ACTA SCIENTIFIC NUTRITIONAL HEALTH (ISSN:2582-1423)

Volume 5 Issue 2 February 2021

Research Article

Seedlessness in Hybrid Fruits is a Low-input Resource for Canning Industry

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Received: November 11, 2020

Published: January 16, 2021

Abstract

So many dual-purpose tropical fruit crops, with both vegetable and fruit use, were unexplored until today to study in-depth about the physiological control of their genomes. These studies would be useful for modern fruit breeding programs aiming 1) seedless hybrid fruits for canning, pulp and food industry 2) dwarf hybrid fruit trees and 3) for improving organic quality of fruit production with pesticide free as well with disease and pest resistance. The mechanism, techniques and various kinds of male-sterility (ms) and their major role in seedless fruit production while connecting ms to the concepts of self-incompatibility, parthenocarpy and poly-embryony were reviewed. The physiological parameters contributing ms, responses and success of ms in different fruit crop's F1 production with seedlessness in their fruits, markers identified for ms and or seedlessness were explained along with manipulating nuclear-mitochondrial gene controlled biochemical pathways at the tissue and cellular level during their flower-bud and fruit development. The routes for possible success in producing hybrid seedless fruits and benefits in the improvement of available unexplored nutritious fruits of tropical origin were detailed in this chapter.

Keywords: Mutation; Parthenocarpy; Perennial Fruit Crops; Morphological and Molecular Markers

Abbreviations

ARF: Auxin Response Factors; CHAs: Chemical Hybridizing Agents; cms: Cytoplasmic Male Sterility; c-gms: Cytoplasmic-Genic Male Sterility or g-cms - gene-cytoplasmic male-sterility; EB: Ethidium Bromide; EMS-Ethyl Methane Sulphonate; EU-European Union; F: Fertile; FAO: Food and Agricultural Organisation; GA: Gibberellic Acid; gms: Genic Male Sterility; Gy: Gray; ha-hectares; IAA-indole-3-acetic acid; JA: Jasmonates or Jasmonic Acid; Kr: Kilo Radians; MH: Maleic Hydrazide; ms: Male Sterility; MMCs: Microspore Mother Cells; mtCMS-Mitochondrial cms; NAA: Alpha Naphthalene Acetic Acid; S: Sterile; t: Tonnes or Tons; US: United States; USA: United States of America; USDA: United States Department of Agriculture.

Introduction

Tropical fruits are with either food and energy values or nutritive and health value. Some fruits that we consume on a daily basis were not even known to us that they are fruits. For example, cashewnut and coconut are commercially valued for nut, the cashew apple and coconut water were consumed in summer for nutrition, instant energy, tasty sweet juice and for rehydration from excessive heat in tropics. Fresh fruits of apple, banana, mango, jackfruit, guvava, sapota, custard apple, pineapple, strawberry, oranges and citrus add vitamins, carbohydrates and sugars to our daily energy. Underdeveloped fruits like for example thati, etha, neredu, regi, usiri, seema-chintha, bael, and tamarind were not even felt the fresh fruit appearance by present day generations, though they have lots of health values. Overall, tropical fruits are a viable economic commodity to choose in the research market with a lot of exploitable diversity in their origin, morphology and taxonomy for development of seedless hybrid fruits (Appendix 1; Figure 1).

Figure 1: Examples of seeded and seedless fruits in parthenocarpic fruits. (A) *Pastinaca sativa*, (B) *Bursera aptera*, (C) *Elaeis oleifera*, (D) *Annona squamosa*, (E) *Actinidia arguta cv Issai*, (F) *Cucumis sativus*, (G) *Citrus clementine*, (H) *Musa acuminata banksia*, (I) *Solanum muricatum*, (L) *Solanum lycopersicum* (seeded and pat-2 fruits in the genetic background of cv Super Marmande).
(Source: Curtosy of [17]).

Global fruit production

The largest fruit-producing states in the United States of America (USA) are California, Florida and Washington. California accounts for 65 percent of the United States (US) non-citrus fruits like strawberries, grapes, bananas, apples, dates, figs, kiwifruit, olives, Clingstone peaches, dried plums, raisins and 30 percent of US citrus fruit (oranges and mandarins) production. The state's contribution of fruit and nuts accounts to 16.7 million tons of $20 billion value, equal to 52 percent of the national total. A typical American consumes around 286 pounds of fruit and tree nuts including fresh and processed products each year [1]. The United States of America and the European Community are the largest import markets for tropical fruits. The major challenges confronting tropical and subtropical fruit exporting countries are issues related to the smallholder production systems, post-harvest handling technologies, sanitary and phytosanitary (SPS) regulations, food safety assurance requirements and quality standards instituted by importing countries [2]. The non-citrus fruit production (169 to 249 thousand tons) and acreage (29 to 39 thousand acres) increased from 2003 to 2012 with an increase in income from $95,000 to $267,000. These include apricot-plum crosses (aprium, plumcot, pluots), black walnuts, chayotes, cherimoyas, chestnuts, feijoas, guavas, jojobas, jujubes, kumquats, limes, loquats, Macadamia nuts, persimmons, pomegranates, prickly pears, quince, citrus by-products [1].

Tropical and subtropical fruits like banana, mango and pineapple increased by 19.2 percent to an estimated 270 million tonnes in 2004 by developing countries including 66 percent contribution from Asia to earn income from exports to developed countries and for nutrition [2]. India produces a vast variety of fruits with high nutritional and health values. The largest producers of the major tropical fruits are India, Brazil and China while the largest exporter of tropical fruits is Mexico. Currently out of 2700 fruit species, 58 percent of world tropical fruit production estimated from four major fruit crops (avocado, pineapple, papaya and mango) originates in Asia and out of which 28 percent contributed by India, 25 percent by Latin America and 16 percent by Africa (www.fao.org, IMF world economic outlook, 2017).

Tropical fruit crops

These may include, but not limited to mango, sugar apple (custard apple), durian, jackfruit, breadfruit, pineapple, papaya, avocado, tamarind, banana and plantain, guava, yellow passion fruit, sweet orange, lychee, loquat, and sapodilla. There were minor fruit trees of tropical origin like Seema-chinthakaya or monkey pod (*Pithicellobium dulce*), Chinni usirikaya (star gooseberry, *Phyllanthus acidus*), Pulla regi pallu and Regipallu (*Ziziphus sp.*), Bilimbi tree (tree sorrel, *Averrhoa bilimbi*), Neredu pandu, Thatipandu, Eetha pandu, Ramaphal and Lakshmanaphal in telugu speaking region of Andhra Pradesh (AP) in India. Most edible fruits belong to the tropical origin and perennial tree group providing instant energy and nutrition to rural poor at low or no cost. Their diverse morphology in flower and fruit structure may be utilized for development of hybrid seedless fruits using the genetically highly conserved genes for physiological mechanisms of male-sterility or self-incompatibility that modify the fruitset and control the gene pathways for seedlessness.

A few among many important tropical fruits (Appendix 1) with diversity in edible fruit and floral morphology were described with an objective of crop improvement in section 3 of this chapter as well as listed below with nutritional value of these fruits, statistics of production, consumption, income and export value with lead producing country or state to explain the attention needed to conserve and exploit the existing diversity for producing table fruits with seedlessness or to induce male-sterility.

Bael or stone apple *(Aegle marmelos, Rutaceae)*

Its synonyms are *Feronia pellucida* and *Crataeva marmelos*. It is a drought-tolerant fruit crop that grows up to an altitude of 1200 m and a temperature ranges of -6.67 to 48.89 ^{o}C in India. It is a sacred fruit of Hindus, cultivated in pagodas and the most common fruit of South India and South-East Asia. Slow-growing spreading tree of medium size (12-15 m) height with short trunk with a clear, gummy sap, resembling gum arabic, exudes from wounded branches and hangs down in long strands, becoming gradually solid. The deciduous, alternate leaves, borne singly or in 2's or 3's, are composed of 3 to 5 oval, pointed, shallowly toothed leaflets of 4 -10 cm long and 2-5 cm wide with a terminal one having long petiole. These leaves are used for religious purposes. Bael fruit is similar but larger in size with few or no seeds compared to wood apple (*Feronia elephantum*) with small size fruits containing more than 500 seeds. The fruit pulp of both has long storage life and made into various edible food items like syrup, sweet and salted snacks and pickles. Pulp powder is used for reducing dysentry with no blood. Both half-ripe and full-ripe fruits are with medicinal use in reducing diarrhea, land-scurvey, fever, typhoid, stimulant, stomach ache, antipyretic, antiscorbutic and beneficial influence on the membrane of the alimentary canal. The leaves (carminative) and gum (demulcent and emollient) of wood apple also medicinally useful besides its fruit (refrigerant). Budding and air-layering are the common methods of propagation. A tree may yield 800 fruits in the peak season [3,4].

Cashew (*Anacardium occidentale, Anacardiaceae*)

Cashew nut and cashew apple are commercial products; commonly grown in coastal areas of South-East Asia, Brazil, Vietnam. Vietnam (860,060 t) is the lead country followed by India (745,000 t) in producing cashew kernels. In AP, cashew is produced in 8 major districts including East-Goadavari with a raw-nut production of 12,500 annually from 46,913 ha area. Around 20,000 people from a district are dependant on cashew industry for employment. Consumption of the 100g of raw cashew nut provides 553kcal energy. India earns around 200 million dollars by exporting 40-50 t of cashew kernels. The largest buyers or importing countries include the USA, Europe, Japan, Australia, Canada, Singapore and the Middle-East countries (www.fao.org; https://www.nda.agric.za/). There are two types of cashew cultivars classified as 1) the common or giant type that includes *A. occidentale* L., and 2) the dwarf type. Common types grow usually to heights ranging from 5 to 15 m in South-East Asian countries, with a crown diameter of 12 to 14 m while the dwarf types grow up to 4m height and with a crown diameter of 6-8m in Brazil.

Coconut (*Cocos nucifera, Arecaceae*)

Copra and coconut water, popularly known as the tree of life. It is a large palm growing up to 30m height with pinnate leaves of 4-6 m long and each pinnae measures 60-90 cm long. Old leaves break away easily leaving the smooth trunk. There are dwarf (Malayan dwarf, Fiji dwarf, dwarf yellow, dwarf orange) and tall (Jamaican tall, Eastcoast tall, Westcoast tall, Hainan tall) varieties. On an average, a tree can give 30 to 75 fruits per month in AP. There are three fruit types yellow, red, green, and a hybrid between red and green. The recent discoveries of coconuts in cooler climates for landscaping purposes profit regions of not less than USDA zone 9. The fruits of these ornamental coconuts (*Syagrus romanzoffiana*, queen palm from the USA; *Beccariophoenix alfredii* from Madagascar) looks similar to coconut and can grow slight cooler climates when compared to the real coconut palm. A full-sized coconut weighs 1.4kg. Coconut stem is used for timber, leaves for thatch roofing and handicrafts, coconut sap from spathe processed into vinegar, wine or sugar, the kernel as a source of oil and cocoa cream/milk, the husks as a source of fiber/coir for various uses and the coconut water for beverage, vinegar and also wine. The coconut shells for charcoal, fuel, handicrafts and are tourist souvenir items in Asia-Pacific countries. Coconut is planted in about 12 million ha globally of which 85 percent are grown in the Asia-Pacific region with Indonesia being a leading country followed by India in production. Most coconut production about 96 percent is in the hands of smallholders tending 4 ha or less of land or practice share-cropping if they do not own the land. Copra is the dried kernel from which oil is extracted and copra yields range from 0.3 to 1.5 t/ha whereas hybrids yield 2-6 t/ha from hybrids. Introduced or exotic cultivars are mostly found in research stations for characterization and on-farm evaluation or for testing as breeding materials. From 1950 to 1993, more than 400 hybrids were developed worldwide under established coconut improvement programs using various breeding strategies [5] (Batugal, 2009). Traditional areas of coconut cultivation in India are the states of Kerala, Tamil Nadu (TN), Karnataka, Puducherry, AP, Goa, Maharashtra, Odisha, West Bengal and, Gujarat and the islands of Lakshadweep and Andaman and Nicobar. The coconut is a national tree of Maldives. It is also grown in the Middle-East, Pakistan, United States, Australia and Bermuda. Coconut is highly tolerant to salinity and drought, but needs high humidity for optimum growth. The temperature and rainfall required for uninterrupted growth are 12-13°C and 1000mm, respectively. They can recover from temperatures of -4°C, but require full and direct sun.

Cherimoya (*Ramaphal, Annona cherimola*), Custard apple (*Annona reticulata*), Soursop (*lakshmanaphal, Annona muricata*) Sweetsop or sugar apple (*Annona squamosa*)

All these three fruit species belong to the same family *Annonaceae* cultivated for their edible sweet juicy fruit pulp. Cherimoya also known as custard apple in some regions. It is the most common fruit weighing 150 to 500gm. The leaves of cherimoya are single and alternate dark green and slightly hairy on the top surface. The tree is evergreen low branching, spreading tree or shrub measuring 5 to 9m tall. The hybrid "Atemoya" from *A.cheirmola* and *A.squamosa*, received attention in West Africa, Australia, Brazil and Florida. It is cultivated in cool high altitudes with Central America being its origin. It is mainly cultivated in the Mediterranean region of Southern Spain, and Portugal, several countries of Africa, the Middle-East and Oceania, Andean regions, Bolivia, Peru, Chile, Equador and Colombia, Americas including Hawaii and California since the 1700s. The fruit of "soursop" are the largest fruit of this genus, ovoid measuring 30cm long with edible juicy sweetish-sour acidic whitish fruit pulp with a moderately firm texture inside emitting aromatic flavour and the fruit is covered with dark green and prickly skin. The *A. muricata* is tolerant of poor soils in tropics and cannot withstand severe frost. Cherimoya seeds are poisonous if crushed open. A neurotoxic acetogenins such as "annonacin" is found in soursop and cherimoya trees, which may cause paralysis in humans if its extract is injected. The pulp from these groups of fruits is used to make nectar, smoothies, fruit juice drinks, candies, sorbets and ice cream flavorings. All these fruits are rich in carbohydrates, protein, vitamins A, B_2, B_6, C, dietary fiber, and readily available sugars for energy. Fruits of Annonaceae were of domestic and local farm income value by filling the local fruit basket. The annual earnings from export are very limited due to its less durability in keeping its phenotypic fruit appearance and tight internal pulp texture from the first day of ripening. Watery and sweet sugary pulp exudes out of over-ripened fruit, it is very tough to transport these fruits to long distances from the site of production, though some income gained in recent years due to its value in flavouring and value added-food industries for making icecreams, and puddings by mixing with other fruit pulps.

