral shoots and wrapping the main stems around the overhead wire system. The controlled environment in net houses allows for tomatoes to develop more vibrant colors compared to field-grown ones. By modifying the natural environment to create optimal conditions for plant growth, net houses facilitate the production of high-quality tomatoes suitable for both domestic consumption and export. Research has shown that net house-grown tomatoes tend to perform better and yield higher fruit compared to those cultivated in open fields. In net houses, tomatoes can be grown successfully during the off-season, resulting in increased fruit yield. Additionally, the risk of frost damage and yield loss, which can be significant in open field conditions during December and January is minimized in protected environments like net houses. Arka meghali, arka surbhi, avtar, FA 180, GS 600 are some other tomato varieties.

Cherry tomato

Cherry tomato plants produce visually appealing and delicious fruits that are highly favored by consumers. They come in determinate, semi-determinate and indeterminate growth habits featuring long racemes and numerous fruits with intense color and flavor, weighing between 10 and 30 grams. Cherry tomatoes exhibit disease resistance and can tolerate high relative humidity levels. They are rich in nutrients particularly vitamin C with content exceeding 57 mg/100 g fresh weight. The number of fruits per cluster can vary significantly ranging from 15 to 50. Cherry tomatoes are commonly consumed as fruits rather than vegetables. They are well-suited for regions with high humidity (above 80%) and exhibit a lycopene content that surpasses 10mg/100g fresh weight which is considered high.

Cherry tomatoes are widely regarded and consumed as fruits rather than vegetables. These tomatoes possess potent anti-cancer properties particularly beneficial for countering mouth cancer and addressing sour mouth issues. Additionally, they can help prevent acidosis, a common condition leading to various health ailments like headaches, fatigue, sleeplessness, absorption problems, arteriosclerosis, muscular aches and loss of calcium from the bones. Incorporating tomatoes into one's diet can contribute to maintaining an alkaline balance and mitigate these problems. Another category known as 'cherry tomatoes' is considered a high-value crop. These tasty and numerous small-sized fruits grow in clusters along the stems and branches of the tomato plants. Cherry tomatoes, being smaller garden varieties, are often favored in salads. The ones that can be eaten whole are typically preferred over those requiring cutting before consumption. For many consumers, fruit size is an important factor. Smaller-sized varieties, ranging from ½ to ¾ inch are ideal for whole consumption, while larger fruits need to be cut. Breeding efforts for cherry tomatoes focus on various aspects such as increasing yield, enhancing the number of fruits, improving fruit shape and weight, ensuring uniform fruit color, promoting earliness in fruiting, enhancing fruit firmness and shelf life, elevating total soluble solids (TSS) content maintaining a favorable solids/acid ratio, and developing resistance to diseases and pests.

The lycopene content (LYC) found in tomato fruit plays a significant role as a source of lipid-soluble antioxidants in the human diet, offering protection against oxidative chain reactions (Rousseaux *et al*., 2005) [37]. On the other hand, the total soluble solid content (SSC) constitutes a crucial aspect of tomato flavor and aligns closely with consumers' perception of internal quality (Arazuri *et al*., 2007) [2]. LYC and SSC are primary indicators of tomato fruit quality. However, the quantitative variation observed across different varieties is influenced by a combination of genetic and environmental factors, making the inheritance of tomato fruit quality a complex process. Consequently, overcoming the genetic linkage between fruit quality traits presents a challenge when using conventional breeding methods. To address these challenges, the application of quantitative trait locus (QTL) mapping is a valuable approach for identifying major genes and functional markers that can enhance the control of quantitative traits and improve tomato fruit quality.

