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## Propagation by Sexual and Budding (Asexual) Methods in Horticulture Crops

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### ABSTRACT

Horticulture crops are propagated mainly by two methods viz. sexual (seed) and asexual (vegetative). The plants produced through former method do not produce seedlings true-to-type wherein, identical characters (true-to-type) are inherited from mother plant through asexual method of propagation. Budding is one of the important methods of asexual propagation. The plants produced through budding have uniform maturity along with quality fruit. Seedling plants are having long juvenile period and gigantic growth habit which hampers plant protection, crop management and harvesting operations. Seed propagation and budding method of asexual propagation with its type is reviewed and presented.

**Keywords:** Propagation, Budding, Types of budding, Seed propagation.

### INTRODUCTION

The types of propagation are grouped as (i) sexual or seed propagation and (ii) asexual (vegetative) propagation. Sexual reproduction (propagation) is known as amphimixis. Multiplication takes place through seed that develop after normal meiosis followed by syngamy. Sexual reproduction refers to multiplication of plants by seed. In sexual process, fusion of male and female gametes takes place to produce seed. Meiotic division take place in course of fusion and chromosome numbers as in parents are reduced to half which after fertilization becomes normal.

Therefore, the new individual formed in this way is not true-to-type e.g. phalsa, karanda, papaya etc. Budding is one of the methods of asexual propagation. Budding is the union of scion (bud) of one plant on or into rootstock of another plant in such a way that a union will be formed and partners will continue to grow e.g. ber, aonla, roses etc.

### Propagation and its type

Plant propagation is defined as multiplication of plants by both sexual and asexual methods. Horticulture crops can be propagated through seed as sexual propagation and grafting as vegetative propagation are given below.

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**Seed formation**

There are four important steps involved in seed propagation are given below.

- (i) Development of gametophytes and gametes
- (ii) Pollination
- (iii) Fertilization (syngamy)
- (iv) Post fertilization development (formation of seed and fruit)

**Development of gametophytes**

A normal flower consists of calyx, corolla, androecium and gynoecium. The last two parts of flowers have great significance in entire process of reproduction. When, androecium and gynoecium both are present in same flower is known as hermaphrodite or bisexual flower e.g. ber, citrus, guava, sapota etc. When, male (androecium) and female (gynoecium) parts of flower are present in separate flower but on same plant known as monoecious condition e.g. aonla, walnut, hazelnut, jackfruit etc. wherein, andromonoecious condition is found in mango, cashewnut in which male and hermaphrodite flower are present in same panicle. Further, the gynodioecious condition is found in some cultivars of papaya e.g. Pusa Delicious, Pusa Majesty, CO3, Coorg Honey Dew (Madhubindu), Sunrise Solo, Taiwan, Surya etc. Wherein, in dioecious condition male and female flowers are found on different plants, separately e.g. pointed gourd, ivy gourd, spine gourd, date palm and Pusa Giant, Pusa Dwarf, Pusa Nanha, CO-1, CO-2, CO-5, CO-6 etc. cultivars of papaya.

**Microsporogenesis and microgametogenesis**

Production of microspore from pollen mother cell (PMC) is termed as microsporogenesis. Generally, each anther has four pollen sacs which contain numerous pollen mother cells (PMCs). After meiosis, each PMC results into four haploid cells or microspores. After thickening of the cell wall of microspore, it is termed as the pollen grain.

On other hand microgametogenesis refers to the production of male gametes. During maturation of pollen, the microspores nucleus divides mitotically to produce a generative and a vegetative or tube nucleus.

The pollen is generally released in this binucleate stage. When the pollen reaches the stigma of flower it is termed as pollination. Shortly after pollination, pollen grains start germination. The pollen tube enters into stigma and grows through the style. The generative nucleus now undergoes a mitotic division to produce two male sperms. The pollen alongwith pollen tube is known as microgametophyte. The process between microspore to microgametophyte is termed as microgametogenesis. Pollen tube finely enters into ovule through a small pore, micropyle and discharges the two sperms into the embryo sac (Shukla et al., 2004).

**Megasporogenesis and megagametogenesis**

Megasporogenesis can be defined as production of megaspore from megaspore mother cell (MMCs). Megasporogenesis occurs in ovule which is present inside the ovary. A single cell in each ovule differentiates into megaspore mother cell. The megaspore mother cell undergoes meiosis to produce four haploid megaspores. Three of them degenerate leaving one functional megaspore per ovule.