Eetha pandu or date palm (*Phoenix dactylifera, Arecaceae*)

Cultivated for edible sweet fresh ripened fruits as well as for dried pitted dates. Originated in the region between Egypt and Mesopotamia. There were 12-19 species in the genus *Phoenix*. Date trees typically reach about 21–23 meters in height, growing singly or forming a clump with several stems from a single root system. The leaves are 4–6 m long, with spines on the petiole, and pinnate, with about 150 leaflets. The leaflets are 30 cm long and 2 cm wide. The full span of the crown ranges from 6–10 m. Cultivated in North Africa, Middle East, South Asia, Mexico (Sonora and Baja California) and the USA (S. California, Arizona and S. Florida). The total annual world production of dates accounts to 8.5 million metric tons. The Middle-East and North Africa are the largest producers. Lead producing countries are Egypt (1,590,414 t) and Iran (1,185,165 t) followed by Algeria (1,058,559 t) in 2017 (UN FAOSTAT:Crops). Dates were introduced to Mexico and California by Spanish in 1765. There were so many developed varieties and hybrids among which Medjool and Degler Nour are the two domesticated date palm hybrids of North Africa. Date palm takes 4 to 8 years to bear fruit and viable yields with economic returns will be only after 7 or 8 years.

Immature dates are used as a vegetable. A mature date palm can produce 150-300 lb (70-140 kg) of dates per harvest season. Dates can be chopped and used in a range of sweet and savory dishes, including puddings, ice-creams, chocolates, bread, and syrup. Vinegar is the traditional product made out of dates during Ramadan in the Middle-East. Dates provide essential minerals and nutrients, dietary potassium and sugars of about 80 percent from ripe dates. Seeds are used as animal feed. The fruit clusters are used as brooms, leaves are served as a source of mats, screens, baskets, roof of a hut, fans, fuel and pulp.

Jackfruit *(Artocarpus integrefolia, Moraceae)*

Fresh fruit and cooked seed are edible. It bears the largest fruit of 55 kg in weight when compared to the other tree-fruits. Its origin is western ghats of Southern India and the rainforests of Malaysia. It is cultivated in South-East Asia, Bangladesh, Americas and Africa. *Artocarpus heterophyllus* Lam., is a member of the family Moraceae too. The family encompasses about 1000 species in 67 genera, most of which are tropical shrubs, trees and a few herbs. The genus *Artocarpus* includes about 110 tropical species.The varieties cultivated in Americas include jac-dura, jaca-mole and jaca-manteiga. A matured jackfruit is a multiple fruit composed of hundreds to thousands of individuals flowers and the fleshy petals of unripe and ripe fruits are edible. The fruit contains 100-500 seeds which are also edible and rich in protein. The jack fruit is a national fruit of Bangladesh and SriLanka whereas the state fruit of Kerala and TN. The raw fruit and seed are used as a vegetable in making curries, fries, sweet syrup, custard, cakes, icecreams and chips. The tree wood is used for manufacturing furniture, musical instruments and house construction. India is the lead producer with 1.4 million tones of fruits followed by Bangladesh, Thailand and Indonesia in 2017. Jackfruit industries are established in Srilanka and Vietnam where the fruit is processed into flour, noodles, papad and icecream or canned and stored for marketing fresh vegetable fresh fruit and to export.

Neredu pandu or Jamun *(Syzygium cumini, Myrtaceae)*

Fresh fruit is sweet and edible. The slow growing tree reaches up to 30m height and lives up to more than 100 years. Its leaves have aroma similar to turpentine, glossy dark green. It is native to the Indian sub-continent, adjoining regions of South-East Asia, China and Queensland. This tree was introduced to Florida in 1911 by the United States Department of Agriculture (USDA) and to Brazil from India during Portuguese colonization. Now it is commonly grown in Suriname, Guyana, Trinidad and Tobago. The fruit is effective against hyperglycemia, rich in vitamin A, C and most frequently used in Ayurveda, Unani and Chinese medicine. It is widely cultivated in Maharastra, AP, TN, Kerala and Karnataka states of India and no available information on lead producing states or countries or exporters or importers of this fruit.

Thati pandu or toddy palm or palmyra palm (*Borassus flabellifer, Arecaceae*)

Its matured ripe fruit's sweet pulp as well as translucent pale-white, sweet jelly seed sockets filled with watery fluid inside a matured unripe fruit are edible. The robust tree reaches a height of 30m with a long trunk with leaf scars. Leaves are fan-shaped. Young palmyra seedlings grow slowly, producing only a few leaves each year giving a substantial stem in no time. It is native to the Indian subcontinent and South-East Asia including Nepal, India, Bangladesh, Srilanka, Cambodia, Laos, Burma, Thailand, Vietnam, Malaysia, Indonesia, the Philippines, Pakistan, and parts of China. Being a minor fruit crop, no lead country for production or export is reported, but major producing regions of India were from Tamil Nadu, Andhra Pradesh, Telangana and Bihar, and in Jaffna, Sri Lanka. This fruit is a rich source of Vitamin A and C. The sap extracted from this tree's top shoots and collected in earthen pots which is used to make sugar, fermented alcoholic beverage of local use or Tadi and jaggary. The fruit pulp is made into various food items (thati garelu and thati chapa in telugu), syrups, candies. The seeds are planted and made to germinate to harvest the fleshy seed sprouts and stems of the underground (thegalu in telugu). These "thegalu" are roasted or boiled to eat as a snack and these are rich sources of fiber, carbohydrates with resistant starch. The germinated seed's hard shell is cut open to have a crunchy kernel (Burragunju in telugu) which is also edible when fresh and eaten as raw. The origin of leaf segment inside a crown is an edible cake known as "Thati adda". The leaves are used for thatching, mats, baskets, fans, hats, umbrellas, and writing materials. In ancient Indian times, these leaves are used to write useful information and were preserved a lot of South-Indian literature and are known as "Thaala-Patra Grandhas". The trunk is used as a fence.

Mango (*Mangifera indica, Anacardiaceae*)

Fresh ripe and unripe fruit are edible and the crop is native to South Asia. Mango trees grow to 35 to 40m tall with a crown radius of 10m. The leaves are evergreen, alternate, simple. The average growth rate in the world's total production and export of mango is 3.48 percent and 4.64 percent, respectively, during 2007-2016. The lead producing continents were Asia, Africa, Latin America and the Caribbean, while the lead countries of production include India, China and Thailand. The lead exporting continents were Latin America and the Caribbean followed by Asia and Africa, while the lead exporting countries were Mexico with 80 percent of the US's entire shipment and Peru. The fruit is consumed mostly by the USA and the European Union (EU) between September and May which gives maximum price to the exporters though the export price in the US dollar per ton of mango dropped gradually since 2007. Matured unripe fruit is used as a vegetable in making curries, fresh and preserved pickles, soups, and whereas ripe mangos are used in making fruit juices, candies, syrups, jellies, fruit and nut bars, sugar-coated glazed dried fruit pieces, etc. The fruit is rich source of carbohydrates, vitamins A, C, minerals, sugars and fiber. Mango leaves are sacred and used during all Indian festivals, marriage ceremonies to decorate their homes. Mango tree's wood is made into useful furniture.

Tamarind (*Tamarindus indica L.*)

Belongs to the family *Leguminosae* subfamily *Caesalpinioideae*. Fresh raw immature, unripe and ripe fruits are edible. It is cultivated in Asia, African countries including but not limited to Ethiopia, Kenya, Uganda, Nigeria, China, India, Indonesia, Malaysia, Myanmar, Nepal, Phillippines, SriLanka, Thailand, Vietnam. India, Thailand, and Mexico are the lead producers and exporters of tamarind fruit. The fruit is pod borne in clusters of 5 to 10 containing 1 to 12 seeds per pod. Fresh fruits are made into vegetable curries, tamarind rice, chutnies while ripe fruits are made into soups, drinks, syrups. The leaves are used for making dahls and chutney. The soft pulp from ripe fruits is processed and stored into large planks or into a thick paste to export. Seeds are edible as snacks after drying and cooking. These fruit and seeds are sources of fresh energy, carbohydrates, protein, vitamin A, B_1, B_2, B_3, and C, folate, many minerals like Zn, Mg, Fe, P, K and sugars. The tree trunk might exude gums of value. The byproducts like husk, dried leaves, twigs are used for agriculture farm manure or animal feed. The tree wood is useful for valuable furniture. In African countries, Tamarind fruits are exported to the Middle East, Italy, India, Thailand, Philippines, Somalia, and Tanzania from where value added products such as flavours, sauce, spice, flour, sweet chutney, kernel oils are made. The Indian exports are confined to Americas and European Union.

Mechanisms of male-sterility and seedless fruit production

Most of the fruit crop production is weather dependent. If the adverse weather is prevailing in growing regions of the potential fruit crops, the fruitset and production of quality and quantity of fruits will be reduced affecting annual farm and national income (www.fao.org). Therefore, the physiological factors affecting viable flower production for successful fruitset and fruit development is the primary requirement in tropical fruit crops. These physiological factors if affected by adverse climatic changes might result in heritable or non-heritable sterility in male floral organs. Some of these male sterilities might be expressed phenotypically as 1) sporogenous ms due to abnormality occurring between early and late microsporogenesis leading to production of nonfunctional pollen, 2) structural ms due to abnormal stamen production leading to no pollen production and 3) *functional ms* due to nonfunctional viable pollen produced which was unable to initiate fertilization and fruitset [6]. This indicates that normal development of pollen grain can be disrupted artificially in a number of ways in a female parent to develop ms lines for hybrid fruit production.

The genotypic expression of ms lines may be genic (gms), cytoplasmic (cms) or genic cytoplasmic (g-cms) depending on the subcellular gene function. The submicroscopic and subgenomic structural variations like inversions and translocations in mitochondria and chloroplast organelles might cause complex gene recombinations leading to genetic variation, polymorphism and infertility in land plants [7]. Many benefitted field crops from all three kinds of male sterility, their applications in autogamous plant species and some examples of cytoplasmic suppression of genic male-sterility were reported [6]. They also strongly pointed out the influence of nuclear genes affecting sporophytic and gametophytic tissues in gms systems. The genetic affect in gms system start from the sporogenous mass stage, the stage 1 of the microspore development, through the stage 10 (the last stage) while in g-cms systems, tapetum is the primary tissue where abnormalities occurring may to lead abortion of male cells.

The cms can arise spontaneously in breeding lines, following mutagenesis, as a result of wide crosses, or interspecific exchange of nuclear and cytoplasmic genomes. Sometimes, cybrids produced by embryorescue technique may result in cms inducing cytoplasm. The cms plants and restorers are very difficult to distinguish, the reasons for which may include 1) Morphological changes associated with male sterility: Premature degeneration of the tapetum layer of the anther causes cms. It has no molecular basis, but this tapetal structure plays an important role in microspore development and the degeneration of which usually begins soon after meiosis I or II, leading to visible identification of fertile and sterile plants 2) Identification of sterility associated genes and their structure: It is very difficult to identify the sterility associated genes due to the larger size of plant mitochondrial genome (200-2400kb). The phenotypic markers such as lesions in either mitochondria or chloroplast are due to the result of cms (Ex. Maize and Petunia). The cms-associated genes are chimaeric as they resulted due to the fusion of portions of known genes with previously unknown sequences. Therefore, cms associated genes may share common sequences that encode hydrophobic domain while cms associated ORFs won't share any such and are purely unrelated. 3) Comparative physical mapping, identification of differences in mitochondrial gene expression patterns between normal fertile(N-), male sterile(T-), restored fertile and fertile revertant plants: This can be achieved by molecular analysis, by employing RFLP analysis of mitochondrial DNA/genome to give details on cytoplasmic differences of cytoplasmic male sterility from fertile line and cms associated ORFs conferring ms. The alterations of these genes restore fertility to male-sterile cytoplasm (T-). The second approach is by tissue culture induced mitochondrial mutations that restored fertility. For example, to identify the cms associated gene in petunia, somatic hybrids of fertile and cms lines were constructed via protoplast fusion. Within these hybrids, the mitochondrial genomes of the two parents were able to recombine. The resulting recombinant genomes then segregated during subsequent cell divisions. Comparative mitochondrial RFLP analysis of ms and fertile progeny revealed a chimaeric ORF consisting of the 5' portion of the atp9 gene, parts of the first and second exons of the Cox II gene, and unidentified sequence "URFS" that co-segregated with ms. 4) Function of genes associated with ms: The ms genes are not directly associated with mitochondrial function. Therefore, correct tissue specific expression and subcellular localization are required for some cms associated proteins to cause either toxin sensitivity or ms [8-10]. The specific phenotypic morphological, genetic and biochemical markers identified were listed in specified sections of this chapter.

In general mutation breeding is used to generate sterility and create variability for high yield, pod and seed traits, plant height and pathogens. Here, selection for plant height is targetted for dwarfness by selecting genotypes with shorter stems in taller cultivars with all the potential yield attributes, so as for selection for seedlessness. About 40 - 60 percent of the sterile and semi-sterile population is discarded in M_1 generation when choosing for yield attributes to avoid any kind of undesirable chromosomal aberrations and physiological damages resulting in less viability of seedling progenies.