Breeding for Cherry tomato

Cherry tomato, scientifically known as *Solanum lycopersicum* var. cerasiforme are small sized fruits that grow in clusters on the stems and branches of the tomato plants. These small sized tomatoes are commonly used in salads, making their size a crucial factor from the perspective of consumers. Breeding objectives aimed at enhancing cherry tomatoes includes increasing yield, increase the number of fruits, fruit shape and weight, achieving uniform fruit color, ensuring early ripening, fruit firmness, extending shelf life, high total soluble solids (TSS) and developing resistance against diseases and pests (Lenecci *et al*. 2006) [24]. In India, the breeding of polyhouse tomato began with the aim of creating pure lines (PTP1, PTP2, PTP3, and PTP4) and a hybrid (PTPH 1) characterized by large fruit size (ranging from 150 to 1,200 grams), a thick pericarp (0.8 to 1 cm), clusters of 4-5 fruits, long fruiting period 8-9 months and plant height (20-25 feet). To achieve this, the Punjab Red Cherry variety was developed using a cross between *Solanum lycopersicum* and S. pimpinellifoilum through the pedigree selection method. This resulting indeterminate variety of cherry tomato features dark green foliage and is well-suited for protected cultivation. In addition, the Punjab Agricultural University (PAU) has recommended two indeterminate table tomato varieties suitable for protected cultivation in the state: 'Punjab Gaurav' and 'Punjab Sartaj.' The Indian Agricultural Research Institute (IARI) in New Delhi has introduced 'Pusa Golden Cherry Tomato-2,' which is a yellow-colored cherry tomato variety. For greenhouse cultivation in India, cherry tomato varieties such as "Sweet 100," "Sun Gold," and

"Yellow Pear" are excellent choices due to their compact growth habits and high yield potential.

Capsicum

The Capsicum genus is native to the Western Hemisphere and was first encountered by Christopher Columbus in the New World. Initially, it was mistaken for black pepper. The indigenous people of the Americas have been consuming Capsicums since around 7000 B.C. It is believed that these plants were later introduced to Europe and subsequently spread to Asia and Africa within a span of approximately 150 years. Currently, it is widely acknowledged that the Capsicum genus comprises about 25 wild species and five cultivated species. Among these, *C. annum* is the most commonly grown species, known for producing both pungent and non-pungent fruits.

The Capsicum genus comprises several groups, including bell pepper, paprika, chilli, and red pepper. All five cultivated species of Capsicum have genotypes with either pungent or non-pungent fruits, making it challenging to assign a particular genotype to a specific cultivated species based on fruit size, shape and pungency. Paprika is produced from mild pungent or non-pungent fruited varieties of Capsicum. However, in international trade paprika specifically refers to non-pungent fruits or their powder or oleoresin extracts (Bosland and Votava, 2000) [6]. Chilli holds a unique place in the world diet, as it is consumed in both its ripe dried form (as a spice) and as green fruits (as a vegetable). Additionally, chilli is used as a natural red colorant serving as an essential condiment in various foods. India boasts a rich variety of Capsicum with diverse quality attributes and is the leading consumer and exporter of chilli. Apart from dried chilli and chili powder, oleoresin of chili with varying levels of pungency is also exported in significant quantities. The chilli colorant, which adds appealing color, delightful flavor and aroma, finds numerous applications in food, pharmaceutical, and cosmetic products.

Pungency is characterized as a sharp, penetrating, stinging, or stimulating quality, capable of exciting the senses (Bosland, 1995) [5]. In the case of chilli, pungency is attributed to specific chemical compounds known as capsaicinoids, which are alkaloid compounds found exclusively in the Capsicum plant genus. Among these capsaicinoids, capsaicin also referred to as n-vanillyl-8-methyl-6-(e)-noneamide, stands out as the most pungent. While capsaicin exhibits limited solubility in water, it is highly soluble in fats, oils and alcohol. Other members of the capsaicinoid group include Dihydrocapsaicin, Nordihydrocapsaicin, Homocapsaicin and Homodihydrocapsaicin. To measure the pungency level of chillies, the Scoville organoleptic test was introduced offering a systematic and refined approach. This test was the initial laboratory method employed for measuring pungency. One ‘Scoville Heat Unit’ corresponds to approximately fifteen parts per million (ppm) of capsaicin. Pure capsaicin has the highest Scoville rating possible, reaching 15 million units (Scoville, 1912) [40]. However, in contemporary practice, the most commonly used and reliable method for estimating pungency is high-performance liquid chromatography (HPLC).