Under the process of megagametogenesis, the nucleus of a functional megaspore divides mitotically to produce four or more nuclei. The exact number of nuclei and their arrangements varies considerably from one species to another species. In most of the cases megaspore nucleus undergoes three mitotic divisions to produce eight nuclei. Three of these nuclei move to one pole and produce central egg cell and two synergid cell. One synergid is situated on either side of the egg cell. Another three nuclei migrate to the opposite pole to give rise to an antipodal cell. Remaining two nuclei in the center are known as polar nuclei and after fusion forms the secondary nucleus. Megaspore develops into a mature megagametophyte or embryo sac and the development of embryo sac from a megaspore is known as megagametogenesis.

**Fertilization (syngamy)**

Fusion of the one of the two sperms with egg cell, producing a diploid zygote is known as

fertilization or syngamy. While, fusion of remaining sperms with the secondary nucleus leads to the formation of a triploid primary endosperm nucleus is termed as triple fusion. When, triple fusion and syngamy takes place simultaneously it is termed as double fertilization.

#### **Post fertilization development**

A series of changes in the ovule follows as the fertilization is over. Synergids and antipodal cells become disorganized, and the egg cell becomes covered with a cell wall, known as oospore. This follows the development of seed. The various part of gynoecium develop into ovary-fruit, ovule-seed, integument outer testa (outer seed coat), integument inner tegmen (inner seed coat), secondary nucleus after fertilization-endosperm (triploid), egg cell (after fertilization) embryo, nucellus-perisperm (nutritive tissue like endosperm).

#### **Asexual propagation**

It does not involve the gametes from parents. It takes place due to mitosis division. Mitosis division continues in shoot tip, root tip and cambium. When, some portion of plants is wounded, mitosis division takes place. Under mitosis division chromosomes divide longitudinally to form two daughter cells. This forms the basis of asexual propagation. It is simply a vegetative or somatic extension of one parent and there is no chance of inheriting a mixture of characters. The plants raised through asexual process are identical to mother plants.

#### **Modes of reproduction in relation to fruit breeding**

The reproduction system is principally responsible for perpetuation and preservation of particular genotype. Further, the mode of reproduction also determines the genetic constitution of fruit crop whether it is homozygous or heterozygous.

#### **Asexual propagation**

- i. By using apomictic seeds. e.g. citrus, mango and apple
- ii. By using vegetative parts of the plant

#### **By using apomictic seeds (Apomixis)**

Apomixis refers to the development of embryo without fertilization. The plants developed

from apomictic seeds are true to type. Obligate apomictic seeds developed with or without pollination but without fertilization e.g. *Malus* sp. Wherein, in case of facultative apomictic seeds both type of embryo may develop i.e. zygotic as well as nucellar embryo e.g. citrus and polyembryonic cultivars of mango (Shukla et al., 2004).

#### **Classification of apomixis**

##### **Recurrent apomixis**

The embryo sac (female gametophyte) develops from the megaspore mother cell where meiosis is disturbed (sporogenesis failed) or from adjoining cell (megaspore mother cell disintegrates). The egg cell is diploid and embryo develops directly from the diploid egg cell without fertilization. Generally, somatic apospory, diploid parthenogenesis and diploid apogamy falls under recurrent apomixis e.g. raspberry (*Rubus* sp.), *Malus hupehensis*, *M. sikkimensis*, *M. sergeant* and *M. toringoides* (Gupta, 2014).

##### **Non recurrent apomixis**

The development of embryo takes place from haploid egg cell without fertilization. Such type of apomixis rarely occurs. Generative apospory, haploid parthenogenesis, haploid apogamy and androgamy fall under non-recurrent apomixes e.g. mango, citrus, jamun.

##### **Adventive embryony / nucellar or polyembryony**

In this case more than one embryo develops in a single seed. In the seed both types of embryo develops i.e. nucellar embryo from nucellar cell and zygotic embryo from egg cell with the result of syngamy. e.g. Olour, Goa, Kurukkan, Bappakai, Vellaikolamban, Chandrakaran cultivars of mango and most of the species of citrus except citron (*C. medica*), pummelo (*C. grandis*) and tahiti lime (*C. latifolia*) (Shukla et al., 2004)

##### **How to differentiate the polyembryonic and sexual seedlings**

It is difficult to differentiate in the nursery. Generally more vigorous seedlings are considered to be polyembryonic. By rejecting about 10 per cent of weaker and weakest seedlings, one can have fairly uniform polyembryonic seedlings.

**Vegetative apomixis**

This is not common in fruit crops. However, in some cases like *Poa bulbosa* and some *Allium*, *Agave* sp. produces vegetative buds or bulbils instead of flowers in the inflorescence.

