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Review Article

A REVIEW ON ADVANCES IN BELL PEPPER

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Abstract- Bell pepper (*Capsicum annuum* L) is a high value solanaceous vegetable grown extensively in India. Germplasm collection and conservation is the major step in improvement of sweet pepper which can further be used in hybridization programme. The goal of increasing productivity in the quickest possible time can be achieved by utilizing heterosis breeding. Male sterility is one of the most important traits used in hybrid pepper breeding. Application of male sterility reduces hybrid production costs by excluding the need for manual emasculation of maternal line and elimination of impurities of the seed material originated from self-pollination. Great loss in yield is caused by soil borne diseases under continuous cropping system. Chemical control is expensive, not always effective and is harmful to the environment. Losses in production due to different environmental conditions can be minimized by grafting. Grafting allows rapid response to new pathogen races and in the short-term provides a less expensive and more flexible solution for controlling soil borne diseases rather than breeding new resistant cultivars in bell pepper. The future outlook of bell pepper is very bright and has a tremendous potential.

Keywords- Bell pepper, emasculation, pollination hybrid, male sterility, grafting.

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Introduction

Bell pepper (*Capsicum annuum* L) is a high value solanaceous vegetable crop grown extensively in central and south America, Peru, Bolivia, Costa Rica, Mexico, in almost all the European countries, Honkong and India. It originated in New World tropics and subtropics and was introduced in India by the British in 19th century in Shimla, Himachal Pradesh and Nilgiri hills of Tamil Nadu [1]. In India, it is cultivated over an area of about 46,000 ha with annual production of 288,000 MT [2]. This crop is extensively cultivated in hills of Himachal Pradesh, Uttar Pradesh, Jammu and Kashmir, Andhra Pradesh and Nilgiris during summer months and as an autumn crop in Karnataka, Maharashtra, Tamil Nadu, Bihar, West Bengal and Madhya Pradesh.In Himachal Pradesh, it is grown as an off-season crop during the summer and rainy seasons and is economically important to small and marginal farmers. It is used either raw as salad, cooked as vegetable, pickled or processed and is appreciated worldwide for its flavour, aroma and colour. Germplasm collection and conservation is the major step in improvement of sweet pepper and this material can be used in hybridization programme. Quality specifications such as size, shape, colour and non-pungency make the task of developing new hybrids/cultivars very challenging. The goal of increasing productivity in the quickest possible time can be achieved by utilizing heterosis breeding. An F_1 hybrid variety is the result of a cross between two homozygous (but genetically distinct) pure lines. The prerequisite is that all the F1 plants should resemble each other phenotypically. The great success of hybrid cultivars is attributed to:

- Sufficiently large flowers, easy emasculation and pollen in abundance.
- Heterosis for yield.
- Large number of crossed seeds/fruit.
- Easy deployment of dominant genes conferring resistance to diseases.
- Highly remunerative price of hybrid seed.

Advances in Hybrid Seed Production:

- **Hybrid Seed Production:**

The F_1 hybrids of sweet pepper have been commercially exploited in India

- Steps involved in production of hybrid seed production are:
    I. Hand emasculation and pollination

A. Emasculation

Emasculation is done in the afternoon hours, a day prior to anthesis, because in some genotypes dehiscence of anthers takes place before anthesis. Emasculation is done with the forceps by gently parting the corolla and picking of anthers. Emasculated flowers are occasionally visited by honey bees and natural cross production ranges from 35 – 38 percent. So, the emasculated flowers are either bagged or protected by thin cotton wad.

B. Collection of Pollen

Fresh powdery pollen grains may be collected on the day of anthesis either by vibrator or by tapping the plucked flowers over a glass container. The pollens could be stored for a period of 1-2 months in desiccators at 0˚C temperature.

C. Pollination

Pollination is done in the afternoon hours, upto 10:00 am, normally on the day of anthesis. Though stigma remains receptive up to 2 days after anthesis, yet pollination should be done on the day of anthesis to achieve maximum fruit set. Stigma also becomes receptive a day prior to anthesis. Fruit sets in flower bud, pollinated a day prior to anthesis, has been found to be satisfactory.
In this bud pollination method, emasculation and simultaneous pollination both are done in the morning hours. Flowering and fruit set requires bright sunlight, hence hybridization should be avoided in cloudy days. Spraying of 10 ppm NAA at

flowering and seven days after, checks flower drop and increases fruit set by hybridization.