Breeding for capsicum

The breeder needs to set objectives and specific goals based on current market trends. In sweet pepper breeding for greenhouse cultivation, the primary objectives are similar to those for outdoor breeding with a few aspects requiring more focused attention. These include aspects like fruit quality, encompassing factors such as shape, size, color, pericarp thickness, plant growth habit, resistance to pests and diseases, early maturity, high yield potential, tolerance to environmental stress, and overall plant morphology. AVRDC-The World Vegetable Centre maintains an extensive collection of accessions in various countries around 7,500 base and active collections. Private seed companies in Asia and North America also maintain substantial collections for use in their breeding programs. Similarly, the Southern Plant Introduction Station in Griffin, Georgia, USA, operating under GRIN-Germplasm Resources Information Networks has evaluated approximately 3,000 accessions for various traits. Numerous other institutes worldwide, such as the Centre for Genetic Resources in Wageningen, The Netherlands (CGN) and the Central Institute of Genetics and Germplasm in Gatersleben, Germany, are engaged in similar germplasm preservation and research activities. In India, the National Bureau of Plant Genetic Resources in New Delhi and the Indian Institute of Vegetable Research primarily focus on germplasm activities related to bell peppers and sweet peppers.

Table 1: Hybrids/varieties of capsicum

Crop	Hybrids/Varieties
Capsicum	Indra, Orobelle, Triple star, Inspiration, Pasarella (Red), Bomby, Sunnyez, Yamuna (Green), Swarna, Bachata (Yellow), Natashaetc, California Wonder, Arka Mohini, Arka Gaurav, Arka Basant, Bharat, Kt-1, Pusa Deepti, Solan Hybrid1

Breeding Methods

Sweet pepper breeding primarily relies on few traditional methods, such as Selection, Pedigree breeding, Single Seed Descent (SSD) method, Backcross breeding, Recurrent Selection, and heterosis breeding. However, in recent times, biotechnological approaches have gained popularity as a significant tool for expediting the development of cultivars or varieties with greater efficiency and assured success. Marker-assisted breeding has also proven valuable, enabling the incorporation or introgression of specific desirable genes from wild relatives into domesticated varieties. It's worth noting that Capsicum, the genus to which sweet peppers belong, can withstand inbreeding to a certain extent, although there is some degree of heterosis observed for reproductive traits, including components related to seed yield. (Mishra *et al*. 1991) [28].

In bell pepper, hybrid varieties have been successfully developed (table-1) but a significant challenge in their adoption for commercial cultivation lies in the substantial seed expenses. This is primarily due to the absence of cost-effective and efficient technology for hybrid seed production. Given that developing resistant or tolerant cultivars is among the most effective strategies to mitigate losses resulting from disease and insect infestations, there is a pressing requirement to create new cultivars endowed with resistance to key pepper diseases and insect pests specifically grown for greenhouse cultivation. (Pereira *et al*. 2011) [33]. Key characteristics of Capsicum varieties bred for polyhouse cultivation includes several factors: Early maturity, fruit quality, diseases and pests resistant, tolerance to various environmental stresses like heat, water scarcity, and salinity, an indeterminate growth pattern, vigor and consistency throughout the season. Desirable fruit attributes include uniformity in size and shape, preferably with four lobes, a substantial fruit weight (> 50 grams), uniform coloring upon full ripeness, extended shelf life (lasting more than 5 days under normal conditions), and short intermodal lengths of 7 to 10 centimeters, with plants reaching a maximum height of 10 feet. Uniform blocky fruit with dark red color, vigorous shoot growth are desired Capsicum hybrids with high yield potential (> 100 t/ha)".