**Apospory**

Sometimes when, embryo sac develops from archisporial or from the nucellus or from other cell. If it is develops from haploid megaspore cell it is known as generative or haploid apospory. On the other hand, if it develops from diploid cell i.e. nucellus of other cells it is termed as somatic or diploid apospory.

**Parthenogenesis**

Development of embryo from egg cell with or without pollination but without fertilization. Depending upon the ploidy levels of egg cell, parthenogenesis can be haploid (non recurrent) and diploid (recurrent) e.g. mangosteen (*Garcinia mangostana*).

**Apogamy**

Development of embryo from synergids or antipodal cells within the embryo sac with or without pollination but without fertilization is termed as apogamy. This type of apomixis is also grouped into haploid and diploid apogamy depending upon the ploidy level of cell. Diploid apogamy is recurrent whereas haploid apogamy is non recurrent.

**Androgamy**

Development of the embryo from male gametes inside or outside of embryo sac is known as androgamy. Since the cells are haploid in nature therefore, it comes under non recurrent type (Shukla et al., 2004).

**Budding**

Budding is one of the type of vegetative (asexual) reproduction in which a new organism develops from an outgrowth or bud through cell division at one particular site. On the other hand, budding is nothing but bud union of one plant (scion) onto another (rootstock). The process of connecting scion, which is bud and rootstock in a manner such that they unite and grow successfully as one plant is termed as budding. Thus, the bud which develops into frame work branches, flowers and fruit is termed as scion. The portion over which bud is united which

provides supportive stem and root system to the plant is termed as root stock. Budding is to be done when sap flow is active and the plant is growing successfully. During this period, taking out bud from scion stick becomes easy because of continuous cambial cell division thereby the chance of bud union increases. Spring, summer and rainy season accordingly, i.e. March- April, May-June and July-September are to be considered suitable for budding. Rootstock 1 to 2 years old along with pencil thickness is to be selected for budding and its types are grouped as such

T-budding or shield budding is a special union technique of rootstock and scion consists of single bud. Successful T budding requires, scion material along with fully-formed, mature, dormant buds. Wherein, the rootstock is to be having features like a) active growth b) bark along with slipping nature c) actively growing vascular cambium and d) easily peeling of the rootstock bark with little twist. T budding can be done in certain fruit tree like peach in June by using cold stored bud sticks and field grown seedling rootstocks. Many deciduous trees are to be budded in late July or early August when the current seasonal buds have developed fully and are dormant by using field grown seedlings that have slipping bark. Bud sticks having plump, healthy buds are considered suitable for union (budding) as a scion. These bud sticks are to be selected from current seasonal growth and leaf blades are to be clipped from the bud sticks leaving the petiole intact as convenient handle for holding the bud while it is removed from the bud-wood. The cut is to be taken about 1.25 to 2.0 cm below the bud and deep enough into the wood so that when the cut is finished, the bud and bark of wood are cut off. A perpendicular cut across the top of the upward cut will separate it from the bud stick. Budding knife should be very sharp for removal of bud from bud wood and incision on stock, as little possible bud damage. Bud is to be inserted inside the incision taken on rootstock, immediately as sap flow is active. A vertical incision on rootstock is to be taken deep enough to ensure easy separation of bark at

cambium. The T is then crossed i.e. a perpendicular cut (horizontally 0.7-0.8 cm) is to be taken at the upper end of the vertical cut. In a situation of heavy rainfall during the budding time or in species in which the rootstock is likely to bleed heavily, inverted T bud can be taken to prevent water or sap from pooling in the graft. The bark is carefully slipped from the rootstock stem exposing a pocket into which the bud shield can be inserted. Care is being taken that not to tear the flaps of bark in the process of spreading them. If the bark does not slip easily thereby the stock is not in active growth and the budding is to be done afterwards when active growth is to be resumed (Yadav & Singh, 2018). The bud shield is carefully slipped in between the bark flaps. The top of the bark strip on the bud shield is to be trimmed to fit tightly against the horizontal cut so that the bud fits within the incision. The bark flaps are to be held tightly against the bud as they are wrapped with polythene tape. Tape is to be removed in 2-3 weeks after the union has healed. As the union has healed, the upper part of the rootstock plant can be cut away to force the bud to grow (for June budding). If the union is done in the late summer, the bud needs overwinter prior to resuming growth. In this case, the upper portion of the rootstock is usually removed during the dormant season either in late winter or early spring. After the upper portion of the rootstock is removed the scion bud grows vigorously. Fruit trees like pummelo, almond, aonla, ber, custard apple, grape fruit, mandarin, sweet orange, peach, plum, olive etc. are propagated by T-budding. The bud along with petiole may indicate the condition of the budding, as the shield bud is shriveled and the petiole does not falls at normal touch then the bud is to be considered possibly dead thereby the budding is to be repeated.

