

## Development of Haploid Production in Fruit Crops

Chandan kumar Rout, Bhagyalaxmi Pahi and Anubhav Biswal

MSc Scholar, School of Agriculture, Lovely Professional University, Phagwara, Punjab

**\*Corresponding Author:** Chandan kumar Rout, MSc Scholar, School of Agriculture, Lovely Professional University, Phagwara, Punjab

### ABSTRACT

Homozygosity at a specific locus is consistently attractive component for crop improvement programs. The production haploids have a remarkable performance for the genetic improvement of fruit trees while breeding takes a long time through parthenogenesis and takes more times to produce a new variety plant. Because of this constraint it is just doable in annuals. As fruit plants are not annuals, they develop in vegetative stage for long term before blossoming to come. Hence, to accomplish homozygosity isn't possible in breeding method. In vitro condition by using androgenesis or gynogenesis, haploid plants can be easily produced.

**Keywords:** Haploid, Homozygosity, Androgenesis, Gynogenesis

### INTRODUCTION

The term haploid refers to which plants have a gametophytic number of chromosomes. Naturally, haploid plants are produced through the process of apomixes and parthenogenesis, but the frequency shallow (0.001 to 0.01%). In Datura plant first observation of the natural haploids and developed through the parthenogenesis. The artificial occurrence of androgenic haploid was first reported in Nicotiana, Crepis, Hordeum, and other species. The first report of induced haploidy through anther culture has been made by Guha and Maheshwari (1964) in Datura species.

With the help of tissue culture haploid plant can be produced through microspore culture, pollen culture, or anther culture i.e. male gamete (e.g. spore) is used so called as androgenesis while such uses of female gametes e.g. ovule is called as gynogenesis.

The production haploids have a remarkable performance for the genetic improvement of fruit trees while breeding takes a long time through parthenogenesis and also extremely heterozygous nature in fruit trees Rajasekaran and Mullins (1979). This practice can be significant with Annona species such as A. Haploid plants produced from A. squamosa by using anther culture on N-6 medium Nair et al., (1983).

Researchers produced haploid embryos by using pseudogamy, wide hybridization and Parthenogenesis under in vivo condition. The

haploid embryo must rescue through embryo rescue techniques and cultured further to produce haploid and subsequently chromosome doubling was done to get doubled haploids. For haploid production androgenesis (anther and microspore culture) and gynogenesis (ovary and ovule) were used, ideally androgenesis is used.

### IN-VIVO METHODS OF HAPLOID PRODUCTION

#### Development of Spontaneous Haploids-

In more than hundred species the unconstrained haploid was appeared however in organic product crop the recuperation rate for haploid creation is low, Zhang *et al.*, (1990). In trees unconstrained and low feasible haploid plants got in apple, pear, peach, plum, apricot, and so on., yet with low recurrence and are not for all intents and purposes pertinent. The creation of unconstrained haploids might be because of parthenogenesis or apogamy.

#### Distant Hybridization

Hybrids can be delivered by disposal of one of the parental genomes, as results of hybridization.

Haploid plants were produced in citrus by cross between diploids and triploids species.

#### Irradiation Effects

In this technique UV ray might be utilized to prompt chromosomal breakage and their resulting end from pollens and utilizing the pollens for preparation to deliver haploids in Citrus, Pear, Apple and so on.

## Development of Haploid Production in Fruit Crops

### Chemical Treatment

Some chemicals like colchicine, nitrous oxide, maleic hydrazide used to eliminate the somatic cells which might result in haploid productions.

### Chromosome Elimination

In this cycle haploid got by specific chromosome end that follows certain inter-specific fertilizations.

### Parthenogenesis

Haploid recovery through unpollinated female gametophytes is typically portrayed as haploid parthenogenesis. The strategy is for all intents and purposes not practical except if explicit markers utilized for choice.

## IN VITRO TECHNIQUES FOR HAPLOID PRODUCTION

### Androgenesis

Haploid culture is the most popular method used to produce Haploid plant through male gametes. It became said that haploid and doubled haploid was efficiently practiced more than two hundred species most of them are belongs to annuals. The fulfilment rate is greater in solanaceae, gramineae as comparison to leguminosae and perennial woody crops. [Wenzel *et al.*, (1995), Bajaj (1990), Sangwan-Norrel *et al.*, (1986), Dunwell (1986), Raghavan (1990)].