Gene action

It has been widely recognized that a single dominant gene C determines the presence or absence of pungency in the fruits (Blum *et al*., 2002) [4]. However, in pungent varieties, the level of pungency is inherited in a quantitative manner and strongly influenced by environmental factors (Zewdie and Bosland, 2000) [6]. Previous studies have suggested that mature red color is dominant over yellow color and controlled by a single gene (Y). According to Hurtadol Hernandez and Smith, 1985 [15] findings, mature fruit color is influenced by three independent pairs of genes cl- codes for pseudo response regulator 2, c2- codes for Phytoene synthase and Y- codes for gene capsanthin capsorubin synthase gene. The presence of dominant alleles at these three loci results in red mature fruits while recessive alleles lead to white mature fruits.

Cucumber

Inheritance of gynoecious in cucumber

Cucumber (*Cucumis sativus* L.; 2n = 2x=14) is a significant vegetable crop worldwide. Its origin can be traced back to India where it evolved from its wild progenitor, *Cucumis sativus* var. *hardwickii* R. which can still be found in the southern foothills of the Himalayas. Cucumber is primarily cultivated for its tender fruits which are commonly used in salads, pickles and rayata preparations. In India, cucumber cultivation is practiced across a range of altitudes from higher elevations to plains both in open fields and under protected conditions. However, the cultivated cucumber has a narrow genetic base with only 3-8% polymorphism within the cultivated genotypes and 10-25% between botanical varieties (Behera *et al*., 2011) [3].

The cucumber plant produces three types of flowers: Male, female, and bisexual. Due to their diverse floral sex types, cucumber and melon have long been important model systems for sex determination studies (Tanurdzic and Banks, 2004) [48]. In the early stages of development, the flower buds of cucumber contain primordia of both stamen and pistil and sex determination occurs when either the staminate or pistillate primordia development is selectively arrested just after the bisexual stage (Kater *et al*., 2001) [20]. Sex determination takes place in flower buds located at 10-12 nodes below the apical meristem regardless of the plant's age (Saito *et al*., 2007) [39]. In commercial cucumber cultivars, the sex form (gynoecious or monoecious) and the intensity of sex expression are important factors as they directly impact the harvesting date and productivity of the crop. The potential for developing tropical gynoecious hybrids in cucumber has also been demonstrated (More, 2002) [29].

Although the cucumber plant is originally monoecious the presence of three flower types on the plant can lead to seven different sex types of cucumber plants (Liu *et al*., 2008) [26]: Androecious (only male flowers), gynoecious (only female flowers), monoecious (male flowers at the base and female flowers at the top of the main stem), hermaphroditic (only bisexual flowers), andromonoecious (male and bisexual flowers), gynomonoecious (female and bisexual flowers), and trimonoecious (male, female, and bisexual flowers). The sex expression in cucumber plants is controlled by three genes, F, M and A (LI *et al*., 2008; LI *et al*., 2009) [25, 27]. The F/f gene regulates the degree of female flower expression while the M/m gene controls bisexual flower expression. The F locus influences the degree of femaleness (FF > Ff > ff) and the M locus determines whether flowers are unisexual (M_) or bisexual (mm). The A locus conditions an increased male tendency when a plant is homozygous recessive aa and ff. Interactions between these loci give rise to the various sex types found in cucumber (Staub *et al*., 2008) [45]. The dominance of gynoecy over monoecy results in highly productive F1 plants. Sex expression in cucurbit species can be influenced by plant hormones and environmental factors (Tanurdzic and Banks, 2004) [48].

Parthenocarpy in gynoecious cucumber

India being recognized as the birthplace of cucumber possesses a wide array of genetic diversity and variability in terms of both growth and fruit characteristics. However, this valuable diversity has not been fully utilized for its genetic improvement. A significant challenge faced by cucumber breeders is the development of gynoecious varieties with parthenocarpic traits. These varieties are sought after as parents in F1 hybrid development to achieve higher yield, early maturity, uniformity and suitability for protected cultivation (Jat *et al*., 2017) [16]. Therefore, there is a crucial need to create gynoecious hybrids with parthenocarpic traits that can be effectively used on a commercial scale particularly in the plains of northern India. This is because most of the Indian cucumber cultivars are monoecious and lack parthenocarpic traits making them unsuitable for growth under protected conditions as they require pollination for fruit set.