The inverted T-budding technique is exactly same as the normal T-budding except that the horizontal cut is to be made at bottom end of the incision. In this case, the bud is to be cut from the bud stick by starting above the bud and exiting below it. Inverted T-budding may be effective due to the downward flow of

hormones that are intercepted below the bud. Therefore, the union will be stronger thereby the healing process will hasten (<https://www.angrau.ac.in>).

Chip budding method is practiced when there is lack of sap flow and bud does not slip easily from the bark. Chip budding does not use the protective bark flaps as T-budding but it also does not use slipping bark. The first step is to make a cut about 1.5 to 2.5 cm long with a depth of  $\frac{1}{4}$  to  $\frac{1}{5}$  the diameter of the stock with a horizontal cut on the bottom, the cutting can be removed. The bud stick and stock must have same diameter. The first cut on both stock and scion is to be taken at a 45- 60<sup>o</sup> downward angle to a depth of about 3 mm. Chip size of the cuts on both the scion and rootstock is to be exactly same. The stock and scion must be placed together in such a way that allows the cambia of the bud and stock to match together as much as possible. Polythene tape wrapping plays key and vital role in chip budding. Desiccation is a high risk in this method; therefore the wound should be wrapped tightly with grafting tape. Healing and sprouting can take place within a month. Budding in early summer, may be desirable for the bud break and grow during the same season. In this case, the stock top should either be removed entirely or broken over within a few weeks of budding to encourage the scion bud break. For plants budded in late summer, the tops is to be removed just before the growth starts. Unwanted sprouts are to be removed periodically to get more per cent success. These sprouts are to be rubbed off as soon as they are visible so that they do not reduce the growth and quality of the budded stock. Chip budding can be done in mid to late summer unlike most budding which takes place in the early spring. *Citrus* sp. and jamun can be propagated by this method.

Patch budding is mostly successful and probably simple methods of budding than other methods due to ease in removing rectangular patches of the bark. It is widely used in plants those having thick bark that can be easily separated from the wood. Fruit plant

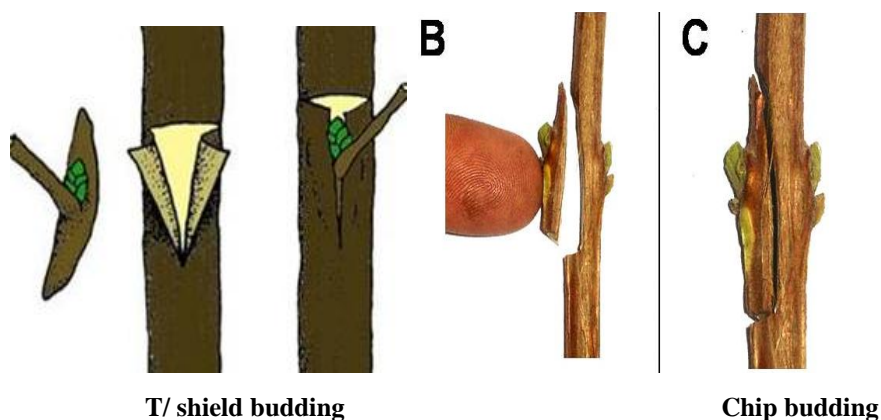
like aonla, bael, jamun, pecannut etc can be propagated by patch budding (Gupta, 2014). In this method, square or rectangular shape bud is to be taken from scion shoot. Similar incision (2-3 cm) is to be made on the rootstock. The bud patch is to be carefully removed intact and inserted into the rootstock followed by polythene tape wrapping, immediately leaving bud's spouting portion exposed. Wrapping prevents desiccation of bud thereby favours bud sprouting.