II. Use of Male Sterility

Male sterility in crop species has been classified in to two major groups, *viz*., genetic (spontaneous or induced) and non-genetic (induced) male sterility [3]. Based on the location of gene (s) controlling genetic male sterility male sterility systems can be classified as (i) Genic male sterility (gms; more precisely nuclear male sterility) and (ii) Cytoplasmic male sterility (cms; more precisely cytoplasmic-nuclear male sterility). Both Genic and Cytoplasmic male sterility have been reported in sweet pepper. Functional male sterility can also be successfully be utilized in hybrid seed production. Male sterility is one of the most important traits used in hybrid pepper breeding. Application of male sterility reduces hybrid production costs by excluding the need for manual emasculation of maternal line and elimination of impurities of the seed material originated from self-pollination.[4]identified male sterile plants in bell-pepper which had normal stigma, smaller flower parts with small, dry and shriveled anthers, as compared to the maintainer line. Cytoplasmic male sterility (CMS) in plants, which fails to produce functional pollen, is a maternally inherited trait. Specific nuclear genes that suppress CMS, termed fertility restorer (Rf) genes, have been identified in several plants. In this study, Rf-linked molecular markers in pepper (*Capsicum annuum* L.) were detected by bulked segregant analysis of eight amplified fragment length polymorphisms (AFLPs). Only AFLP8 was successfully converted to a cleaved amplified polymorphic sequence (CAPS) marker. For the fast and reliable detection of restorer lines during F_1 seed production in bell pepper, markers will prove to be very useful in future. [5].

Exploitation of male sterility

The practical importance of the CMS system in breeding is highly dependent on the presence of a restorer of fertility (Rf) gene and stable response in various environments. CMS pepper lines frequently display unstable, sterility under low temperatures. Commercial use of CMS requires high stable fertility restoration to assure high F1 hybrid seed production. [6] could develop two hybrids in bell-pepper by using CGMS system. In China, two hybrids, ‘Jiyan No. 4’, in sweet pepper [7] and ‘ Biyu’, in hot pepper[8] have been developed by using GMS and CMS lines, respectively.

[9] had also observed that sterility and fertility could be modified to a large extent by environment fluctuations, especially temperature. [10] selected ‘Sweet Pepper Parental Line Nou-1' from a cross between a male sterile plant of 'Tokyo Piman' and '101(2.4)-11-1-4' (a seed parent line of 'Beruhomare'). The cross showed genic male sterility, resistance to Phytophthora blight and to MV. It is suggested that genic male sterility is controlled by a recessive gene and the resistance to Phytophthora blight is controlled by a few incompletely dominant genes. Since genic male sterility is able to improve in F1 seed production systems, 'Sweet Pepper Parental Line Nou-1' is expected to be used as a seed parent for F1 cultivars of sweet peppers. Genetic and cytoplasmic male sterility are used on a limited scale for hybrid seed production in bell peppe [11]. Jongshu No. 5, a new sweet pepper F1 hybrid was developed by crossing CMS line with the restorer line [12]. Jiyan 8, a hybrid pepper was developed by crossing genetic male sterile line with the ox-horn pepper inbred line [13]. (Nongda-082, a sweet hot pepper F1 hybrid was developed by crossing a male sterile line with a restorer line which was slightly pungent [14]. Similarly, Jingla No.2, a hot pepper F1 hybrid was bred by crossing cytoplasmic male sterile lines 181A with the restorer line 98199 [15].

A study was conducted by [16] for conversion of the genic male sterility (GMS) system of bell pepper (*Capsicum annuum* L.) to cytoplasmic male sterility (CMS). Non-pungent bell pepper (*Capsicum annuum* L.) lacks the cytoplasmic male sterility (CMS) nuclear restores allele, R*f,* and CMS cannot be employed in its F_1 hybrid seed production. To demonstrate that the genic male sterility (GMS) system in non-pungent bell pepper can be converted to the CMS, male sterility system, the conversion of GMS to CMS for non-pungent bell pepper line GC3 was conducted by introgression of S-type cytoplasm and the R*f* allele from tropical pungent donar. After evaluating morphological traits two lines from BC_1F_1 containing S-type cytoplasm and four lines from BC_2F_2 containing R*f* allele, phenotypically similar to GC3, were obtained and could be employed as CMS male sterile line and restorer line for non pungent bell pepper. This is the first time that GMS has been successfully converted to CMS in bell pepper, a significant contribution for bell pepper hybrid seed production.