Gynoecy combined with parthenocarpy in cucumber is a crucial factor affecting both yield and quality. This high-value vegetable crop is particularly well-suited for off-season cultivation under protected conditions, as parthenocarpic varieties do not require pollination for fruit setting. Additionally, these fruits are known for their mild flavor, seedlessness and thin skin that eliminates the need for peeling. The expression of the parthenocarpic trait is also influenced by plant growth regulators, and its stability is significantly affected by various environmental factors. This physiological process is intricate and can be influenced by environmental, physiological and genetic factors. Some studies have suggested that low temperature, light exposure and exogenous hormone application can induce parthenocarpy. However, the exact genetic mechanism responsible for parthenocarpy in cucumber remains unclear.

Understanding the genetics of parthenocarpy is of paramount importance for an effective breeding approach aimed at developing stable parthenocarpic cucumber lines. Considering all the aforementioned factors and recognizing the significance of cucumber as a vital vegetable crop for protected cultivation, it was deemed essential to conduct an experiment to study the inheritance of parthenocarpy in a cross involving a gynoecious parthenocarpic line (Figure 1: gynoecious line GBS-1).

Fig 1: Gynoecious line GBS-1

Breeding for Cucumber

Parthenocarpy with gynoecy in cucumber (*Cucumis sativus* L.) was reported long back by Sturtevent (1890). Parthenocarpy can be defined as the ability to develop fruits without pollination and fertilization. Parthenocarpy term was coined by Noll (1902) in cucumber. True breeding parthenocarpic lines in cucumbers are reported from GBPUA&T, Pant Nagar (Singh 2012) [44], MPKV, Rahuri and IARI, New Delhi (More and Budgujar 2002) [30]. These lines were employed in a heterosis breeding program to create F hybrids. The growth habits play a crucial role in breeding efforts as they significantly contribute to increased yield and a prolonged availability period. Parthenocarpic gynoecious cucumber varieties are well-suited for polyhouse cultivation because they produce fruits automatically without the need for pollination (Karlsson 2016) [19]. However, the primary focus is on developing parthenocarpic gynoecious hybrids. The 'Pc' gene, responsible for parthenocarpy, and the 'F' gene, responsible for short inter-nodal length, can be effectively harnessed using pure line and backcross breeding techniques to establish these traits in parthenocarpic cucumber populations (Hou *et al*. 1995) [13].

Consumers demand cucumbers having green color, cylindrical shape, tender skin, sweetness (without cucurbitacin bitterness), and a crisp fuit (More and Munger 1986) [31]. Two parthenocarpic cucumber, i.e. Pant Parthenocarpic Cucumber-2 and Pant Parthenocarpic Cucumber-3 and tomato variety Pant Polyhouse hybrid tomato-1 and Pant Polyhouse hybrid tomato-2 were released. In cucumber gynoecious parthenocarpic variety Pusa Seedless (DPaC-6) has been release by IARI for growing in protected conditions (More 2002) [29]. Cucumber Varieties like "Marketmore 76" and "Diva" are long and slender, making them ideal for vertical trellising in net houses.

Several hypotheses were formulated regarding causes and function of abortion (Stephenson 1981) [46], but parthenocarpy has received much less attention. Hypotheses in relation to abortion can be placed into three groups,

1. environmental uncertainty
2. male role of hermaphroditic flowers, and
3. Improvement of the quality of seed produced through selective abscission (Stephenson 1981) [46].