Gamma radiation has been widely used in citrus breeding to create variability through radiation-induced chromosomal aberrations resulting in female sterility in seedless mutants. These seedless mutants when intercrossed with its wild type, resulted in efficient fertilization as well as in seedless fruit growth due to strong female sterility [11]. A chemical mutagen, ethyl- methane sulphonate (EMS, 0.1-0.2%) was used to induce mutations by using cotyledons as explant in pomegranate and banana, while seeds were treated with gamma rays (10-25 Gy) in pomegranate cultivars "Kesar" and "Ganesh" in an effort to generate populations for bacterial blight resistance. The mutant types of dwarf, compact, prostrate, gigas, white flower non-flowering and sterile were induced by EMS in chickpea. The female sterility in induced mutants was controlled by a single recessive gene(fs) in chickpea [12]. Mutation treatment of 30 Kr to Hy-2 variety of pigeonpea showed a drastically changed plant type with complete female sterility and partial pollen fertility [13]. A lot of mutation work done in cereal, food legume crops and very few fruit crops, the information of which may form the basis for improvement of presented tropical drupe, pome and multiple fruited crops of dual-purpose use.

Haploid plants were produced, from culturing microspore of F_1 hybrid and without the use of mutagenic agents, for improvement of crop production by selection from a large number of variable gene combinations for oil quality, seed meal, and herbicide resistance. Haploids express both dominant and recessive genes showing their role in plant breeding, selection, transformation, and mutagenesis as well as for tissue manipulations. These haploids produced invitro are genetically fixed for plant production by treatment with colchicine for diploidy to have fertile true-breeding lines [14]. Similarly, when female parents carrying msms with the cytoplasm (S) was crossed with male parent carrying msms with the cytoplasm (F) will produce all sterile phenotype [(S), msms], while this female parent when crossed with male parent carrying MsMs with either S or F cytoplasm will result in fertile phenotype.

Commercially important traits developed by mutation are listed below [15]:

- Compact tree growth in apple (*Malus dometsica*) and peach (*Prunus persica* L.)
- Disease and pest resistance in apple (*Malus dometsica* L), grape (*Vitis vinifera* L.) and pear (*Pyrus communis* L.)
- Self-incompatibility in sweet cherry (*Prunus avium* L.) and apple (*Malus domestica* L).
- Fruit color in apple (*Malus domestica* L.) and grapefruit (*Citrus paradisi* L)
- Low acidity in apple (*Malus domestica* L.)
- Seedlessness in citrus (*Citrus* Sp. L.), watermelons (*Citrullus lanatus* L.), grapes (*Vitis vinifera* L.) and pomegranate (*Punica granatum* L.)

Therefore, hybrid seed production in fruit crops is adopted for a variety of reasons starting from plant canopy's morphological trait improvements to improved fruit yield and its quality attributes. Conventional breeding methodologies like introduction, mass and pedigree selection, various cross-breeding techniques of superior parents for any chosen trait are a common practice followed in fruit crops for development of hybrids by pomologist. Advanced molecular marker technology is applied for the marker-assisted selection of important phenotypic traits developed in various cultivars by mutation and tissue culture. Multiplication of improved fruit cultivars or F_1 hybrids is done by vegetative propagation methods like budding, grafting, cuttings and tissue culture for true-to-type fruit quality.

Male sterility (ms) in crop production is the mechanism of utilizing the male-sterile female parents for hybrid seed production to favor certain quality traits of phenotype or disease resistance or qualitative yield by genetic improvements through cross-fertilization with chosen fertile heterozygous male parent. Joseph Gottlieb Kölreuter, a German botanist first identified plant fertilization and observed hybrids in plants by discovering self-incompatibility and male sterility mechanisms in 1763 when working with tobacco and oranges.

Seedless crop plants must have the inherent ability to sustain fruit development in the absence of seed development during parthenocarpy or stenospermy. Parthenocarpy is the development of fruit without fertilization as seen in cultivated pineapples, bananas and in some citrus cultivars. The stenospermy is the development of fruit after pollination and fertilization, but without embryo formation or with aborted embryo due to independent genetic factors to complete normal seed formation and was observed in watermelons and seedless grapes [16].

The actual reduction in seed number in parthenocarpic plants is often achieved by combining mechanisms of parthenocarpy with self - incompatibility or male-sterility. Traits related to seedlessness and parthenocarpy can be introduced into genetic accessions through conventional cross-breeding methods. However, conventional breeding in *Citrus* species was observed with several limitations. First, these species have long juvenile (non-flowering) periods during which a relatively thick canopy develops, which limits the size of seedling populations that can be maintained for further evaluation. Second, they have a narrow genetic base, which limits the availability of alternative alleles that could be introgressed into other lines for the formation of a particular phenotype such as parthenocarpy. Third, breeding efforts are limited by the lack of knowledge of the mode of inheritance of specific characteristics. Fourth, and last, breeding efforts are limited by the polygenic nature of many important traits [15].

Approximately 20 sources of seedlessness have been described in tomato. Excluding the EMS induced mutation parthenocarpic fruit (pat), the parthenocarpic phenotype always emerged in biparental populations derived from wide crosses between cultivated tomato and wild relatives [17]. This indicates that wild ancestors are sources of conserved genes contributing seedlessness as these genes were lost or deleted during domestication and breeding cultivated genotypes of the present-day farming use.

Physiological factors contributing to the male-sterility and seedlessness

Pollen is produced inside an anther, the distal structure of filament in a stamen of the flower. Failure to produce functional anthers, pollen, or male gametes due to defects in the reproductive cycle during its major developmental stages leading to the sterility of functional male organelles in crop plants is known as male sterility. The ms may result in failure to release mature pollen grains due to defective sporogenous cell production, specification of stamen primordia, tapetum and microspore mother cells (MMCs) development, meiosis, formation of free haploid microspores, and degeneration of tapetum [18-20]. These physiological and functionally defective factors contributing to pollen sterility were discussed at the cell's hormonal and molecular level below.

Pollen is the career of male genetic factors (male gametes) namely polar nuclei. Plant growth hormones including maleic hydrazide (MH), Jasmonates (JAs), gibberellins, ethylene, abscisic acid, indole acetic acid (IAA) have a serious impact on pollen grain production and inducing ms [11]. The ms can be induced by over or under expression of genes controlling stamen development and thus producing defective anthers. The arrested stamen development at anthesis might result in short filaments, indehiscent anthers and non-viable pollen grains leading to ms.

In *Arabidopsis*, the Jasmonic acid (JA) deficient mutants, the overexpression lines of JA catabolism gene (CYP94B3), the JA signaling mutant "COI 1" and some transgenic lines are male sterile due to arrested stamen development at anthesis. The JA treatment might restore stamen development in JA biosynthesis deficient mutants, but not in JA signaling mutants [21]. The chemical hybridizing agents (CHAs) are known to produce male-sterile female-fertile flowers in larger proportions [11].

The mechanisms of ms and SI were most commonly used for the production of seedless fruits. During the transition from flower to fruit development stage, parthenocarpy or stenospermy is controlled by the complex changes in gene expression and regulation. The IAA, GA_1 and GA_3, brassinosteroides are positively and synergistically correlated with the growth of ovary and fruit after pollination and fertilization. The facultative parthenocarpy was resulted due to downregulation of chalcone synthase (Chs), and silencing of down-regulated genes coding for 53-aminoacid long peptides. The Auxin/IAA repressor proteins are ubiquitinated and degraded via proteasome to allow auxin response factors (ARF) mediated regulation of auxin-responsive genes and to form complexes that repress auxin signaling and action during initial phases of fruit growth [22].

Fruit morphology along with seedlessness is possible with negative regulators of GA signal transduction pathways that regulate the growth of ovary at anthesis, over-expression of ethylene biosynthesis genes before pollination which were negatively correlated with fruit development and positively correlated with parthenocarpy.

Cytokinins were increased during the seed and embryonic fruit development stages due to their key role in cell division, although their transcription regulation was still unknown to reveal their role in parthenocarpy. Alterations of brassinosteroid pathway were helpful to induce seedlessness when GA applied on unpollinated flowers [22].

Therefore, genetic regulation of auxin and GA biosynthetic pathways prior to anthesis might be a useful scientific approach for the induction of male sterility or production of sterile, aborted or nonfunctional male or female organelles in the flowers of fruit crops. The continuous supply of functional and active hormones, rate of supply, and their strength of expression as controlled by genetic factors are viable parameters to be considered due to their sensitivity in action at very low concentrations on fruit development and quality.

The concentration of endogenous GA in 'Satsuma' (obligatory parthenocarpic) reaches its highest level at anthesis, in contrast to the situation in self-incompatible cultivars like clementine (stimulative parthenocarpy) in which endogenous GA levels are not enhanced at anthesis. The concentration of ABA was high during the petal stage in mandarin. Thus, the relative balance between growth-promoting GAs and the growth inhibitor effect of ABA appears to play an important role in the regulation of fruit development in seedless mandarin. In pear fruit, after pollination, the level of GA increased indicating that seedlessness and parthenocarpy are controlled by GA. The parthenocarpic progeny, with cms mandarin cv. Satsuma as a female parent when grafted onto different pollen donors, segregating in the ratio of 1:1 to 15:1 and controlled by two dominant complementary genes unlike those were three dominant complementary genes favoring nonparthenocarpy in diploid Banana. Here all the pollen donors Clementine mandarin, Ellendale, Fortune, displayed parthenocarpy when grown in isolation indicating their self-incompatibility while Shani, Trovita, Niva and Wilking displayed self-compatibility [15]. Thus, seedlessness is not only controlled by triploidy and ms, but also controlled by growing conditions, GA and self-incompatible nature of cultivars.

Genetic and molecular factors directly control the biochemical pathways by altering the membrane and cellular enzyme biosynthesis. Proteins and major enzymes in the tapetal layer are controlling the growth-promoting pathways of developing pollen grain. The ms might result from the presence of mitochondrial-open reading frames (ORF) encoding cytotoxin peptides that are produced during the meiotic generation of microspores (pollen grains) inside an anther [11].

Cherimoya (*Annona cherimola*), sugar apple (*Annona squamosa*) Soursop (*Annona muricata*) were used as a food source in the tropical world including central and south America. Among these three, sugar apple was identified with seedless mutants for commercial production. This was due to the conservation of INO ortholog for "fruit without fertilization/auxin response factor 8" *(fwf/arf8)* mutation in mutants of sugar apple [16].

Spontaneous mutation of ploidy, colchicine treatment of monoembryonic citrus cultivars, and protoplast fusion lead to the production of autotetraploids in many polymebryonic citrus cultivars. These tetraploids displayed characteristics of the increased leaf thickness, differences in color and shape, slower growth rates, and longer juvenile periods besides the double times the actual number of chromosomes unlike diploid citrus cultivars. This feature made avenues for the development of somatic hybrids, cybrids (Ex: pumelo). Here, protoplast derived embryogenic, apomictic calli are fused with mesophyll derived propolasts, which usually do not divide leading to the production of triploids in citrus. Production of triploids occurs naturally in citrus and induced in watermelon [23].

Role of male-sterility in producing unique quality

More than 150 plants were identified with male sterility and utilized for various purposes in crop production. The majority of the work benefitted very few table fruit-crops, many temperate vegetable crops including cole crops, fruit vegetables like tomato, and cucurbits where male-sterility is induced by artificial means of chemical growth regulator sprays, by genetic means of pollen suppression, by mutation and natural selection.

The wide occurrence of seedlessness or parthenocarpy in fruit-crops (65%) is likely the result of selective pressure for seedlessness during their domestication and breeding. The pomologist's selection for parthenocarpic fruits is due to (i) release of fruit set from environmental constraints, (ii) advantageous for fruit processing, (iii) improvement in fruit quality, or (iv) simply represents a feature appreciated by consumers (Figure 1) [17].

The most commercial citrus fruits, mandarins, oranges and lemons were seedless with better fruit quality traits such as tender fresh texture with few slags, low acidity, and flavor. In commercially accepted cultivars of these fruit crops, with better fruit quality and excellent fruit shape, were induced with seedlessness caused by many factors such as male or female sterility, defective ovules and embryo-sac abortion, pollen self-incompatibility, polyploidy, abnormal climate, as well as the application of plant growth regulators [24].

The seedless trait in watermelons is a result of the cross-breeding of a normal seeded diploid parent with a tetraploid parent. Although fruits from these plants are considered seedless, when these plants were exposed to stress, fruits may develop pips-thin, edible, whitish, ovules - along with an occasional typical black seed. Here, in this kind of crop's physiological stress situations, the critical role of ms supports producing good quality seedless fruits.

Triploid citrus cultivars are thornier and have longer juvenile period, therefore seedlessness in citrus is introduced into cms and SI breeding lines by parthenocarpy. Seedlessness in "Satsuma" is caused by the combination of male sterility, female sterility, and parthenocarpy. To date, cms in citrus has been found only in 'Satsuma' mandarin and 'Encore' (*Citrus nobilis* Andrews x *C. deliciosa* Ten.). The ms in 'Satsuma' is used in citrus breeding program for producing seedless varieties or to introduce ms through the transfer of its cytoplasm for cybrid formation [25-27]. The seediness is less in ms progenies and self-incompatible cultivars compared to male fertile progenies and self-compatible cultivars. The seedlessness in citrus fruits was more in open field conditions, might be due to the availability of more pollen from heterozygous male-fertile cultivar, and this can be used to estimate the degree of female fertility and sterility. Kiyomi and Egami Buntan (*Citrus grandis*) are seedless due to ms and self-incompatibility. The Satsuma mandarin, Kiyomi and sweet Spring were used as parents to investigate the ms, fertility, seediness to develop seedless cultivars [28].

Seedless rasp-berry, eggplant, tomato and cucumber fruits with obligate and facultative parthenocarpy, early fruit growth, normal size and shape were observed due to ovule-specific transgene expression by modification of "DefH9" promoter and "*IaaM*" gene ((tryptophan mono-oxygenase) which has a functional role in auxin biosynthesis [22,29]. The literature presented here on genetic or physiological aspects of ms do not pertains only to tropical fruit crops because of its limited availability during literature search or might be due to limited parthenocarpic fruit or mutagenic research achievement in these crops as of today compared to other crops. The knowledge on mechanisms of ms is highly useful for the development of seedlessness in unexplored tropical fruit crops of nutritional and health value.