### Anther Culture

Examination on haploid production has been completed on various fruit trees through anther culture, Ochatt and Zhang (1996). flower bud was collected from fruit plant at a particular state of pollen grains development. After that the floral buds were sterilized with 70 % (v/v) ethyl alcohol then followed by inundation in sodium NaOCl (about 1.5% dynamic chlorine in water) containing a couple of drops of Tween 20, and lastly washed 3 times for five minutes with sterile refined water. Petals were removed carefully with little forceps, and anthers are deliberately dismembered and put into the culture media. Then culture medium placed in a culture room with maintaining 12-18 hours light and 6-12 hours dark at 28 °C. After some day's anther produced callus, then callus later produced an embryo and finally mass development of haploid plant.

### Microsporeculture

Production of haploid plants can be possible by using male gametophytic cells (microspores). Flower bud were collected from fruit plant then pollens should be separated from anthers by

crushing with glass rod. Then remove the anther waste by filtering the pollen suspension, generally this filtering is done because smaller size pollen did not germinate only large pollens were considered for culturing, after that pollen were washed and assembled. Now these pollens were ready for culturing, then these pollens were cultured in liquid medium. After some days pollen produced callus first later produced an embryo and finally mass development of haploid plant.

The couple of reports about this technique in fruit crops are with respect to citrus, Germana *et al.* (1996), olive Bueno *et al.* (2004), and apple Hofer (2004). Exploration is in progress on microspore culture of a few genotypes: cherry, loquat, pear, olive. Germana *et al.* (1996).

### Gynogenesis

Unfertilized egg cells are used for gynogenesis. Haploid plant life can be developed from ovary or ovule cultures. It is possible to trigger megaspores of angiosperms to develop into a sporophyte. The flowers so produced are known as gynogenic haploids. Gynogenic haploids have been first advanced by means of San Noem (1976) from the ovary cultures of *Hordeum vulgare*. This method changed into later carried out for raising haploid plants of rice, wheat, maize, sunflower, sugar beet and tobacco. Haploid induction through gynogenesis is popular in onion and sugarbeet but for other crops it is not preferred due to its low efficiency Forster *et al.* (2007).

## APPLICATION OF HAPLOID PRODUCTION IN FRUIT CROPS

First haploid plant was successfully developed in kiwi from Gynogenesis Fraser *et al.*, (1991). At the point when mortally illuminated pollen dust from flower and capricious male were utilized as pollinator, the offspring got decreased ploidy level in numerous genotypes Pandey *et al.*, (1990). Dust germination was influenced; number of reasonable seeds were diminished because of low natural product set and helpless natural product development. It was demonstrated that parent genotype impacts the amount and nature of seedling and haploids (Chalak and Legave, 1997). Pandey *et al.* (1990) and Fraser *et al.* (1991) had the optioned to develop haploid plants but they failed to deliver fruitful dihaploids.

Papaya is polygamous in nature however here likewise anther culture was fruitful. Haploid plantlets were created under invitro condition

through anther culture [Litz and Conover (1978), Tsay and Sue (1985)].

Anther culture was most successful and popular in citrus to produce haploid plants in various cultivars. Germana *et al.* (1996) studied on microspore culture on various species of citrus like orange, sour orange, lemon etc. The results showed that small proembryos, and Multi-nucleated structures were observed, without obtaining embryos and plantlets. Haploid plants were first developed in citrus by using gamma rays by Karasawa (1971).

The scientists studied on 9 varieties for anther culture, to see the response of those cultivars and they saw out of these cultivars 4 cultivars gave positive results. The cell changes advanced in the in vitro refined anthers had been portrayed by using magnifying lens investigation and showed the existence of pollen dust determined multi-cell formation that demonstrate re-programming of pollen Germanà *et al.*, (2006).