Breeding against virus

Backcross

Backcross breeding is a method utilized to introduce specific easily inherited traits from unadapted donor parents into recipient lines. It involves a series of repeated crossings with the recipient line (recurrent parent), followed by selecting and retaining the desired trait being transferred (Kenaschuk, 1975) [21]. To achieve successful backcrossing, the transferred trait must remain effective across several generations and a sufficient number of backcrosses must be carried out to regain the desirable traits of the recurrent parent (Harlan *et al*., 1992) [12]. The concept of backcrossing for crop plant development was first proposed by Harlan and Pope in 1922 [12]. They observed that backcrossing had long been utilized in animal breeding to stabilize traits and highlighted its potential value in agriculture (Harlan *et al*., 1992). [12] After conducting a single round of backcrossing they demonstrated that a portion of the resulting offspring displayed the phenotypic characteristics of one parent while inheriting the desired genetics from the other. This provided breeders with an effective and cost-efficient means to introduce the desired trait into the preferred genetic background (Fehr, 1991) [9].

Procedure for backcross method

The backcross breeding method involves crossing a donor parent (possessing a gene of interest) with a recurrent parent (an elite line that could be improved by incorporating the gene of interest). The resulting progeny from this cross is then crossed back to the recurrent parent hence the term "backcross." This cycle is repeated multiple times, with each successive generation being selected for the specific trait of interest and then crossed back to the recurrent parent. This process continues until a line is developed that closely resembles the recurrent parent while carrying the desired gene from the donor parent. By the BC4 generation, the lines are more than 96% genetically similar to the recurrent parent. To expedite the backcrossing process marker-assisted backcrossing, also known as background selection can be employed. This technique uses genetic markers to efficiently identify and select progeny with the desired trait and genetic background. The backcrossing method is also utilized for resistance gene pyramiding which involves incorporating multiple resistance genes into a single genetic background for enhanced disease resistance.

Demerits

- The newly developed variety will not surpass the recurrent parent in any aspect except for the specific transferred trait.
- The process requires extensive crossing efforts and conducting 6-8 backcrosses can be challenging and time-consuming.
- Occasionally, along with the desirable gene undesirable genes may also be transferred due to genetic linkage.
- Throughout the backcrossing program, the recurrent parent may have been replaced by other varieties that exhibit superior yield and other favorable characteristics.

Gene pyramiding

The concept of gene pyramiding was initially proposed by Watson and Singh in 1953. Gene pyramiding is a breeding method that involves the deliberate incorporation of multiple desirable genes or QTLs from different parents into a single genotype to enhance specific or multiple traits through conventional breeding (Yunbi Xu, 2010) [49].

Advantages of gene pyramiding

1. Enhancing trait performance by combining two or more complementary genes.
2. Remedying deficits by introgressing genes from other source.
3. Increasing the durability.

Types of gene pyramiding

1. Conventional technique.
2. Molecular technique.

Molecular markers also known as DNA tags play a crucial role in this process by identifying genes linked to specific traits of interest. This method is particularly beneficial for incorporating genes affected by environmental factors, disease and pest resistance genes and for accumulating multiple resistance genes within a single cultivar a process known as gene pyramiding.

At AVRDC, a three-parent cross (denoted as CLN3241 in June 2012) was created using the pedigree selection method in October 2008. The parents involved in the cross were CLN2777G, which carries homozygous resistance genes for Bwr-12 (bacterial wilt), Ty-2 (tomato yellow leaf curl disease) and Tm22 (TMV); G2-6-20-15B, which is homozygous for Ty-3 (tomato yellow leaf curl disease resistance); and LBR-11, which carries homozygous resistance genes for Ph-2 and Ph-3, I2 (resistance to race 2 of the fusarium wilt pathogen) and Sm (resistance to the gray leaf spot pathogen). Through the breeding program, five F7:8 lines were developed that showed high yield and resistance to multiple diseases. These lines have the potential to be released as inbred line cultivars, hybrid parental lines or utilized as breeding stock (Hanson *et al*., 2016) [10].