Few examples of seedless fruit production by genetically engineering hormone-related genes are listed below (Source: [22]; Note: Sl=*Solanum lycopersicum*; At=*Arabidopsis thaliana*):

- ***DeH9-iaaM* gene:** Its function is in "auxin synthesis" and genetic modification of "Ovule specific transgene expression". The fruit phenotype is observed without any vegetative alterations but with obligate and facultative parthenocarpy, seedless, early fruit growth, normal size and shape, especially in crops plants like tobacco, eggplant, tomato, rasp-berry and cucumber.
- ***rolB* gene:** Its function is in "auxin response" and genetic modification of "Ovary/fruit specific transgene expression". The fruit phenotype observed without any vegetative alterations and with obligate and facultative parthenocarpy, seedless, size and shape similar to weight in tomato.
- ***SlAA9* gene:** Its function is auxin signaling and genetic modification of "Antisense downregulation, constitutive promoter". The fruit phenotype observed with vegetative alterations and with parthenocarpy, seedless, early fruit growth, normal size and shape in tomato.
- ***AtARF8* gene:** Its function is in "auxin signaling" and genetic modification of "Expression of mutant AtARF8-4 gene". The fruit phenotype observed is parthenocarpy, seedless/pseudoembryos, size and shape similar to weight in tomato, and *A. thaliana*.
- ***SlARF7* gene:** Its function is "auxin signaling" and genetic modification of "RNAi-mediated silencing, constitutive promoter". The fruit phenotype observed without any vegetative alterations is parthenocarpy, seedless, altered shape in tomato.
- ***SlAUCSIA* gene:** Its function is auxin response and genetic modification of "RNAi-mediated silencing, phloem-specific promoter". The fruit phenotype observed with vegetative alterations is facultative and obligate parthenocarpy, seedless, reduced size in tomato.

- ***SlChs* gene:** Its function is flavonoid biosynthesis (auxin transport) and genetic modification of "RNAi-mediated silencing, constitutive promoter ". The fruit phenotype observed without any vegetative alterations is parthenocarpy, seedless, altered size, shape, and color in tomato.
- ***SlDELLA* gene:** Its function is Gibberellin signaling and genetic modification of "Antisense downregulation, constitutive promoter". The fruit phenotype observed with vegetative alterations is facultative parthenocarpy, seedless, reduced size, altered morphology in tomato.
- ***SlTPR1* gene**: Ethylene signaling and genetic modification of "Overexpression". The fruit phenotype observed with vegetative alterations is parthenocarpy, seedless, altered morphology in tomato.
- The data on genes related to seedlessness indicated down-regulation of biosynthesis genes and ethylene signal transduction pathway genes after fruit initiation which is related to the release of the dormant state of the ovary. The function of ethylene in this respect is antagonistic to that of auxin and gibberellin in that it would prevent ovary growth before pollination while on the otherhand enhanced ethylene response leads to parthenocarpic fruit development [22].

Hybrid seed production using male-sterility in tropical fruit crops

Natural and artificial induction methods of male sterility were in practice by plant breeders to produce hybrid seeds based on the purpose or economic importance of the product. Identification of natural mutants from a random population or progeny of a breeding line will take more than 12 years. Mutagenic agents and chemical hybridizing agents (CHAs) were the artificial means of inducing male sterility. The development of hybrids depend on the kind of mutagen, crop, time and rate (dosage) of the application when using CHAs like maleic hydrazide (MH), alpha naphthalene acetic acid (NAA), beta indole acetic acid (IAA), the gamma rays and ethidium bromide (EB). These are the best effective mutagenic agents to induce genetic male sterility, though their effectiveness vary due to crop by mutagen by application rate and their interaction effects.

Mechanisms employed in the induction of male-sterility in fruit crops

Mutation of homozygous fertility restoring nuclear gene (MsMs) result in heterozygous (Msms) individuals and help to identify the genotypes carrying recessive alleles (Msms or msms) to have male-sterile forms from progeny with homozygous genic (msms) male sterility, while mutation of mitochondrial genome result in cytoplasmic male sterility (cms). The cms is the most useful technique for those fruit crops where seed is not the main product, but cms needs male hybrid parent as maintainer line that produces heterotic hybrid progenies [11].

The cytoplasmic-genic male sterility (c-GMS) is the mechanism where both mitochondrial and nuclear genes involved in inducing male sterility. The sterility or fertility of different restorer lines/genes is affected by photoperiod, temperature or both, light illumination and soil nutrient stress. The c-GMS is controlled mostly at the molecular level and by mitochondrial genes. To create male sterile lines, interspecific hybridizations are the best choice to have both sterile and maintainer lines. The genomic studies on the mitochondrial genome of pigeon pea A_4 cytoplasm recognized 13 ORFs which can trigger ms [30] though the reversion of sterility is influenced by cytoplasmic genes only, but not nuclear genes [11]. The aberrant recombinations in cms associated mitochondrial genome resulted in the production of novel chimeric polypeptides specific to cms line and nuclear maintainer lines. These were identified in several field crops for hybrid seed production [31].

The cms in crop plants is determined by the mitochondrial genome. Mitochondira is the site of essential metabolic pathways. Therefore, cms is associated with a pollen sterility phenotype that can be suppressed or counteracted by nuclear genes known as fertility restorer (fr) genes. The mitochondrial signaling pathways, including those involved in regulating cell death and nuclear gene expression, are controlled by nuclear genes that restore fertility (fr). It was identified that genes encoding pentatricopeptide-repeat proteins are the key regulators of plant mitochondrial gene expression [19].

The use of gms for F_1 hybrid seed production first used in horticultural crops to improve vegetable or fruit size, seedlessness, and yield. Little advancement today, male-sterile 'Satsuma' cytoplasm was transferred into seeded cultivars of citrus by producing cytoplasmic hybrids or somatic hybrids (cybrids in Pumelo) by symmetrical fusion between protoplasts derived from embryogenic callus and mesophyll tissues have the nuclear genome of the mesophyll parent and the mitochondrial genome of the callus parent [15]. The majority of seedless fruits were benefitted from parthenocarpy induced naturally or spontaneously or by chemicals during anthesis but only a few crops were benefitted from cms and nuclear-mitochondrial gene interactions. A combination of both is viable mostly for mandarin cultivars (Table 1).

Crops and cultivars	Improved variability in qualitative and quantitative traits of economic importance in horticultural crops	F_1 Hybrids with seedlessness (mechanism of cytoplasmic male-sterility and fertility restoration, parthenocarpy)
Vegetable crops: 1) Onion 2) Tomato 3) Cucumbers 4) Chilies (Capsicum) 5) Field beans	Vegetable crops: 1) bulb size, quality and yield 2) fruit size, seedlessness 3) no viable seed 4) color, size, shape, yield 5) flower color, yield	1) T-cytoplasm and genic male sterility for hybrid onion production 2) cgms for F_1 hybrid tomatoes and independent mutations-*pat*, *pat*-2 and *pat*-3/*pat*-4 genes induce facultative parthenocarpy by increasing active GA in 13-hydroxylation pathway for seedless fruits of tomato. 3) Cucumber with seedlike structures but lack embryo and endosperm 4) c-gms type of male sterility 5) Cytoplasmic suppression of expression of gms
Field crops: 1) Rice 2) Maize 3) Rapeseed or Canola (*Brassica napus*) cv. Polima, F_1 from *Brassica rapa* cv. Shogoin-kabu x *Raphanus sativus*)	Field crops: 1) Accelerate or delay panicle differentiation or heading depending on the photoperiod length and temperature and determines if the pollen is sterile or fertile. The photoperiod sensitive rice can propagate by itself under short-day conditions. 2) Multiple color cob 3) high seed and oil yield	1) Photoperiod sensitive recessive male steriles to produce hybrid rice. Long days induce sterility. 2) gcms- T, S and C-cytoplasm depending on the type of fertility restorer genes 3) Pol-cytoplasm, dominant male sterile for SI family recurrent selection programs; polyploidy
Flower crops: 1) Petunia 2) Lillies	Flower crops: 1) Flower 2) size	1) Cytoplasmic male-sterility and mitochondrial mutants (Hanson,1991) 2) Self-incompatibility; Nitrous oxide to induce polyploidy
Fruit crops: 1) Watermelon Varieties are Tetra 1, Tetra 2 and Tetra 3, W872 and W1068A and also serve as parents for seedless watermelon production	Fruit crops: seedlessness, size, texture, sugar/TSS, tough rind and high keeping quality; Triploid (3x) watermelons were distinguished from tetraploids by the presence of triangular (lobed) shape when viewed from the blossom end.	1) Watermelon: ms, triploidy and facultative parthenocarpy; Triploid seedless F_1 hybrid seed of watermelons produced in 1962 by crossing diploids (VBL 59-1 and VBL 59-6) with tetraploids (Tetra 1-3)
2) Citrus Few cultivars {e.g., Satsuma mandarin (*Citrus unshiu* Marc.), Washington Navel orange (*C. sinensis*), Tahiti lime [*Citrus latifolia* (Yu. Tanaka) Tanaka], and Oroblanco (*C. grandis* X *C. paradisi*)}; Mandarin seedless cultivars "Orri-seedless Orah. and Moria- seedless Murcott"; Seedless cybrid pumelo, and grapefruit; A sweet self-incompatible seedless mandarin cv."Clemenules".	Mandarins, Pumelo, Oranges, limes with variability in fruit color, size, shape, seedlessness, yield	2) Citrus: Satsuma mandarin with cms (Satsuma), c-GMS, cybrids by protoplast fusion of ms lines, gamma irradiation, budmutation-grafting, female sterility, obligatory parthenocarpy. Washington Navel with female and male sterility, obligatory parthenocarpy. Navel orange with genetically controlled degradation of pollen mother cells during the first meiotic division. Tahiti and Orblaco with parthenocarpy, chromosomal irregularity during meiosis. Wilking mandarin with a recessive gene to induce asynapsis during meiosis resulting in pollen and ovule abortion. Clementine with stimulative parthenocarpy when SI lines grown in isolation (Vardi., *et al*. 2008) Pumelo with triploidy from interspecific crosses

3) Sugar apple- Thai seedless (Ts)	Seedlessness	3) Sugar apple: spontaneous mutation
4) Grapes cv. Corinth cv. Thomson seedless cv. Talisman is a cross between Frumoasa albe and Vostorg.	Seedless small berries due to parthenocarpy and large size berries from stenospermy and GA application	4) Grape: with stenospermy while Cv. Corinth with facultative parthenocarpy. cv. Thompson seedless seed abortion after fertilization cv. Talisman produces parthenocarpic fruits if isolated before anthesis and contains only female flowers, has stenospermy and parthenocarpy.
5) Banana	Seedlessness, size, shape and length	5) Banana: with triploidy and facultative parthenocarpy (parthenocarpy gene, *Pi*)
6) Pomegranate - Seedless/soft-seeded: cv. Jalore Seedless cv. Ever sweet Soft seeded or seedless cultivars from India include 'Bedana', 'Kandhari', 'Alandi' 'Dholka', 'Kabul', 'Muscat Red', 'Paper Shell', 'Poona', 'Spanish Ruby', 'Vellodu', 'Muscat White' Medium to hard seeded: Ambrosia, Wonderful, Redskin, Grenada and Kashmir and cultivars from Turkey, Spain, Israel, Iran and Middle East countries.	Sweet and seedless, sweet-sour and sour, early, mid season and late, juicy and table fruit, soft-seeded and hard-seeded or major and minor with various skin colors red, yellow or brick pale red.	6) Pomegranate: seedless or soft seeded fruits selected by recurrent selection and observed with facultative parthenocarpy and apomixis; Spontaneous mutation during fruitset produced fruits lacking viable seed.
7) Pear	lack seed or no viable seed	7) Pear with parthenocarpy
8) Pineapple 9) Strawberry	8) lack seed or no viable seed 9) fruit size, shape, sweetness	8) facultative parthenocarpy 9) Polyploidy
10) Apple	lack seed or no viable seed	10) facultative parthenocarpy
11) Peach, Apricot, Nectarine, Plum, Cherry, Litchi, Loquat	no data available	11) No data available
12) Kiwi, fig, blackberry	lack seed or no viable seed	12) Parthenocarpy in natural open populations of these three crops. Male sterility in Kiwi.

Table 1: Variability in qualitative and quantitative traits of F_1 hybrids in horticulture and few field crops produced using male-sterility or parthenocarpy.

Source: [6,15] Note: In all the crops where cms or cgms are used for F_1 production, male fertile nuclear restorers were used to produce seed.

Therefore, the review presented is mostly in support of work done in fruit crops of citrus species where a lot of research has done in hybrid seed production with improved quality. This might benefit future fruit crop researchers. The research that was done in the past in the vast majority of horticultural and field crops using other methods for induction of seedlessness or parthenocarpy was also included as necessary for the conceptual understanding of the subject matter on ms benefits (Table 1 and 2).

Tropical fruit crops identified with male-sterility for commercial cultivation

Selected varieties of watermelons, grapes, citrus, pineapples and bananas are some examples of commercial table purpose fruit-crops where seedlessness is frequently used due to polyploidy. Hybrid triploid watermelons, commonly called seedless watermelons, are more difficult to grow than diploid varieties. However, they can be cultivated successfully with some extra-care and management by planting diploids as pollinizers.

There were very limited reports on fruit crops identified with ms or utilization of ms or self-incompatibility for the production of edible fruit crops. Mandarin is the most benefitted crop from ms. There are tamarind clones (NBN1-3, RDB patna and JRK) in India identified with variability in pollen sterility with values ranging from 0.88 percent to 1.38 percent. The natural fruit set is very low (3-5%) compared to 70-90 percent in controlled conditions and honeybees are the major pollinators during the peak season of flowering between May and July [32]. Major fruit crops benefitted from three mechanisms of male sterility were presented (Table 2).