The researcher found no plantlet were produced, when they used un-fertilized ovule and ovary to induce plantlet in vitro condition, Zhang and Lespinasse (1988). Portion of gamma ray didn't influence the dust feasibility and in vitro dust germination yet it influences fruit set and number of reasonable seeds. The quantity of suitable seeds, void seed and seed with just endosperm were rely upon female genotype [Zhang *et al.* (1992), De Witte and Keulemans (1994), Zhang *et al.* (1987), James *et al.* (1985), Verdoodt *et al.* (1998) and Nicoll *et al.* (1987)]. Many scientists studied on anther culture of apple, they successfully produced plantlets through androgenesis but the induction rate was very low. [Xue and Niu (1984), Fei and Xue, (1981)].

Homozygous plants can't be produced through selfing in Mulberry, because of dioecious in nature. It was difficult to produce haploid plant through anther culture [Jain *et al.* (1996), Sethi *et al.* (1992)] but when researcher used unpollinated ovaries, they observed haploid plantlets were produced in Mulberry through gynogenesis. Dennis *et al.* (1999).

In 1996 Kerbellec was successfully developed haploid plant in *Musa acuminata* by using anther culture. Then later in *Musa balbisiana* banana 41 haploid plantlets were developed from anther culture. Assani *et al.* (2003). Bueno *et al.* (2004) studied effect of microspore culture on in two cultivars of olive i.e. Arbequina and Picual. They found that multi-nucleate microspores development with division of the microspore.

The researchers used anther of sweet cherry and successfully developed Haploid callus but failed to produce plantlets [Höfer and Hanke (1990), Seirlis *et al.* (1979)]. like sweet cherry some researcher practiced anther culture in *Prunus persica*, also they got no plantlet through androgenesis only observed production of callus, Hammerschlag (1983). Though many researchers produced haploid plants by using androgenesis Pratassenja 1939). Kadota *et al.* (2002) studied on anther culture in pear, but they were failed to develop plantlet but they produced 2 embryos through androgenesis.

Many researchers developed haploid callus by using the anther of grapes and anther culture is most popular method in grapes from long ago, [Kim and Peak, (1981), Gresshoff and Doy, (1974), Cersosimo (1986)].

Many scientists studied on culturing anther in grape and successfully developed haploid plantlets [Bouquet *et al.* (1982), Hirabayashi and Akihama (1982), Mauro *et al.* (1986)]. Mauro *et al.* (1986) found that 2n somatic embryoid was produced by using anther culture in grapes. Mullins and Rajasekaran (1979) studied on compositions of growing media and condition of culture; affect the anther production in grapes.

## CONCLUSIONS

Several generations of selfing is required to develop a pure line through conventional breeding method. Some constraints of fruit crops are heterozygosity in nature, **long** reproductive cycle, and sometimes some fruit crops self-incompatibility in nature, so in breeding method it is difficult to produce haploid plants. So improve the efficiency and the speed production of haploid plant, some researchers used androgenesis or gynogenesis in vitro condition and successfully haploid plant in fruit crops like citrus, kiwi, apple, pear, plum etc.

## REFERENCE

- [1] Bajaj YPS (1990) In vitro production of haploids and their use in cell genetics and plant breeding. In: Bajaj YPS (ed) Biotechnology in Agriculture and Forestry: Haploids in Crop Improvement. Berlin: Springer, Vol. 12. Part I. pp. 1–44.
- [2] Bouquet AB, Piganeau B, Lamaison AM (1982) Influence du génotype sur la production de cal, d'embryons et de plantes entières par cultures d'anthères in vitro dans le genre *Vitis*. CR Acad Sci Paris 259: 569–574.
- [3] Bueno MA, Pintos B, Prado MJ, Gomez A, Manzanera JA (2004) Androgenesis: A tool for