Bell Boy and No. 12 were two F_1 hybrids developed by using GMS in Bell Pepper.

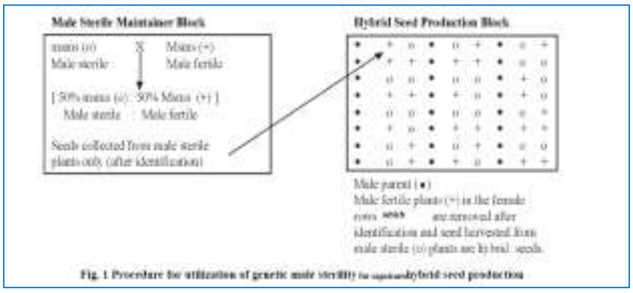

Fig-1 Reported male sterility genes in *Capsicum*

Nutraceutical Breeding Approaches in Bell Pepper

Nutraceutical is a term consist of two words:

1. Nutrition
2. Pharmaceutical

People are now more concern with the quality attributes and demand varieties which are rich in nutrients and have health benefits also so neutraceutical breeding is of great importance. Nutraceutical is any substance that may be considered as food or part of food and provides medicinal or health benefits, encompassing, prevention and treatment of diseases. The inheritance of the character “high beta-carotene content” was studied. A fertile series of lines was developed by means of reciprocal selection with orange-red pericarp colour, differing in their plasticity, fruit shape, length of vegetation period, tolerance to *Verticillium* wilt. The best fertile lines with complex economic-valuable characters were pyramided by genes of sterility and high beta-carotene content. The new hybrid cultivar ‘Vitamin’ was developed [17]. A New Sweet Pepper Variety, ‘Kyoto Manganji No. 2’, with Non-pungent Fruit was developed in Kyoto in a study by [18] In this study, a molecular marker (SCY-800) linked to single recessive gene locus controlling pungency was developed. A continuous backcross of [(a bell pepper variety ‘Tongari’ × ‘Kyoto Manganji No. 1’) × ‘Kyoto Manganji No. 1’] was carried out with the help of marker-assisted selection in order to develop a novel non-pungent variety, ‘Kyoto Manganji No. 2’, by transferring the recessive gene to the original variety ‘Manganji’. A study was also conducted by [19] at Vegetable Research Block of GB Pant, Uttrakhand to gather information about the potential and characteristics of the experimental material of bell pepper. A wide range of heterosis over better parent and standard check was observed in F_1 generation for most of the studied traits. The F_1 crosses California Wonder x SSP and PRC-1 x SSP exhibited highest significant heterosis for days to first harvest in desirable direction each over standard check. The highest economic heterosis was recorded in cross combination PRC-1 x Rani Sel-1 to the tune of 170.30% over standard check followed by California Wonder x SSP (140.64%) and Rani Sel-1 x Sel-12-2-1 (108.62%) respectively.

Breeding for Biotic and Abiotic Stress Resistance:

Need for resistance breeding:

Heavy application of pesticides increases the cost of cultivation as well as residual toxicity of the chemicals in the produce causes health hazards upon consumption.

Excess use of pesticides causes environmental pollution.

The crop suffers from many diseases like *Phytophthora* leaf blight and fruit rot, leaf spot, powdery mildew, wilt, virus complex.

A major area of our country is covered by different stresses such as moisture, temperature, both high and low and salinity. (Peppers are moderately sensisitive to saline condition – Bernstein, 1954)

Stress cause reduction in growth of plants, early senescence and ultimately reduces the yield resulting from low photosynthetic rate and rise in respiration.