Name of the edible fruit crop	Mechanism of male-sterility or methods employed for a specific trait	Hybrids produced and methods of seed multiplication	Parents of hybrids	Qualitative and quantitative traits that were improved
Meiguicheng orange (*Citrus sinensis*) (Source: 21., *et al.* 2017 a)	Radiation-induced chromosomal aberrations: Female sterility in seedless mutant induced by ^{60}Co gamma rays, causing abortion of megagametophyte during megasporogenesis and early megagametogenesis	Fruits developed from the flowers of cross-pollinated with wild type pollen still produced few seeds with small nucellar embryos inside ovules of seedless mutant.	Polymebryonic Meiguicheng orange (Seeded) as a female X Seedless mutant as a male parent, created from Meiguicheng by gamma rays. Both are diploid.	A diploid cultivar with female sterility, seedlessness in fruits, less number of hybrid seeds from mutant seedless cultivar, no change in flower structure of both parent and improved cultivar.
Polymebryonic acid citrus fruit -Grapefruit (*Citrus Paradisi*) produced by crossing Sweet orange (*Citrus sinesis*) and pumelo (*Citrus maxima*) (Source: [33].	Cytoplasmic-genetic male sterility (c-GMS)	Hybrids of cross "Hy16 X Foster Pink" grapefruit produced 53 male fertile and 48 male sterile seedlings	"Fosterpink" (*Citrus paradisi*), HY16 (*Citrus hanaju* X *Citrus junos*) and "Kiyomi" (*Citrus unshiu* X *Citrus sinensis*) tangerine are used as seed parents to ms genotypes for seedlessness.	Precocious flowering in nucellar and zygotic seedlings of citrus species to determine ms in a shorter period of time. Seedless hybrid fruits.
Mandarin [25]	cytoplasmic male-sterility	Protoplast fusion of nuclear genome and chloroplast DNA from leaf parent {Hirado Buntan pumelo (*C. grandis* L. Osbeck) (HBP)} and mitochondrial genome of callus parent {Satsuma cultivar Guoqing No.1 (G1)}.	Male-sterile hybrid line G1+ HBP	hybrid is seedless and male-sterile. Seedlessness achieved due to severe degeneration of stamen, petals and aborted pollen grains inside anther
HB Pumelo (*Citrus grandis* L) [34]	cytoplasmic male sterility	Male-sterile cybrid (G1+HBP) pumelo. Multiplied from somatic fusion between embryonic callus protoplasts of G1 (*Citrus unshiu* Marc.cv.Guoqing No.1) and mesophyll protoplasts of HBP {Hirado Buntan pumelo (*C.grandis* L.Osbeck)}.	This diploid cybrid (G+HBP) was regenerated with nuclear genome and chloroplast DNA from leaf parent HBP (a seedy variety with attractive fruit quality) and mitochondrial genome from callus parent G1 (Satsuma mandarin cv.Guoqing No.1)	This ms diploid pumelo hybrid is seedless cultivar with foreign mitochondrial genome, a variant of male fertile pumelo. Reduced petal development in size and width, retarded stamen primordia development, disorganized cell proliferation in stamen like structures fused to petals or carpel. Alterations in mitochondrial DNA or gene expression resulting in altered nuclear gene expression in cms lines.

Annona fruit -Family: *Annonaceae*[16]	Mutation - ovules (seed precursors) of the mutant lacking the "*ino"* locus coding for outer of the two normal integuments of the ovary	Thai seedless (Ts)	spontaneous seedless mutants of Annona squamosa	Useful agronomic trait -Seedless fruits in sugar apple; Conservation of gene locus"ino" for outer integument specific gene expression and regulating the ovule development between eudicots and more ancient lineages of angiosperms.
Fruit crops [15]	1) Induced mutations using fast neutrons, x rays or gamma rays 2) Buds of old and newly released high-quality selections of citrus cultivars were irradiated with different doses of ^{60}Co 3) Budwoods were gamma-irradiated with 13.3 Gy/min ^{60}Co and grafted onto "Troyer" citrange 4) Ploidy manipulation by crossing diploids with tetraploids	1) Apple, peach, grape, pear, sweet cherry, citrus 2) Citrus 3) "Troyer" citrange (*C. sinensis* x *Poncirus trifoliate*) Mandarin seedless cultivars developed by bud wood irradiation are Orri-seedless Orah. and Moria-seedless Murcott. 4) Oroblanco and Melogold; Winola	1) Induced mutations of these fruit crop species or hybrids 2) Four different citrus cultivars 3) "Troyer" citrange rootstocks were budded with cobalt treated individual buds to form the first generation of mutation vegetative shoot(mV1). The buds of mV1 were grafted to new Troyer citrange rootstock to form mV2 generation to avoid chimeras and select seedless fruits by counting seeds during the second or third flowering season of mV2 generation. 4) *C. grandis x C. paradisi*	1) compact tree growth, self-incompatibility, improved fruit color, low acidity, seedlessness and disease and pest resistance 2) complete seedlessness and few numbers of seeds 3) seedlessness, pollen sterility and avoided the formation of chimeras. This technique reduced the time for selection of seedlessness to less than one year (takes time from march to November) 4) Seedless triploid fruits

Table 2: Edible seedless fruit crops and cultivars identified with male-sterility.

Source: [15,16,21,25,33,34].

Therefore, among horticultural crops with cultivars that were improved using the techniques of ms: 1) Seedless citrus- Mandarin cv. Satsuma (*Citrus clementine*) and cv. Clemenules 2) Seedless custard apple (*Annona squamosa*) cv. Brazilian seedless and Thai seedless, 3) Seedless kiwi (*Actinidia arguta* cv *Issai)*, 4) Triploid seedless watermelon (*Citrullus lanatus*) and seedless cucumber (*Cucumis sativus*), 5) Seedless parthenocarpic fruits in citrus cultivars, pineapple and banana (*Musa acuminata banksia*) 6) Stenospermic- seedless grapes and watermelons 7) male-sterile Mandarin cv. Satsuma hybrids and seedless mutants or mutant grafts in pumelo, grapefruit, and orange 8) seedless facultative parthenocarpic fruits in grapes, and citrus 9) Seedless fruits from clonally propagated in pomegranate (*Punica granatum*) cv. Jalore seedless/soft-seeded and cv. Eversweet are available for table purpose fresh fruit consumption use and were all benefitted as of today due to seedlessness in fruits (Figure 1).

Molecular markers identified for male sterility

Genetically induced male-sterile plants and their restorer lines were produced using recombinant DNA technology and asexual recombination using somatic hybridization and transformation. The regenerated plants derived from cultivar G_1 (callus parent with cms) produced both diploid and tetraploids and were analyzed by 23 simple sequence repeat markers (SSR) markers to map citrus genome [11]. In grapefruit, five random amplified polymorphic DNA (RAPD) markers amplified in most of the male-fertile individuals and 'Foster Pink' while two RAPD markers were found in most of the male-sterile individuals and 'Foster Pink'. The linkage analysis showed that the two and five RAPD markers flanked with R_1 or R_3 gene with the closest distance of 15.5 cM and 8.4 cM respectively in grapefruit [33]. The same authors also reported that a dominant nuclear fertility-restorer gene system comprising one epistatic gene R_1, and two complementary genes R_2 and R_3 controls restoration of male fertility in acid citrus plants with sterile cytoplasm (S). R_1 acts on the perfect restoration of fertility, whereas the complementary genes R_2 and R_3 act on the restoration of anther development and pollen formation. Information on segregation of "ms and mf" hybrid citrus seedlings derived from seven crosses (Table 3) indicates that HY16 and Kiyomi are ms genotypes and generated only ms seedlings when crossed with Foster Pink or three orange cultivars (Ruby Blood, Shamouti orange and Trovita) and one acid citrus cultivar Sudhachi. This proves that cytoplasm of the seed parent is sterile and pollen parents have heterozygous genotypes for the male-fertility restoration.

Cross (msxmf)	Number of Seedlings sowed	Number of seedlings with flowers (mf:ms)	Precocious flowering (%)
HY16 x Foster Pink	1119	158 (80:78)	14.1
HY16G2 x Foster Pink	324	55 (12:43)	17.0
HY 16 x Ruby Blood	31	7 (5:2)	22.6
HY16 x Shamouti	422	2 (0:2)	0.5
HY16 x Sudachi	102	1 (0:1)	1.0
HY16 x Trovita	3588	24 (11:13)	0.7
Kiyomi x Foster Pink	5901	175 (134:41)	3.0

Table 3: Segregation of male-sterile (ms) and male-fertile (mf) hybrid citrus seedlings derived from seven crosses.

Source: [33].

In Satsuma mandarin (*Citrus unshiu*), QTL mapping of progeny from cross Okitsu No.46 and Okitsu No.56, the QTL marker for the number of pollen grains per anther (*MS_P1*) and apparent pollen fertility (*MS-F1*) on linkage groups 8 and 6 respectively as an index of ms. Further studies on the cross "Okitsu No.46 X Kara (O46-K)" confirmed the role of QTL loci *MS-P1* in reducing the number of pollen grains and *MS-F1* in decreasing apparent pollen fertility. These loci (nuclear genes) were derived from Kunenbo (*Citrus nobilis*) through "hassaku (*Citrus hassaku*)" and "Sweet Spring", while the male-sterile cytoplasm of "Sweet Spring" and "Okitsu No.46" was contributed by Kishu (*Citrus kinokuni* hort. ex Tanaka) [25].

The self-compatible cultivars grapefruit and "Dancy" tangerine carry self-incompatible gene useful to study inheritance pattern of self-incompatibility to use in seedless cultivar development. There was very little known information on the inheritance of female sterility, although there was a close relationship observed between seedlessness and female sterility [28].

In grapes, for seedlessness, a dominant gene "I" was identified regulating three independently inherited complementary recessive genes a1, a2 and a3 and contributing stenospermocarpy as confirmed by RAPD and sequence characterized amplified region (SCAR) markers [35].

Morphological markers identified for male-sterility

The morphological traits that might indicate inherent gms associated with nuclear controlled recessive (msms) alleles are pigmentation in the stem, translucent anthers, sparse podding and delayed flowering. But, this mostly benefitted self-pollinated crops and field crops where the hybrid seed is required to produce. Further, evolution has a lot of impact on the development of seedlessness in fruit crops over time due to natural mutation of the original genome in a fruit crop. A unique outer integument is one synapomorphy of angiosperms separating them from other extinct more ancient lineages of seed plants by regulating the development of ovule in eudicots. A candidate gene approach framed the molecular basis for seedlessness in mutant "Thai seedless" and disruption of the conserved genetic pathway in angiosperm ovule development [16].

Homeotic conversions of third whorl floral organs in somatic hybrid (G1+HBP) leading to abnormal floral morphology was observed in cms lines of citrus. The morphology of flower buds in G1+HBP was similar to that of HBP during early stages of floral development, but later phenotypic differences were observed during early stages of stamen differentiation by retarded development of stamen primordia with few distorted stamen-like structures fused to petals to carpels with reduced petals in size and width. In this floral mutant phenotype with genes *ap3* and *pi* with foreign mitochondria were controlled by *MADBOX genes* resulting in ms and lead to the conversion of stamens to carpel like structures and replacement of petals with sepals [34]. The grapefruit seedlings with a high rate of precocious flowering within several months after germination unlike the usual method of more than five years and were used to determine the ms in a shorter period of time. Nucellar seedlings of grapefruit and zygotic seedlings from some citrus species and cultivars often form a flower in the shoot terminal within several months after germination and this temporary phenomenon is called precocious flowering [33].

The production of *A. squamosa* fruits of normal shape and size, the peculiar structure of the ovule, the occurrence of fertilization and embryo degeneration were notable morphological features along with sterile seeds in a seedless fruit (Fig.1). Thus, in custard apple only after fertilization fruit development occurred and the sterile seeds promoted the physiological growth of fruit [36].

Gamma radiation has been widely used in citrus breeding to create diploid seedless cultivar. Gamma radiation mutagenesis created chromosomal aberrations that induced seedlessness to reduce pollen viability in the mutant of genus citrus, "Meiguicheng" cultivar of orange (*Citrus sinensis*). Inter crossing between seedless mutant and its wild type revealed efficient fertilization by mutant pollen grains and thus male fertile mutant was used in hybridization breeding programs [24].

Thus, the two main mechanisms (genic or cytoplasmic-genic) of male sterility while understanding their physiological factors of control in phenotypic expression will help to develop DNA markers as well as morphological markers for breeding fruit crops with seedlessness.

A summary of distinguishable traits identified as markers in the genic, cytoplasmic and gene-cytoplasmic male sterile lines in horticulture crop species

Morphological and physiological features including growth: Phenotypic markers

1) Leisons in mitochondria or chloroplast of cms lines of tobacco; 2) Vacuolated cytoplasm during meiosis in cms line of Petunia 3) Embryo degeneration and sterile seeds in seedless fruit of *A. squamosa* 4) Stenospermy in watermelons, grapes and sugar apple 4) Degenerated flower organs like stamens and petals with aborted pollen grains in cybrids produced from cms cultivars of mandarin and pumelo. Because of nuclear-mitochondrial incompatibility, cms lines exhibited developmental defects in male gametophyte and or floral organs. 5) Polymebryonic acid citrus fruit, grapefruit with c-GMS were observed with precocious flowering and zygotic seedlings to ms in short span of time 6) Triangular (lobed) shape in seedless triploid watermelon when viewed from the blossom end. Seedless watermelons have tough rind to resist shipment and semiseedlessness or no seed content will keep the fruits from overripening.

Biochemical features including enzymes and proteins

Barnese-Barster system in tomato, tobacco. 2) Gibberellins (GA) regulated flower organ development by antagonizing the function of *DELLA* proteins and partially activating the expression of floral homeotic genes ap3 and pi. GA has no role in floral organ differentiation but essential for normal growth and development of the floral organs. GA levels regulate the expression of key enzyme "GA-20 oxidase" of the final step in the biosynthetic pathway of bioactive GAs (GA_1 in citrus). The repression of GA-20 oxidase was observed in G1+HBP whose expression is required for elongation or vegetative growth and floral bud formation. In pumelo, this enzyme was at very low concentration in reproductive organs compared to vegetative shoots confirming its role in stamen development of citrus fruits as similar to jasmonates (JA) to promote expression of R2R3MYP transcription factor (MYB108, MYB21, MYB24) for stamen development and maturation. In citrus cybrid G1+HBP, expression of MYB108 is necessary, but its activated expression profile might contribute to retarded stamen development.