Marker Assisted Selection: Marker Assisted Selection (MAS) is a method of selecting plants with desired traits based on the banding pattern of linked molecular markers (DNA). MAS relies on the concept that the presence of a marker closely linked to the gene of interest can indicate the presence of that gene. In breeding programs, MAS has been used to combine resistance genes against various pathogens in crops. Similarly, resistance to Tomato spotted wilt virus in tomatoes has been achieved using CAPS markers (Langella *et al*., 2004) [23]. In another example, monogenic recessive resistance to Pepper leaf curl virus in Capsicum was developed through an interspecific cross with the assistance of a marker (Rai *et al*., 2014) [36].

Problems

Root knot nematode: The interaction between *Meloidogyne* species and tomato has been ongoing for many centuries. These nematodes are responsible for causing significant economic losses globally in various agricultural crops including tomato. This review aims to offer insights into the damage caused by *Meloidogyne* spp. on tomato cultivars (*Solanum lycopersicum*) and to compile various studies conducted on managing *Meloidogyne* spp. on tomato with a specific focus on the Mi resistance gene.

Numerous research studies have been conducted to evaluate the impact of root-knot nematodes on different tomato cultivars revealing potential yield losses ranging from 25% to 100%. Various management approaches including the use of synthetic nematicides and soilless cultures have been explored and made available for addressing *Meloidogyne* spp. issues. Successful management has been achieved through the deployment of resistant commercial cultivars and rootstocks carrying the Mi gene against *Meloidogyne incognita, M. javanica* and *M. arenaria*. However, it is essential to note that virulent populations have also been identified. The conventional reliance on a single strategy for managing root-knot nematodes is outdated and it is more effective to adopt an integrated management approach that considers the entire disease management system. In the future, it is crucial to exercise caution in directly extrapolating tolerance limits determined in other contexts, as several factors such as initial inoculum type physiological races of *Meloidogyne* spp. environmental conditions, cultivar types and experimental approaches can influence the outcome. There are so many varieties/hybrids developed which are resistant to root knot nematode which are mentioned in table-2.

Table 2: Nematode resistant varieties of vegetable crops

Crop	Varieties
Tomato	Nema Mukt (SL 120), Punjab Chhuara (PNR 7), Hisar Lalit
Capsicum	The Hybrid -19 and BSS-89
Brinjal	ARBH-786, Navkiran, BSS-790, Kanhiya, Hybrid-183, Sandhaya and PB-324

Future thrust

In the future, one of the main thrusts should be directed towards the development of tomato rootstocks containing heat-stable genes which will be instrumental in controlling Meloidogyne spp. in tomatoes cultivated in high-temperature soils. Additionally, there is a need for international collaboration and focus on the development of Capsicum varieties with high colour but low or no pungency. This can be achieved through extensive research on germplasm, genetics and breeding techniques. Furthermore, efforts should be made to identify specific areas and seasons that allow for efficient screening of large populations under natural epiphytotic conditions. For instance, screening against tomato leaf curl disease can be particularly effective during the autumn season (August planting) in the North Indian plains as this period experiences high pressure from viruliferous whiteflies. By prioritizing these research directions and collaborative efforts, we can make significant strides in addressing challenges related to nematode control and enhancing the quality of vegetable crops like tomato and Capsicum.

Conclusion

In conclusion, the potential of protected house-grown crops such as tomatoes and Capsicum has been demonstrated in terms of better performance and higher fruit yields compared to open field conditions. To further enhance the genetic potential of these crops, gene pyramiding with marker technology can be effectively integrated into existing plant breeding programs. This approach allows researchers to access, transfer and combine desirable genes leading to the development of high-yielding and disease-resistant varieties. By adopting these advanced breeding techniques and protected cultivation methods vegetables with superior productivity and uniform quality can be produced. These high-quality produce can not only meet domestic demands but also have the potential to be exported to global markets enabling farmers to fetch higher returns on their investments. Embracing innovative breeding practices and protected cultivation technologies is crucial for sustaining agricultural productivity, meeting market demands and ensuring food security in an ever-changing world.

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