It is imperative to concentrate on the development of cultivars that are genetically resistant to biotic and abiotic stress.
A study was also conducted by [20] at Experimental Research Farm of Department of Vegetable Science at Dr Yashwant Singh Parmar University of Horticulture and Forestry for screening of 15 genotypes of bell pepper against *Phytophthora* leaf blight and fruit rot disease under natural epiphytic conditions and revealed, genotype Feroz and LC-1 as moderately resistant to the disease reactions with lowest apparent infection rate and Kannaul Collection as highly susceptible to the disease reaction with highest apparent rate of infection.
A study was conducted [21] at Faisalabad Pakistan for the assessment of salinity tolerance in bell pepper genotypes on the basis of germination, emergence and growth attributes and the results revealed, genotype Zard as the most tolerant one while the genotype PEP-311 as the most sensitive.

Table-1 *Source of resistance in Bell pepper*

Species	Disease resistance
C. chinense	Root rot, TMV, potato virus X, spotted wilt virus, poty virus, aphids, thrips, drought
C. chacoence	Bacterial leaf spot, anthracnose, TMV, poty virus, aphids, thrips
C. baccatum var. pendulum	Powdery mildew, CMV, potato virus Y, drought, high temperature
C. ciliaum	Root rot, water logging
C. frutescens	Fruit rot, TMV,CMV, tobacco leaf virus, poty virus
C. eximium	Potato virus Y, tobacco leaf curl virus, drought, high temperature
C. flexuosum	CMV, potato virus X, soil salinity
C. cardenasii	Aphids, thrips, low temperature

Use of grafting to develop biotic stress resistance in bell pepper

Though grafting has been practiced in fruit trees for thousands of years, vegetable grafting has been only recently widely adapted on a commercial scale. Grafting was introduced to Western countries in the early 1990s and is currently being globally practiced using local scion cultivars and introduced rootstocks. Sweet pepper is affected by various biotic (diseases, nematodes) and abiotic stresses (cold, heat, water stress, salinity). Great loss in yield is caused by soil borne diseases under continuous cropping system. Chemical control is expensive, not always effective and is harmful to the environment. Losses in production due to different environmental conditions can be minimized by grafting. Grafting allows rapid response to new pathogen races and in the short-term provides a less expensive and more flexible solution for controlling soil borne diseases than breeding new resistant cultivars. Grafting is a propagation method where the tissues of two plants are fused together. The bottom part of the plant that contributes roots and support is called the rootstock. It involves the joining together of plant parts by means of tissue regeneration which achieves the union and grows as an independent plant. In India, grafting work has been started in IIHR Bangalore, NBPGR regional station, Thrissur, Kerala and CSKHPKV, Palampur. IIHR, Bangalore, has identified rootstocks for waterlogged conditions and imported semi-automated grafting machine for grafting.

Variety selection

Wise selection of rootstock based on fruit quality is very important. Rootstock is selected based on resistance i.e. resistance to various soil borne diseases, root knot nematode and tolerance to salt, cold and water logging conditions. In some cases, rootstock may be selected on the basis of vigorous root system which results in increased vigour of the plant. Scion is selected on the basis of fruit quality i.e. for high TSS, flavour, antioxidants and ascorbic acid content etc. Rootstock and scion varieties must be genetically compatible. Healthy, uniform rootstock seedlings with 2–4 true leaves and a minimum stem diameter of about 1.5 mm to 2.5 mm are best for grafting. Choose rootstock and scion seedlings whose stem diameters are similar. Mainly cleft, tongue, tube, splice and pin grafting is done in sweet pepper.

Types of grafting:

Cleft Grafting

In Cleft grafting, cut a 0.5 cm long vertical incision into the centre of the rootstock.

Tongue approach grafting/ Side grafting

The grafting cut for rootstock should be made in a downward direction and the scion cut in an upward direction at an angle, usually 300-400 to the perpendicular axis and deep enough to allow the fusion of as many vascular bundles as possible and then specially designed clips are placed to fix the graft position. After five days, the rootstock top and the scion roots from the
grafted plants are removed.

Tube grafting

This is the most widely used grafting technique for tomatoes and sweet pepper. The rootstock should be grafted when cotyledons and the first true leaf start to develop (about 7 to 10 days after sowing). One cotyledon and the growing tip are removed

Pin grafting

In pin grafting specially, designed pins are used to hold the grafted position in place. The ceramic pin developed by Takii Seed Company is about 15 mm long and 0.5 mm in diagonal width of the hexagonal cross-section. The pins are made of natural ceramic so it can be left on the plant without any problem. The price of ceramic pin is fairly high so the alternative methods are being sought like bamboo pins, rectangular in cross-sectional shape, could successfully replace the expensive ceramic pins at much lower price.