Molecular features including genetic and genomic markers

1) Expression of TA-29 RNAse genes-Barnase and T_1 facilitated by 5' region of tapetum -specific gene along with activation of GUS marker gene. 2) In citrus, ms genotype was estimated to be (S)r1r1R2r2r3r3 for male sterile 'HY16', (S)r1r1r2r2R3r3 for male sterile 'Kiyomi' and (F)r1r1R2R2R3r3 or (F)R1r1R2R2r3r3 for male fertile 'Foster Pink' grapefruit. Here, ms is a recessive character; r2 and r3 are complementary genes with upper stream expression of r1, and r1 is an epistatic gene to r2 and r3 genes. Five markers identified flanked with R1 or R3 gene with the closest distance of 8.4 cM in grapefruit and the two markers flanked with r1 or r3 gene with the closest distance of 15.5 cM in grapefruit. 3) Absence of "*ino*" (inner no outer) gene locus from seedless mutant variety Ts (Thai seedless), but its presence is noticed in wild type *A. squamosa* and other species. 4) The citrus class-B MADS-box genes responsible for cms retrograde signalling and a disfunctional mitochondria in cybird pumelo. This lead to reduced expression of PISTILLATA (pi) in stamen like structures of cybrid pumelo which was correlated with down regulated expression of APETALA3(ap3). Here cms resulted in disturbed mitochondrial-nuclear gene interactions where as pi and ap3 transcripts encoded key transcription factors for stamen identification.

Barriers for success when producing male-sterile (female), male-fertile, and restorer (maintainer) lines

Interspecific hybridization is a challenging task, due to frequent occurance of reproductive barriers either pre- (before fertilization) and post- zygotic (after fertilization) origin, to maintain genetic integrity or to prevent gene flow. The natural habitat (geographic isolation), flower structure (i.e., shape or color), flowering time or pollen mediator are prezygotic barriers where as interspecific incompatibility due to inhibition of pollen tube germination or growth, pollen germination, embryo abortion, abnormal growth of endosperm, and hybrid sterility (cms) caused by epistatic (nuclear or mitochondrial) gene interactions are post-zygotic barriers [37]. Understanding the molecular mechanisms of reproductive barriers expands the possibility of hybridization in fruit crops.

The pre-zygotic interspecific barriers can be overcome in *Brassicaceae* by bud pollination (pollinating stigmas 2-3 days before anthesis) while in *Liliaceae* by cut-style pollination (pollinating to a cut stigma or style) or heating pistils and pollen. The post-zygotic embryo abortion during interspecific crossing can be overcome by embryo-rescue techniques like ovary ovule or embryo culture. The hybrid sterility, due to reduced chromosomal pairing during meiosis, can be restored by zygotic or somatic chromosomal doubling through cell cycle disruption by antimitotic agents such as colchicine, oryzalin, trifluralin or nitrous oxide (N_2O). All these techniques are useful for producing hybrid seeds by overcoming interspecific reproductive barriers [37].

Among diploid, triploid and tetraploid plants, only triploid plants were observed with high percentage of survival, high number of seedless fruits and this predominance of triploidy relates to a remnant of autosterility in tetraploids with respect to seedlessness [38].

Conditions of crop growth that pose risks in the development ms lines

High or low temperature causing hybrid necrosis was reported in both intra- and inter-specific hybrids of vegetable crops and was caused by the interaction between alleles at two different loci of two chromosomes or two different arms of the same chromosome. Using genome imprinting and high throughput sequencing techniques, the endosperm abnormalities and associated epigenetic gene interactions can be revealed to understand the disturbances of the genome balance that occured either due to maternal or nuclear gene component.

The genomic disturbance that occur after allopolyploidization might be i) *major* by elimination, disjunction or chromosomal doubling or ii) *minor* by deletion or inclusion of DNA sequence, gene conversion and rDNA (ribosomal DNA) loci exchanges. These disturbances occur both in distantly as well as closely related hybrids. Chromosomally distant species hybrids generally show sterility due to chromosomal rearrangement on meiotic pairing and thus form bivalents or multivalents for novel genetic exchanges between species. Bivalent formation was observed when allopolyploid hybrids were generated in banana *Musa accuminata* X *Musa bulbisiana* [39]. These transcriptional chromosomal changes may result in changes in gene expression affecting the phenotype of the hybrid plant.

A candidate gene approach by deletion of "ino" locus in Thai seedless (Ts) mutant revealed the apparent molecular basis of a useful agronomic trait "seedless fruit" in a crop species like sugar apple (*Annona squamosa* L. a member of magnoliids). This indicate conservation of the role of a critical regulator of ovule development between eudicots (*Arabidopsis thaliana* L.) and more ancient lineages of angiosperms (magnoliids), as the Ts mutant lacks the outer two normal integuments of the ovule which are precursors for seed development. In *Annona*, outer integument is supporting the embryosac and embryo development without which the seed development is impossible. The region coding for deleted "*ino"* locus was "s" not estimated yet. Pollen from Ts plants was fully fertile when used to pollinate wild type Annona cherimoya, demonstrated that infertility did not result from polyploidy or from other severe chromosomal aberrations [16].

Method of seedlessness obtained in grapes by using gibberellic acid sprays result in increased number of berries, improved size of berry with reduced or no seed, and elongated clusters. Here, the success in achieving seedlessness is mainly dependant on weather, labour intensive as well as labour expensive factors besides leaving potentially hazardous synthetic chemicals into human diet [35].

Crop physiological or biochemical processes hindering the ms induction

The tapetal cell differentiation and or function is essential for pollen development. The cytoplasmic or nuclear mutations may prevent normal pollen development resulting in male sterility. Though this ms is useful in hybrid seed production, but it is limited to crops or lines that can restore fertility and in crops where emasculation and pollination is possible.

Transcriptional events during anther development may activate unique gene sets within tapetum thereby regulating gene expression of anther's tapetal cells. These tapetal cells surrounding the pollen-sac can be destroyed by expression of two kinds of chimaeric robonuclease genes viz., TA-29 RNAse genes-Barnase and T_1. The expression of these two different RNAse genes can be facilitated by 5' region of tapetum-specific gene along with activation of GUS (beta-gucuronidase) marker gene. This leads to prevention of pollen formation resulting in ms. The function of these genes is similar to those in self-incompatable lines. It is beneficial only in nonfruiting type crop plants such as leafy vegetables (cabbage) where fertility restoration is not required. But in fruiting type crops like tomato and other fruit crops where complete and true fertility restoration is required, antisense RNA technology and existence of Barstar- a protein inhibitor of Barnase, are essential for fertility restoration in ms lines [10].

A diploid plant that carries ms cytoplasm and is heterozygous for a resotorer will produce two classes of pollen grains, those that carry the restorer and those that do not. In genetic terms, there is a lot of diversity between and within plant species for restoration of fertility either by gene action prior to meiosis in sprophytic tissues (sporophytic restoration system) or after meiosis in gametophytic tissues (gametophtic restoration system) of microspores or pollen grains. In case of sporophytic restorer, both genotypic classes of gametes will be functional. In case of heterozygous plants for gametophytic restorer (Eg.S-cytoplasm of Maize), only those gametes that carry the restorer will be functional. Further, the number of restorer genes may also vary with the crop, though a single restorer gene is the most common. For example, two unlinked restorers are required for T-cytoplasm of onion to restore fertility while duplicate restorers with overlapping functions are common in Phaseolus indicating multiple mechanisms associated with fertility restoration. In Phaseolus, only in presence of nuclear fertility restorer gene (Fr) deletion of cms-gene (pvs) or physical loss of cms associated gene from the mitochondrial genome is possible [8].

Fertility-restoring genes reduced the sterility-causing proteins by 80%, because fertility restoration is associated with genes encoding pentatricopeptide repeat proteins [11]. Female fertility and sterility in citrus varies with the cultivars and pollination methods while seediness varies with time and planting area as influenced genetic and environemntal factor interactions. The female fertility and sterility are heritable characters. The cultivars Okitsu wase, Shirayanagi Navel are both male and female sterile while T132 Tankan is female sterile, Kiyomi is completely male sterile. The degree of female fertility in Kiyomi increases when cross pollinated by male fertile cutlivars. Therefore, female sterility coupled with ms or self-incompatibility is essential for development of seedless cultivars in citrus group [28].

Formation of multivalents and univalents sometimes cause sterility when transfering genes from wild relative to cultivated tomato. It was observed during first generation of fusion hybrids with low to moderate levels of pollen fertility in tetraploids and with little or no pollen viability in hexaploids. This might be due to precocious cell division of microspores, laggard chromosomes due to formation of bridges during anaphase I and II and multivalent configurations in diakinesis [40].

Auxin signal induced by pollination in fruit crops leads to phosphorylation and inhibition of aux/iaa complex protein involved in fruit development process. In mutant arf8 seedless progeny, this complex formation is completely absent leading to parthenocarpy [15].

For development of fruits, fruit number and fruit weight, availability of nitrogen (N), potassium (K), calcium (Ca) and magnesium (Mg) are required during all the growing months. For example leaf potassium is essential for flowerbud initiation and for transport of metabolites to sink (fruit) while Zinc (Zn) has negative effects on fruit yield and fruit weight in Mango [41]. Thus mineral nutrients had essential roles before, during and after fruit set in fruit crops. An imbalance in mineral availability during the period from flower bud initiation to matured fruit may result in hormonal imbalance or less quality seedless fruits.

Advanced techniques competitive in place of male-sterility to produce uniform hybrid fruits with best fruit quality and productivity

Tissue culture

Genetic improvement of fruit crops using tissue culture was successful in shoot tips as explants in datepalm (*Phoenix dactylifera* L.), and fig (*Ficus carica* L.), epicotyle in rough lemon, nodal segment in aonla and jackfruit, leaf, nodal, hypocotyle and cotyledon segments in pomegranate. Usage of Murashig and skoog (MS), NLN and woody plant media (WPM) were in practice for best vegetative growth of explants in fruit crops though differences in callus formation and growth were observed due to physiological state of explant and quality of media [42].

Somatic hybridization by protoplast fusion was used in transgenic citrus plants expressing green floursecent protein (*gfp* gene) as parent material for complete regeneration of hybrid products and overcoming the traditional breeding problems as well as the reproductive barriers among species and genera [43].

Cybrids are the cytoplasmic hybrids possesing the nucleus of one species and the mitochondria and/or chloroplast of another species formed by somatic fusion [44]. These are the best choice in offering improved quality of citrus and uses a non-GMO technology. The mt DNA of cultivar G_1 (Guoping No.1) of Satsuma mandarin (*Citrus unshiu* Marc.) was successfully introduced into the three seedy sweet orange cultivars (Early gold, Taoye and Hongjiang) for potential seedlessness via symmetric fusion (Figure 2) [11]. This non-GMO technology can be adopted for potential seedlessness in Custard Apple, Moringa, Sapota, Tamarind fruits and other unexplored tropical fruit crops.

Figure 2: The strategy to create cybrid by transfering Satsuma CMS to seedy varieties by symmetric fusion. (Source: Courtosy of [11])

New grapefruit cultivars with enhanced canker resistance were generated from several putative cybrids that were created by protoplast fusion of embryogenic suspension culture-derived protoplasts of canker resistant 'Meiwa' kumquat (*Citrus japonica* Thunb), with mesophyll-derived protoplasts of three grapefruit (*Citrus paradisi* Macfad.) cultivars namely 'Marsh,' 'Flame,' and 'N11-11' somaclone of 'Ruby Red'. All the cybrids had the mitochondrial genome of kumquat and most had the chloroplast genome of kumquat revealing the valuable role of cytoplasmic organelles to plant disease resistance [45].

Advanced techniques of symmetric somatic hybridization, haploid-diploid fusion, targeted cybridization to transfer cms (mtCMS) from Satsuma mandarin, and triploidy via interploid crosses using somatic hybrid allotetraploid breeding parents were employed for scion improvement of citrus and for development of seedless fresh fruit varieties. Similarly, rootstock somatic hybridization were used for protecting improved gene combinations, improving disease and insect resistance, soil adaptation and tree size control. Rootstock breeding and selection at the tetraploid level using somatic hybrid parents and resynthesis of important rootstocks at the tetraploid level via fusion of selected superior parents was the best in citrus rootstock improvement [46,47].

Aspermia in natural mutants, 'Brazilian seedless' and "Thai seedless", is of the stenospermy type controlled by single recessive '*ino*' gene resulted in loss of the outer integument of ovule which affects seed development in *Annona squamosa* L. resulting in seedless fruits (Figure 3) [36]. Application of GA (1500ppm) during flowering enhanced fruitset resulting fruits with larger size and seedlessness. The gibberellic acid stimulated the fruit growth in all dimension and fresh weight when applied on tree fruit while when it is applied on ovaries lead to the formation of parthenocarpy without pollination and fertlisation [48].

Figure 3: Longitudinal sections of Annona squamosa fruits from normal (left) and aspermic mutant accession, with the location of sterile seeds marked (arrows). se, seed. Scale bar: 3 cm. (Source: Curtosy of [36]).

Pre-zygotic self-incompatibility reaction between pollen and ovule was observed in tamarind controlled by multiple alleles at the single gene loci. Reduced pollen fertility and pollen tube growth observed in open flowers with indehiscent anthers due to prezygotic SI and high percentage of aborted seeds in fruits produced from self pollination due to post-zygotic SI [49]. Gametiphytic SI in watermelon facilitated in good quality, sweet and seedless fruit production by treatment of pollen with soft x-rays followed by fertilizing female flowers with irradiated pollen [50].

Seedlessness achieved successfully with an optimum GA_3 concentration of 0.15mM when applied during pre- and post-anthesis time on field grown high quality seeded table grape culitvar "Swenson Red" both by dipping and spraying treatments [51].