- woody plant breeding. *Rec Devel Genet Breed* 1: 365–383.
- [4] Cersosimo A (1986) Coltura in vitro di antere in *Vitis* sp. (I contributo). *Rivista di Viticoltura e di Enologia di Conegliano* 39(12): 520–531.
- [5] Chalak L, Legave JM (1997) Effects of pollination by irradiated pollen in Hayward kiwifruit and spontaneous doubling of induced parthenogenetic trihaploids. *Sci Hort* 68: 83–93.
- [6] De Witte K, Keulemans J (1994) Restriction of the efficiency of haploid plant production in apple cultivar „Idared“, through parthenogenesis in situ. *Euphytica* 77: 141–146.
- [7] Dennis Thomas T, Bhatnagar AK, Razdan MK, Bhojwani SS (1999) A reproducible protocol for the production of gynogenic haploids of mulberry, *Morus alba* L. *Euphytica* 110: 169–173.
- [8] Dunwell JM (1986) Pollen, ovule and embryo culture, as tools in plant breeding. In: Withers LAP, Alderson G (eds) *Plant Tissue Culture and Its Agricultural Applications*. London: Butterworths, pp. 375–404.
- [9] Fei KW, Xue GR (1981) Induction of haploid plantlets by anther culture in vitro in apple. cv. “Delicious”. *Sci Agric Sin* 4: 41–44.
- [10] Fraser LG, Harvey CF, Kent J (1991) Ploidy manipulations of kiwifruit in tissue culture. *Acta Hort* 297: 109–114.
- [11] Froelicher Y, Bassene JB, Jedidi-Neji E, Dambier D, Morillon R, Bernardini GG, Costantino G, Ollitrault P (2007) Induced parthenogenesis in mandarin for haploid production: Induction procedures and genetic analysis of plantlets. *Plant Cell Rep* 26: 937–944.
- [12] Germana MA, Scarano MT, Crescimanno FG (1996) First results on isolated microspore culture of Citrus. *Proc Inter Soc Citriculture (South Africa)*. 2: 882–885.
- [13] Gresshoff PM, Doy CH (1974) Derivation of a haploid cell line from *Vitis vinifera* and the importance of the stage of meiotic development of anthers for haploid culture of this and other genera. *Z. Pflanzenphysiol* 73: 132–141.
- [14] Guha S, Maheshwari SC (1964) In vitro production of embryos from anthers of *Datura*. *Nature* 204: 497–498.
- [15] Hammerschlag FA (1983) Factors influencing the frequency of callus formation among cultivated peach anthers. *HortScience* 18: 210–211.
- [16] Hirabayashi T, Akihama H (1982) In vitro embryogenesis and plant regeneration from the anther derived callus of *Vitis*. In: Fujiwara A (ed) *Plant Tissue Culture*. Tokyo: Maruzen, pp. 547–548.
- [17] Hofer M (2004) in vitro androgenesis in apple – improvement of the induction phase. *Plant Cell Rep*. 22: 365–370.
- [18] Höfer M, Hanke V (1990) Induction of androgenesis in vitro in apple and sweet cherry. *Acta Hort* 280: 333–336.
- [19] Jain AK, Sarkar A, Datta RK (1996) Induction of haploid callus and embryogenesis in vitro cultured anthers of Mulberry (*Morus indica*). *Plant Cell Tiss Org Cult* 44: 143–147.
- [20] James DJ, Passey AJ, Mackenzie KAD, Menhinick EC (1985) The effect of pollen irradiation on the development of the post-fertilization ovule of apple (*Malus pumila* Mill.). In: Chapman et al., (eds) *Experimental Manipulation of Ovule Tissue*. London: Longman, pp. 210–224.
- [21] Kadota M, Han DS, Niimi Y (2002) Plant regeneration from anther-derived embryos of apple and pear. *HortScience*, 37(6): 962–965.
- [22] Karasawa K (1971) On the occurrence of haploid seedlings in Citrus natsudaoides Hayata. *Bull. Sakushingakuin Junior Col Woman* 1: 1–2.
- [23] Kim SK, Peak KY (1981) Study on anther culture of grape. 1. Varietal differences in callus formation. *J Korean Soc Hort Sci* 22(2): 89–91.
- [24] Litz RE, Conover RA (1978) Recent advances in papaya tissue culture. *Proc Florida State Hort Soc* 91: 180–182.
- [25] Mauro MC, Nef G, Fallot J (1986) Stimulation of somatic embryogenesis and plant regeneration from anther culture of *Vitis vinifera* cv. Cabernet Sauvignon. *Plant Cell Rep* 5: 377–380.
- [26] Mullins MG, Rajasekaran K (1979) Embryos and plantlets from cultured anthers of hybrid grapevines. *J Exp Bot* 30: 399–407.
- [27] Nair S, Gupta PK, Mascarenhas AS (1983) Haploid plants from in vitro anther culture of *Annona squamosa* L. *Plant Cell Rep* 2: 198–200.
- [28] Nicoll MF, Chapman GP, James DJ (1987) Endosperm responses to irradiated pollen in apples. *Theor Appl Genet* 74: 508–515.
- [29] Pandey KK, Przywara L, Sanders PM (1990) Induced parthenogenesis in kiwifruit (*Actinidia deliciosa*) through the use of lethally irradiated pollen. *Euphytica* 51: 1–9.
- [30] Pratassentja GD (1939) Production of polyploid plants, haploid and triploids in *Prunus persica*. *Camp. Rend. Acad. Sci. (Doklady) URSS*, 22: 348–351
- [31] Raghavan V (1990) From microspore to embryo: Faces of the angiosperm pollen grain. In: Nijkamp HJJ, van der Plas LH, J van Hartigik (eds) *Progress in Plant Cellular and*