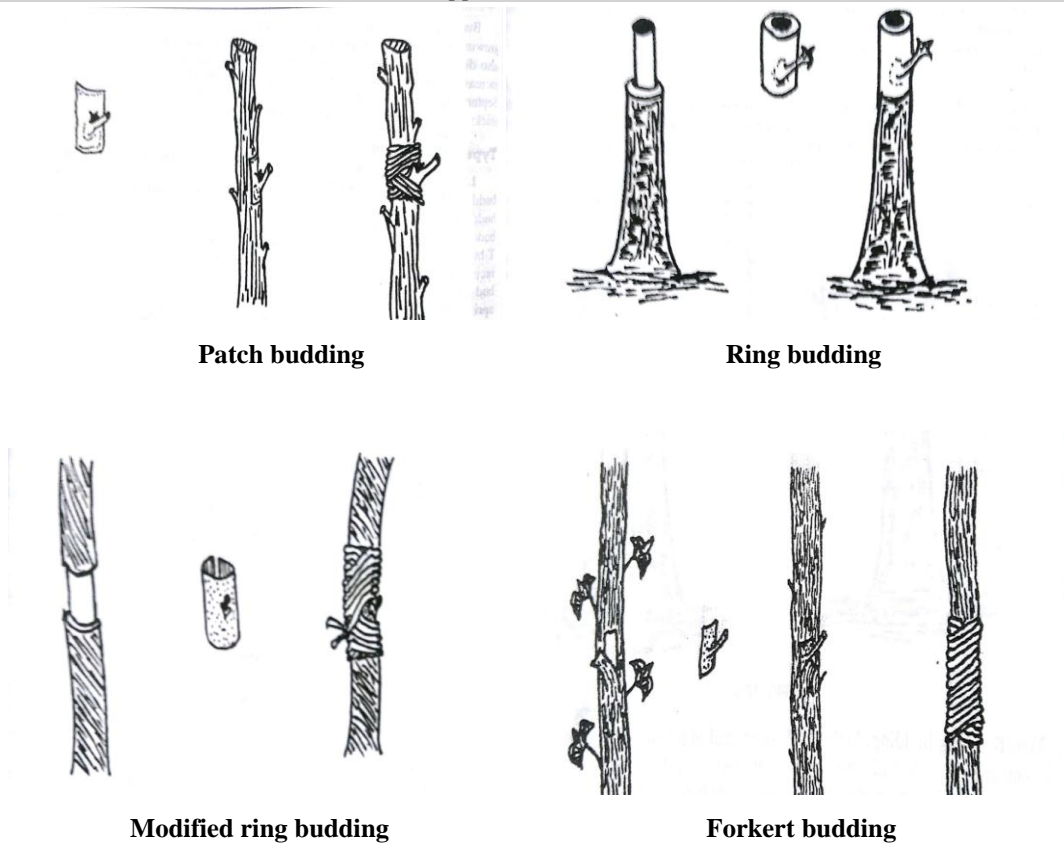
I-budding is a method, in which incisions in the shape of English capital alphabet 'I' is to be made in the bark of the rootstock by a single vertical cut and a horizontal cross-cut at both the ends. A rectangular bud patch similar to that in patch budding is then inserted in this incision (Yadav and Singh, 2018).

Ring or annular budding is rarely used because of bud union failure thereby plant get die. Ring budding can be adopted for propagation of fruit plant like ber, peach etc. In this method, ring shape bark consist of bud along with 2.5 to 3 cm length is to be taken from scion shoot. At terminal end of rootstock, incision similar in size of bud is to be made. Then bud wood is to be placed on rootstock, tightly in the central portion e.g. ber (Singh, 2011 and Gupta, 2014). On the other hand side modified ring budding (Flute budding) is one

of the methods of propagation. In this method instead of taking out complete circular ring (2.5 cm ring) of bark consists of bud is to be taken from scion shoot by taking a vertical slit in bud wood. At suitable portion of rootstock similar size bark is to be removed. The scion is to be fitted on rootstock and wrapped using polythene 300 guage, immediately e.g. ber, walnut, pecan nut etc.

Forkert budding (form of patch budding) is one of the budding methods. In this method, patch shape bud having 2 to 3 cm length and 0.5-1.0 cm width is to be taken from scion shoot. Similar size incision is to be made on rootstock at a height of 5 to 10 cm. While making incision the vertical flap of the bark is left intact with lower portion of the rootstock. The patch of bark consists of bud is fitted in the incision made on rootstock. The bud is to be covered with flap of the bark and then wrapped using polythene tape. After 15-20 days, union can be completed and the wrapped polythene is to be removed (Singh, 2011). Fruit crops like mango, jackfruit and cashew can be propagated by this method. Modified forkert budding is one of the methods of budding in this method, the vertical flap of the bark covers the base portion of bud only. Thus, to remove flap as in forkert methods is not required and remaining all procedures are similar to forkert budding.





**Patch budding**

**Ring budding**

**Modified ring budding**

**Forkert budding**

**Plate: Methods of budding**

Source of images (Yadav and Singh, 2018), (Singh, 2011) and (<https://www.angrau.ac.in>)



**Slipping bud**



**Budded rootstock**

Image source (personal photo collection)

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