Securing graft union

It is done by using glue and clips mainly.

Robotic Grafting

Grafting machines have been developed and commercialized in Korea. The pin grafting robot developed by Rural Development Administration, Korea for solanaceous crops can graft 1200 seedlings per hour. The simple and economic grafting machine was developed and has been very popular in Korea. This machine priced about US $500, has been exported for more than 10 years to many Asian countries and some European countries. This machine can graft up to 600 seedlings per hour by tongue approach grafting. However, an experienced operator is needed to run this machine effectively and efficiently. This machine has also been actively exported to many foreign countries in recent years because of the reasonable price, multiple functions and convenient handling. More recently, a fully automated grafting robot (1000 grafts per hour) has been developed and used commercially for tomato in Netherlands.

Healing and acclimatizing the graft

The healing and acclimatization are very important for grafted plants to survive. A healing chamber is commonly placed in a greenhouse. Both the width and length of the healing chamber should be 5 feet or less to provide easy management when opening and closing and to maintain high humidity throughout the chamber. In healing chambers, the grafts are kept for 5 – 7 days. The grafted plants take 7-10 days for acclimatization. The plants must be re-acclimated for 3-4 days to the full sun conditions of the greenhouse environment. Start increasing the light exposure by removing the opaque plastic sheets. Finally, while planting the grafted plants into the production house, it is important to keep the graft union above the soil line, because if the scion roots into the soil, the plant will become susceptible to soil-borne diseases.

Field management of grafts

Planting is done on the raised beds covered with polythene sheet in order to minimize the transmission of disease through rain splashes. Raised beds are highly recommended to minimize flooding. While transplanting care should be taken that graft union remain 2.5 cm above the soil line. Timely removal of shoots developed from the rootstock and adventitious roots from scion is very important after transplanting in the field. Grafted plants should be staked three to four weeks after transplanting in tomato. This will prevent the scion stem contacting the soil. Proper water management practices are very important after transplanting to minimize the wilting of the grafted plants.

Germplasm Sources:

Southern Plant Introduction Station, Griffin, Georgia, USA-approximately 3000 accessions, evaluation data entered in the Germplasm Resources Information Network (GRIN), a centralized computer database system.

- World Vegetable Centre (WVC), Taiwan-about 7500 base/active collection.
- Centre for Genetic Resources, Wageningen, the Netherlands.
- Centre Institute of Genetics and Germplasm, Gatersleben, Germany.
- National Bureau of Plant Genetic Resources, New Delhi, India.
- Indian Institute of Vegetable Research, Varanasi, India.

Conclusion:

Exploitation of heterosis in vegetable crops has attracted attention of vegetable breeders to the vegetable requirements of the increasing population.

- There are alternative systems to produce hybrid seed, but the production of bell pepper hybrid seed commercially still relies on making crosses between two parents by conventional hand pollination, a very labour intensive and expensive process.
- Male-sterility in peppers (*Capsicum annuum* L.) was first documented in the 1950's. Since then considerable knowledge has been accumulated on the nature of the trait, the means of its identification and induction, inheritance of both genic and cytoplasmic genic male-sterility, its line maintenance, and the potential for breeding hybrid cultivars. Today, several internationally known seed companies use the genic mechanism msms on a large scale for producing hybrids, whereas the cytoplasmic genic source is used mainly for breeding pungent (S) Rfrf hybrids.
- Hybrid seed technology has the tremendous potential today. The future outlook for bell pepper is good and should be exploited commercially for the benefit of the farmers

Future strategies:

- Heterosis should be exploited commercially in most of the vegetables.
- Multiple disease resistance should be developed in hybrids.
- Hybrids of export quality should be developed.
- Emphasis on male sterility should be given.
- MS with linkage marker should be used.
- Standardization of research on molecular level.
- Public sector research organizations to develop parental lines with authenticated resistance and genes

Application of review: The review will be helpful in understanding the recent advances in the bell pepper.

Review Category: Breeding of Bell pepper

Abbreviations:

NHB- National Horticulture Board

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