## Development of Haploid Production in Fruit Crops

- Molecular Biology. I.A.P.T.C., Dordrecht: Kluwer, pp. 213–221.
- [32] Sangwan-Norrel BS, Sangwan RS, Pare J (1986) Haploïdie et embryogenèse provoquée in vitro. Bull Soc Bot Fr 133, Actual Bot 4: 7–39.
- [33] Seirlis G, Mouras A, Salesses G (1979) Tentatives de culture in vitro d'anthères et de fragments d'organes chez Prunus. Ann Am Plant 29: 145–161.
- [34] Sethi M, Bose S, Kapur A, Rangaswamy NS (1992) Embryo differentiation in anther cultures of mulberry. Indian J Exp Biol 30: 146–148.
- [35] Tsay HS, Sue CY (1985) Anther culture of papaya (*Carica papaya* L.). Plant Cell Rep 4: 28–30.
- [36] Verdoodt L, Van Haute A, Goderis IJ, De Witte K, Keulemans J, Broothaerts W (1998). Use of the multi-allelic self-incompatibility gene in apple to assess homozygosity in shoots obtained through haploid induction. Theor Appl Genet 96: 294–300.
- [37] Wenzel G, Frei U, Jahoor A, Graner A, Foroughi-Wehr B (1995) Haploids – an integral part of applied and basic research. In: Terzi M et al., (eds) Current Issues in Plant Molecular and Cellular Biology. Dordrecht: Kluwer, pp. 127–135.
- [38] Xue GR, Niu JZ (1984) A study on the induction of apple pollen plants. Acta Hort Sin 11: 161–164.
- [39] Zhang YX, Boccon-Gibod J, Lesinasse Y (1987) Obtention d'embryons de pommier après culture d'anthères. Comptes rendus de l'Académie des Sciences, Serie III 305: 443–448.
- [40] Zhang YX, Bouvier L, Lespinasse Y (1992) Microspore embryogenesis induced by low gamma dose irradiation in apple. Plant Breed 108: 173–176.
- [41] Zhang YX, Lespinasse Y (1988) Culture in vitro d'ovules non fécondés et d'embryons prélevés 8 jours après pollinisation chez le pommier cultivé (*Malus x domestica* Borkh.). Agronomie 8: 837–842.
- [42] Zhang YX, Lespinasse Y, Chevreau E (1990) Induction of haploidy in fruit trees. Acta Hort 280: 293–304.

**Citation:** Chandan kumar Rout, Bhagyalaxmi Pahi and Anubhav Biswal, "Development of Haploid Production in Fruit Crops", *International Journal of Research in Agriculture and Forestry*, 7(11), 2020, pp. 01-05.

**Copyright:** ©2020 Chandan kumar Rout, Bhagyalaxmi Pahi and Anubhav Biswal. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.