Grafting is the most common method employed to improve quality production by true to type plant produce in a very less time by reducing pre bearing period of tropical fruit crops. The first grafted vegetable seedlings used were for watermelon (*Citrullus lanatus* L.) plants grafted onto bottle gourd (*Lagenaria siceraria* L.) rootstock to overcome Fusarium wilt. The rootstock and scion cultivars must be chosen with care to avoid loss [52]. Grafting is useful to multiply the superior plantation crop cultivars at a faster rate with extreme uniformity in yield and quality compared to those from seed. Grafting was successfully practiced in both tropical and temperate fruit crops including cashew, citrus, jackfruit, sapota and annonas. Once a cultivar is developed for seedlessness in fruit crops through ms induction or embryo-rescue, grafting technique can be employed for mass multiplication of the plantlets for crop production purposes. Invitro grafting is also employed during development of best hybrids with seedlessness and fruit quality. For example, rootstock improvement using symmetric somatic hybridization provides an opportunity to hybridize complementary rootstocks without breaking up successful gene combinations [46].

Knowledge of the floral morphology of tropical fruit crops to induce male-sterility for seedless fruits or dwarf fruiting plants

The floral morphology of the perennial fruit crops with vegetable use are described below for consideration of their improvement for hybrid seedless fruit production by using growth regulators, mutation or by male-sterilty and self-incompatibility systems. The specified crops received limited or no research attention with regards to hybrid seedless fruit production and were observed with less fruit quality, loss in fruit quality due to lot of constraints in production due to pests and pathogens. Understanding the floral taxonomy besides plant growth and production factors will help to rescue these crops using tissue culture or mutation breeding approaches to produce crops with good quality seedless fruits as well as to benefit both fresh and processed fruit consumers worldwide.

Tamarind

The flowers are hermaphroditic with three stamens which are shorter than the style so that the spacial arrangement of anthers and stigma may limit autogamoous and insect mediated self pollination. These protogynous flowers borne in clusters on racimose inflorescence, each flower composed of five petals (2 reduced to bristles) which are yellow with orange or red or pink streaks and superior ovary with up to 14 ovules. Flower buds are distinctly pink due to the outer color of the 4 sepals which are shed when the flower opens. Pollen grains are dimorphic, symmetrical, tricolporate, oblate speroidal in shape, compact together, sticky and measure 14 to 100 µm. The two major pollen sizes are 40-42µm and 22-25µm. Pollen sterility in tamarind is less than 2 percent which is very low. Under ambient conditions of 37-40^{o}C pollen viability was observed to be 88 percent for three days where as 97 percent pollen viable up to 100 days when stored at 4^{o}C. The breeding system might be a combination of facultative or obligate allogamy or some times frequent geitonogamy leading to low fruit quality. The fruits are borne on new shoots in clusters. They are dry indehescent long bean like pods with fleshy mesocarp, each schizocarpic (lomentum) fruit has many locules, multiseeded protected by hard brown shell. Seeds are hard, glossy-brown, square shape enclosed by a thin membrane.

Tamarind crop improvement is necessary to address development of dwarf tree types, seedlessness, high pod yield, cultivars with uniform pod quality, cultivars with less flower drop and high percent fruitset, cultivars with improved pod size and quality free from pink pod borer and other pests.

Jackfruit

Jackfruit is monoecious, having male and female flowers on separate inflorescences. Individual flowers are borne on an elongated axis forming a racemoid inflorescence, which is termed as a spike or head, and is enclosed in a protective pair of stipules. There are many light to dark-green male flowers arranged in densely crowded clusters of either sterile or fertile flowers. Female inflorescences are also light- to dark-green in color, but they are larger and have a thicker peduncle than the male inflorescences. The surface of the male inflorescence is smooth, while that of the female is granular. Stigmas are normally short and fresh in appearance for 1 to 2 weeks, but when inflorescences are bagged, the stigmas continue to elongate and remain fresh for a longer period. Male inflorescences are produced first, and are usually more numerous than females on the trees. At anthesis, the male flowers emit a sweet smell often compared to over-ripe fruit. They typically rot before abscission of the entire male inflorescence [53].

Jackfruit crop improvement is necessary to address dwarf cultivar development, seedlessness, high quality fruit yield with more number of fruits, cultivars with uniform fruit size and number, cultivars with less flower drop and high percent fruitset, separate cultivars for seed, fruit, culinery and fodder use.

Bael fruit

Fragrant flowers are borne in clusters of 4 to 7 along the young branchlets. These flowers have 4 recurved, fleshy petals, green outside, yellowish inside, and with 50 or more greenish-yellow stamens. The fruit is round, pyriform, oval, or oblong measuring 5-20 cm in diameter and is gray-green turning into yellowish with hard shell when fully ripe. It is dotted with aromatic, minute oil glands. Inside, there is a hard central core and 8 to 20 faintly defined triangular segments, with thin, dark-orange walls, filled with aromatic, pale-orange, pasty, sweet, resinous, more or less astringent, pulp. Embedded in the pulp are 10 to 15 seeds, flattened-oblong, about 1 cm long, bearing woolly hairs and each enclosed in a sac of adhesive, transparent mucilage that solidifies on drying [3].

Bael crop improvement is necessary to address reduced rind thickness, dwarf varieties free from pests and diseases.

Cashew nut

The giant or common cashew bears three kinds of flowers namely hermaphrodite (bisexual), male and sterile flowers. Sexually propagated cashew usually flowers from 24 to 36 months after planting. The flowers are small, polygamous born in clusters, which together form a panicle. Female flowers are only found in a bisexual form. The sterile flower is abnormal as it possesses neither stigma nor anthers with pollen. The color, shape and mean size of the panicles vary with the cultivars. The sexually propagated dwarf cashew flowers between 6 and 18 months after planting. The real cashew fruit is achne (nut) hanging from a fleshy peduncle of varying colors and size. The pseudo or false fruit, cashew apple, is a modified peduncle called receptacle is popularly known for its sweet juice.

Cashew nut crop improvement is necessary to address development of dwarf tree types, cultivars with high percent of bisexual flowers, separate cultivars for apple and nut, high nut yield, cultivars with uniform nut size and quality, cultivars with less flower drop and high percent fruit set.

Coconut

These coconut palms are largely cross pollinated. Some dwarf varities are self pollinated. At the age of 4-6 years, the coconut tree produces sweet-scented yellow flowers. Flowering occurs continuously. Flowers grow in clusters and are held by slightly branched stalks. The tree is monoecious by producing male and female flowers that grow on the same inflorescence. The flowers are protandrous, male flowers release pollen before the large female flower becomes receptive. After fruit set, the developed fully matured fruits are coconuts, botanically called as a drupe. The endosperm is initially in its nuclear phase suspended in within coconut water, later slowly this deposits edible coconut flesh along the walls of the coconut known as copra of the endocarp. The exocarp and mesocarp makes up the husk. The shell has three germination pores (micropyles) or eyes that are clearly visible on its outside surface once the husk is removed.

Coconut crop improvement is necessary to address drought tolerant cultivar development, dwarf tree types, Separate cultivars for copra, coconut-water, fibre and shell yield, uniform quality and high yield, cultivars with less flower and fruit drop, fruits with high percent fruitset, cultivars with improved nut size and nuts free from damage due to drought or pathogens. The fruitset, fruit quality and yield in coconut is highly influenced by growth factors.

Custard apple or Cherimoya (*Annona sp.*)

The flowers are hermaphroditic, pale green, and fleshy with mechanism to avoid self-pollination. Each flower has three outer, greenish, fleshy, oblong, downy petals and three smaller, pinkish inner petals with yellow or brown finely matted hairs outside, whitish with purple spot and many stamens on the inside. Flowers appear on the branches opposite to the leaves, solitary or in pairs or groups of three on flower stalks that are covered densely with fine rust-colored hairs of 8–12 mm in length. Flower buds are 15–18 mm in length and 5–8 mm in width at the base. The short lived flowers open as female then slowly progress to male stage in few hours later. The pollen shed as a permanent tetrad. Require pollinators (beetles) or hand pollination to collect pollen from male stage and deposit in female stage for successful fruit set. The fruit is round, oval, heart-shaped or kidney-shaped. The seeds are enclosed in the carpels and do not detach easily, the flavour balances intense sweetness with slight acidity and the soluble sugar content exceeds 17° Brix. The cultivar "Fino de Jete" gives fruits measuring 10-20cm in length with smooth skin having overlapping scales or knobby warts in appearance. The flesh of the cherimoya fruit contains numerous hard, inedible, black, bean-like, glossy seeds measuring 1cm to 2cm in length and about half as wide.

Soursop

The soursop is small upright, evergreen tree can grow upto 9.1m hight, young branches are hairy, leaves are oblong, glossy to oval 8-16cm in length. Flower stalks known as peduncles are 2-5mm long and woody. These appear opposite from the leaves or as an extra from near the leaf stalk containing 1 to 3 individual flowers on stout and woody pedicels with small bractlets nearer to the base which are densly hairy. The petals are thick and yellowish. Outer petals meet at the edges without overlapping and are broadly ovate (2.8 cm x 2.1cm to 2.5 cm) tapering to a point with a heart shaped base. They are evenly thick, and are covered with long, slender, soft hairs externally and matted finely with soft hairs within. Inner petals are oval shaped and overlap measuring 2.5cm to 2.8cm and are sharply angled and tapering at the base. Margins are comparatively thin, with fine matted soft hairs on both sides. The receptacle is conical and hairy. The stamens are 4.5 mm long and narrowly wedge-shaped. The connective-tip terminate abruptly and anther hollows are unequal. Sepals are quite thick and do not overlap. Carpels are linear and basally growing from one base. The ovaries are covered with dense reddish brown hairs, 1-ovuled, style is short and stigma truncate. Its pollen is shed as permanent tetrads.

Custard apple or Annona group of fruits require improvement in seedlessness, dwarf tree phenotype, high yield with uniform fruit size and quality with reduced annonacins or toxins, cultivars with transport durable quality of epidermis or outer green rind, cultivars with less flower drop and high percent fruitset, cultivars with improved fruit size and quality free from damage due to pests and diseases.

Date palm

The date palm is dioecious, having separate male and female plants. Both seed and cuttings are used for propagation as 50% of seedling progeny may give female trees. Date fruits (dates) are oval-cylindrical, 3 to 7 cm long, and about 2.5 cm in diameter, ranging from bright red to bright yellow in colour, depending on the variety. Plants grown from cuttings will fruit with in 2 to 3 years. Dates are wind pollinated and natural pollination occurs with an equal number of male and female plants. Manual pollination is usually preferred due to potential of one male parent's ability to pollinate 100 females. Therefore, male plants are not cultivated in an orchard as abundant pollen is available in market during the season.

Datepalm (eetha-pandu or south-Indian date palm) fruits require improvement in seedlessness, dwarf tree phenotype, high yield with uniform fruit size and quality with thick seedcoat, cultivars with transport durable quality of epidermis or outer green rind, cultivars with less flower drop and high percent fruitset, cultivars with improved fruit size and quality free from damage due to pests and diseases as well as resistant to strong winds or cyclone or drought.

Neredu tree

Flowering starts from March to April. Flowers are fragrant, small, about 5mm in diameter. The fruits are berries develop by May or June. These are drupaceous oblong and oval. Unripe fruit looks green and when it ripes color changes to pink to crimson red and then finally to black color.

Neredu require improvement in seedlessness, dwarf tree phenotype, high yield with uniform fruit size and quality, cultivars with transport durable quality of epidermis, cultivars with less flower and fruit drop, cultivars with high percent fruitset, cultivars with improved fruit size and quality free from damage due to pests and diseases, and drought tolerant cultivars.

Toddy palm

B. flabellifer is dioecious with male and female flowers on separate plants. The male flowers are less than 1 cm long and form semi circular clusters which are hidden beneath the scale-like bracts within the catkin-like inflorescences. In contrast, the female flowers are golfball-sized and solitary, sitting upon the surface of the inflorescence axis. After pollination, these blooms develop into fleshy fruits 15–25 cm wide, each containing 1-3 seeds. The fruits are black to brown with sweet, fibrous pulp and each seed is enclosed within a woody endocarp.

Toddy palm require improvement in special features of fruit traits (seedlessness in ripened fruits), dwarf tree phenotype, high yield with uniform fruit size and quality in both young mature and ripe fruit-cultivars, cultivars with transport durable quality of epidermis or outer black rind, cultivars with high percent fruitset, cultivars with improved fruit-size and quality free from damage due to pests and diseases, drought, ripe-fruit cultivars with long-term storage ability.

Mango

The flowers are produced in terminal panicles of 10 to 40cm in length. Each flower is small and white with five petals measuring 5 to 10mm in length with a mild to sweet fragrance. Out of 500 varieties of mango, some fruit once and some fruit twice in a year. The fruit takes five months from flowering to ripen. The fruits either monoembryonic with yellow, orange, red or green fruits (Indian type mangoes) or polymebryonic with bright yellow ripe fruits (Carabao mango of the philippines). The stone (endocarp) has single seed and it does not separate easily from the fibrous pulp of the mesocarp. The mangos have recalicitrant seeds which do not survive freezing and drying and germinate with high success when seeds are obtained from fresh mature fruits.

Mango require improvement in seedlessness without a spongy tissue and without loss in sweet or sour taste in a specific cultivar after improved, dwarf tree phenotype, high yield with uniform fruit size and quality, cultivars with transport durable quality of epidermis or outer rind and thick pulp with lot of fibre (Ex. Mango cv.Kottapalli-kobbari), cultivars with less flower drop and high percent fruitset, cultivars with improved fruit size and quality free from damage due to pests and diseases as well as drought tolerant.

In the above ten fruit crops, ms can be induced by 1) mutation breeding and using DNA markers for traist associated with seedlessness, such as parthenocarpy, male and female sterility and self-incompatibility. Through mutageneisis is not possible to target mutation at a specific gene locus. Thus requires laborious large scale population screening for selecting individual plants exhibiting desired phenotypes with seedlessness in fruits and dwarf plant types in these fruit crops at initial stages of crop improvement by mutation breeding. 2) The transgenic approaches at tapetal cell and seedcoat destruction using suicide gene approach during early pollen and seedcoat development respectively may help to reduce the time in achieving ms genotypes for seedless F_1 hybrid fruit or cultivar development. This approach requires isolated blocks of self-incompatible cultivars with specific growing conditions to eliminate cross pollination. Here parthenocarpy is not required for seedlessness 3) By induction of female sterility which can be achieved by destruction of ovule or stigma by suicide genes that could trigger seedless fruitset. Here there is no requirement of isolated blocks as fertilisation would be impossible and therefore, this approach is limited to the cultivars with parthenocarpic traits. 4) Increased expression or increased sensitivity to GA in the ovary or ovule. This was successfully used in citrus by over expression of GA20-oxidase or ovary-specific GA-I silencing and may help in producing obligatory seedless citrus cultivars (Ex: cybrid pumelo "G1 x HBP"). 5) Seedlessness may also be induced by managing environmental conditions such as low or high temperatures, chemical treatments, chromosomal aberrations and gene manipulations at meiosis or by self-incompatibility. It might be beneficial to use mutation breeding or genetic engineering and tissue culture methods for production of ms lines followed by adopting conventional breeding methods to produce seedless fruits from these ms lines for improved fruit quality.

Out of 96 seedless taxa, 66% belonged to plurispermic species with a maximum percentage of cultivated species than wild species, suggesting a selective pressure for parthenocarpy during domestication and breeding. In monospermic taxa, wild and cultivated species were similarly represented. The occurrence of parthenocarpy in wild species indicated that seedlessness may have an adaptive role due to high occurnace of polyploidy suggesting wide hybridization between cultivated diploid and wild species following the disruption of synchrony in time and space of reproductive developmental events, from sporogenesis to fruit development [17]. Though it is challenging to overcome reproductive barriers, based on the extreme success achieved in seedless citrus cultivars, inter-specific hybrid plants can potentially serve as an elite material for plant breeding, produced through the merging of genomes of parental species by allopolyploidization. Today, it is very easy and quick to align different genomic sequences to identify structural genetic variations using next generation DNA sequencing technologies with advances in bioinformatics that allows analysis at high resolution at a genome-wide level to screen the specific nuclear or mitochondrial gene loci coding for seedlessness by ms, SI or any enzymes involved in biochemical pathways. Thus, modern breeding methods that control microspore mother cell stage to a huge diploid plant, will help to induce seedlessness by induction of ms or SI and by identification of wild relatives in the under-developed and unexplored tropical fruits of nutritional and health values.

Conclusions

Male sterility is available option for induction of seedlessness in tropical fruit crops through mutation and tissue culture embryo-rescue techniques. The best fruit crops suggested for induction of male sterility and seedlessness include the listed perennial fruit crops in this chapter. For crop improvement point of view, these tropical origin perennial fruit crops can be categorised into four groups, 1) Jackfruit, breadfruit, durian, pine apple, sugar apple (custard apple), guava, sweet orange, papaya 2) Coconut, cashew nut 3) Sapodilla, lychee, loquat, mango, and 4) Tamarind, monkey pod or seema-chintakaya. Production of seedless tamarind and jackfruit would be more beneficial for fresh fruit as well as fruit processing industries. Because every part of tamarind tree (pulp, seed, leaves, flowers, bark and roots) and jack tree have nutritional, and industrial value. The research investment in development of seedless cultivars to scale up these fruit crop's production is viable necessity and this affort is a way to boost national income, benefit industry, conserving naturally available soil and environmental resources, farmers income and in conserving seed for farming every year as well as scientists to protect the transgene encoding seedlessness for future research.

Acknowledgement

I like to thank my husband, Mr.Pradeep Kumar Sripathi, IT professional for providing computer with highspeed internet facility to search available literature and our kids in providing ample time in successfully completing this review article on time.

Bibliography

1. CASR-California Agricultural Statistics Review. "Fruit and nut crops. California agricultural statistics review 2013-2014". (2014): 56-80.

2. Ahmad I and CP Chwee. "An overview of the world production and marketing of tropica and subtropical fruits". *Acta Horticulturae* 787 (2008): 47-58.

3. Morton J. "Bael Fruit". In: Fruits of warm climates. Julia F. Morton, Miami, FL (1987): 187-190.

4. Bahadur MSK. Materia medica of Madras. Printed and published by the Madras Government as per the recommendation of the Madras working committee, Calcutta International Exhibition (1891): 1883-1884.

5. Batugal P., *et al*. "Coconut Breeding". In: Jain S.M., Priyadarshan P.M. (eds) Breeding Plantation Tree Crops: Tropical Species. Springer, New York, NY (2009).

6. Horner HT and Palmer RG. "Mechanisms of genic male sterility". *Crop Science* 35 (1995): 1527.

7. Saxena KS., *et al*. "Structural variation in plant genomes". *Briefings in Functional Genomics* 13.4 (2014): 296-307.

8. Patrick S., *et al*. "Molecular basis of cms and fertility restoration". *Trends in Plant Science* 3.5 (1998): 175-180.

9. Hanson MR. "Plant mitochondrial mutants and male-sterility". *Annual Review of Genetics* 25 (1991): 461-486.

10. Celestina M., *et al*. "Induction of male-sterility in plants by a chimaeric ribonuclease gene". *Nature* 347 (1990): 737-741.

11. Xiao SX., *et al*. "Production and molecular characterization of diploid and tetraploid somatic cybrid plants between male-sterile Satsuma mandarin and seedy sweet orange cultivars". *Plant Cell, Tissue and Organ Culture* 116 (2014): 81-88.

12. Kumar S., *et al*. "Mutation breeding in chickpea". *Advances in Plants and Agricultural Research* 9.2 (2019): 355-362.

13. IAEA, International Atomic Energy Agency. Imrpovement of grain legume production using induced mutations. Proceedings of a workshop organized by the joint FAO/IAEA division of isotope and radiation applications of atomic energy for food and agricultural development. Held in Pullman, Washington fromm july 1-5 in 1986 (1988): 544.

14. Kott LS. "Production of mutants using the rapeseed doubled haploid system. Induced mutations and molecular techniques for crop improvement". Proceedings of an international symposium on the use of induced mutations and molecular techniques for crop improvement jointly organised by IAEA and the FAO of the UN held in Vienna, 19-23 June 1995. (1995): 505-515.

15. Vardi A., *et al*. "Induction of seedlessness in citrus :from classical techniques to emerging biotechnological approaches". *Journal of the American Society for Horticultural Science* 133.1 (2008): 117-126.

16. Lora J., *et al*. "Seedless fruits and the disruption of a conserved genetic pathway in angiosperm ovule development". 108.13 (2011): 5461-5465.

17. Picarella ME and A Mazzucato. "The occurence of seedlessness in higher plants: Insights on roles and mechanisms of parthenocarpy". *Systematic Review, Frontiers in Plant Science* 9 (2015): 1-11.

18. Akter S., *et al*. "Application of Single Nucleotide Polymorphism markers for the selection of male-sterility in crop plants". *Plant Breeding and Biotechnology* 4.4 (2016): 379-386.

19. Chase CD. "Cytoplasmic male sterility: a window to the world of plant mitochondrial-nuclear interactions". *Trends in Genetics* 23.2 (2007): 81-90.

20. Goldberg RB., *et al*. "Anther development: Basic principles and practical applications". *Plant Cell* 5 (1993): 1217-1229.

21. Huang H., *et al*. "Jasmonate action in plant growth and development". *Journal of Experimental Botany* 68.6 (2017): 1349-1359.

22. Pandolfini T. "Seedless fruit production by hormonal regulation of fruitset". *Nutrients* 1 (2009): 168-177.

23. Grosser JW and FG Gmitter Jr. "Protoplast fusion and Citrus improvement". *Plant Breeding Reviews* 8 (1990): 334-379.

24. Huang J-H., *et al*. "Abnormal mega-gametogenesis results in seedlessness of a polyembryonic Meiguicheng orange (Citrus sinesis) mutant created with gamma-rays". *Scientia Horticulturae* 217 (2017a): 73-83.

25. Goto S., *et al*. "QTL mapping of male-sterility and transmission pattern in progeny of Satsuma mandarin". *PLOS ONE* 13.7 (2018): e0200844.

26. Yamamoto M., *et al*. "Segregation for aborted anthers in hybrid seedlings using Citrus nobilis x C. deliciosa cv. Encore as the seed parent". *Journal of Japanese Society of Horticultural Science* 60 (1992): 785-789.

27. Yamamoto M., *et al*. "Aborted anthers of citrus result from gene-cytoplasmic male-sterility". *Science Horticulture* 70 (1997): 9-14.

28. Yamamoto M., *et al*. "Relationship between sterility and seedlessness in Citrus". *Journal of Japanese Society of Horticultural Science* 64.1 (1995): 23-29.

29. Mezzetti B., *et al*. "The DefH9-iaaM auxin-synthesizing gene increases plant fecundity and fruit production in strawberry and raspberry". *BMC Biotechnology* 4 (2004): 1-10.

30. Tuteja R., *et al*. "Cytoplasmic male-sterility associated chimeric open reading frames identified by mitochondrial genome sequencing of four Cajanus genotypes". *DNA Research* 20.5 (2013): 485-495.

31. Heng Sh., *et al*. "Comparative analysis of mitochondrial genomes between the hau cytoplasmic male-sterility (cms) line and its iso-nuclear maintainer line in Brassica juncea to reveal the origin of the cms-associated gene orf288". *BMC Genomics* 15 (2014): 322-334.

32. El-Siddig K., *et al*. "Book on Tropical fruit trees "Tamarind (Tamarindus indica L.)" Edited by Williams J.T., Smith R.W., Haq N., and Dunsiger. Published in 2000 by International centre for underutilized crops, University of Southhampton, Southhampton, SO17 1 BJ, UK Revised in 2006 by Southhampton centre for underutilized crops, Southhampton, UK (2006).

33. Dewi PS., *et al*. "Genotyping for male-sterility (ms) and ms gene mapping with RAPD markers in citrus, especially with precocious flowering seedlings from a cross of HY 16 X Grapefruit". *Acta Horticulture* 1065 (2015): 475-486.

34. Zheng B-B., *et al*. "Comparative transcript profiling of a male-sterile cybrid pummelo and its fertile type revealed altered gene expression related to flower development". *PLOS ONE* 7.8 (2012): e43758.

35. Varoquaux F., *et al*. "Less is better: New approaches for seedless fruit production". *Biotopics: Trends in Biotechnology* 18 (2000): 233-242.

36. Santos RC., *et al*. "Stenospermy and seed development in the "Brazilian seedless" variety of sugar apple (Annona squamosa)". *Anais da Academia Brasileira de Ciências* 86.4 (2014): 2101-2108.

37. Tonosaki K., *et al*. "Review: The importance of reproductive barriers and the effect of allopolyploidization on crop breeding". *Breeding Science* 66 (2016): 333-349.

38. Andrus CF., *et al*. "Production of seedless watermelons". Technical bulletin 1425. United States Department of Agriculture (USDA)-National Agricultural laboratory (NAL), current serial records (2019): 16.

39. Jeridi M., *et al*. "Homoeologous chromosome pairing between the A and B genomes of Musa spp. revealed by genomic in-situ hybridization". *Annals of Botany* 108 (2011): 975-981.

40. Stommel JR. "Barriers for introgression of Solanum ochranthum into tomato via somatic hybrids". *Journal of the American Society for Horticultural Science* 126.5 (2001): 587-592.

41. Padmaja M., *et al*. "Leaf nutrient status of mango Cv.Baneshan in relation to yield". *The Andhra Agricultural Journal* 48 (2001): 273-275.

42. Sangamesh K. "Micropropagation and mutation studies in Pomegranate (Punica granatum L.,)". MS Thesis submitted to the Department of Crop Improvement and Biotechnology, KRCC Horticulture, Arabhavi, UAS, Bagalkot (2014): 117.

43. Olivares-Fuster O., *et al*. "Green fluorescent protein as a visual marker in somatic hybridization". *Annals of Botany* 89.4 (2002): 491-497.

44. Bassene JB., *et al*. "Influence of mitochondria on gene expression in a citrus cybrid". *Plant Cell Report* 30 (2011): 1077-1085.

45. Omar AA., *et al*. "Production of three new grapefruit cybrids with potential for improved citrus canker resistance". *In Vitro Cellular and Developmental Biology - Plant* 53 (2017): 256-269.

46. Grosser JW and FG Gmitter. "SIVB Congress Symposium Proceedings "Thinking Outside the Cell": Applications of Somatic Hybridization and Cybridization in Crop Improvement, with Citrus as a Model". *In Vitro Cellular and Developmental Biology - Plant* 41 (2005): 220-225.

47. Guo WW., *et al*. "Targeted cybridization in citrus: Transfer of Satsuma cytoplasm to seedy cultivars for potential seedlessness". *Plant Cell Report* 22 (2004): 752-758.

48. Santos RC., *et al*. "Gibberellic acid induces parthenocarpy and increases fruit size in the 'Gefner' custard apple (Annona cherimola x Annona squamosa)". *Australian Journal of Crop Science* 10 (2016): 314-321.

49. Diallo BO., *et al*. "Breeding system and pollination biology of the semi-domesticated fruit tree, Tamarindus indica L., (Leguminosae: Caesalpinioideae): Implications for fruit production, selective breeding, and conservation of genetic resources". *African Journal of Biotechnology* 7.22 (2008): 4068-4075.

50. Qu H-y., *et al*. "The mechanism of seedlessness in watermelon generated using soft-x-ray irradiated pollen". *African Journal of Agricultural Research* 11.25 (2016): 2200-2204.

51. Fellman C., *et al*. "Gibberellic acid induced seedlessness in field-gron vines of "Swenson Red" Grape". *Horticultural Science* 26.7 (1991): 873-875.

52. Lucas AG., *et al*. "Grafting in Vegetable Crops: A Great Technique for Agriculture". *International Journal of Vegetable Science* (2017): 18.

53. Elsawa S. "Pollination and breeding of jackfruit (Artocarpus heterophyllus L) in South Florida". Thesis submitted to Florida International University, Miami, Florida. FIU Digital commons (1998).