




The origin of mangosteen: A review

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Abstract Mangosteen (*Garcinia mangostana* var. *mangostana*) is an obligate apomictic species which does not require a fertilization process to produce seeds. The absence of male plants of mangosteen brings about an interest to find out the origin of mangosteen. Determining the parents is important in any improvement program to produce better quality products. Several hypotheses were proposed since the 1990s, and the most popular and frequently cited one stated that mangosteen was a product of hybridization of *G. mangostana* var. *malaccensis* and *G. celebica*. This hypothesis then became the groundwork for later experiments to further study the origin of mangosteen. The methods used included morphological comparison between *Garcinia* species, molecular

marker analyses from the internal spacer, chloroplast sequencing, and microsatellite and internal transcribed spacer (ITS) sequence analysis. This review discusses the different approaches taken to study the origin of mangosteen and suggestions for future work in order to verify the original parents of mangosteen.

Keywords Mangosteen origin · *Garcinia* · Agamospermy · Tropical fruit

Introduction

Mangosteen is the most important cultivated species in the genus of *Garcinia*. The species was recognised by Linnaeus and named as *G. mangostana* L. (Linnaeus 1753). However, the latest taxonomic revision by Nazre et al. (2018) concluded that the cultivated mangosteen was *G. mangostana* var. *mangostana* and another two varieties of mangosteen were wild species: *G. mangostana* var. *malaccensis* and *G. mangostana* var. *borneensis*. The main reason for its popularity for cultivation is the fleshy portion of the pulp which accounts for roughly 30% of fruit weight, and it is eaten fresh (Cruz 2001). Fairchild (1915) has recognised that this fruit is known as ‘the queen of fruits’ because of its appetizing and finest taste.

Whitmore (1973) explained that the crown of *Garcinia* trees is generally monopodial, conical shape (pagoda-like) and dense. Monopodial branching can be described as a growth pattern by which the

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secondary shoots and branches grow from behind the growing point but remain subsidiary to the main stem, enabling the plant to grow upwards without limit (Nazre et al. 2018). According to Palakawong and Delaquis (2018), the height of a mangosteen tree may reach up to 25 m, but it requires several years to reach such a height because of its slow growth rate. According to Lim (1984), based on his observation on the cultivated tree of mangosteen, *Garcinia* species generally has a slow growing pattern. The tree would likely take 10 to 15 years to reach the flowering stage. However, a study by Palakawong and Delaquis (2018) suggested that the period for mangosteen to reach flowering and fruiting stage may even be less than 7 years. Based on the record, *Garcinia* trees are usually restricted to the interior forest or shady places (Whitmore 1973) which do not receive direct light from the sun. Some species of *Garcinia* like *G. celebica* can be found in coastal forest, preferring to grow within a shady area rather than in an open area in that particular forest. According to Nazre et al. (2018), mangosteen, *G. penangiana*, and *G. diospyrifolia* are among *Garcinia* species that are most likely to grow in lowland or hill forest (approximately 900 m and below) and not confined to montane forest.

Osman (2006) divided mangosteen production into major and minor production areas. Four countries from Southeast Asia have been recognised to be the major production areas of mangosteen: Malaysia, Thailand, Philippines and Indonesia (about 85% of the total production is produced by Thailand). Other minor production areas include Australia, Vietnam, United States of America and Brazil. Recently, it has also been cultivated in other tropical areas, including Cuba, Dominica, Equador, Gabon, Ghana, Guatemala, Honduras, India, Jamaica, Liberia, Myanmar, Puerto Rico, Singapore and Zanzibar (Cruz 2001; Lim 2012; Murthy et al. 2018). Mangosteen requires fertile, well-drained and high organic content with slightly acidic soils combined with high humidity (at least 1200 mm annual rainfall) to grow optimally (Osman 2006). Osman (2006) also emphasised that a short dry period is essential to induce flowering, and the range of optimum temperature is between 25 and 35 °C.

In Malaysia, several states have been identified to have suitable conditions to grow mangosteen (especially in small orchards in rural areas), including Penang, Perak, Johor, Kedah, Negeri Sembilan and Sabah

(Osman 2006). The total area used for mangosteen cultivation in Malaysia was estimated to be between 7000 and 8000 ha (Osman 2006). There are two fruiting seasons every year, with a major and minor fruiting season whereby more fruits are produced during the major fruiting season. The major fruiting season usually occurs between May and September (peak season between July and August) while the minor fruiting season occurs between November and February (peak season in December). However, the duration and timing of the fruiting season are influenced by the location and climatic factors. Thus, the fruiting season may differ from one location to another (Osman 2006).

Mangosteen's large economic potential in medicine, such as having anti-inflammatory, anti-tumour and anti-oxidative properties (Pedraza-Chaverri et al. 2008), has prompted researchers to improve its cultivation. The latest study confirmed that mangosteen possessed various types of beneficial chemical compounds. Ansori et al. (2020) listed up to 78 different chemical compounds present in the entire mangosteen tree, with xanthones derivatives as the major type of bioactive secondary metabolites. Other compounds found in mangosteen fruit include gartanin (related to inducing cell death in lung cancer cells) as well as isogarcinol which helps in maintaining immune system regulation (Ansori et al. 2020).

Despite having multitude of benefits, growing mangosteen can be long and intricate. Mangosteen has a long juvenile period, and it usually takes 8 to 15 years to bear fruits (Poerwanto 2000). Its slow growth is caused by poor growth of its root system due to no root hairs, poor branching, easily broken and disturbed roots by adverse environment that results in very small contact surfaces between roots and soil; poor nutrient and water uptake; low photosynthetic rate; low cell division rate in the apical meristem; and long shoot dormancy period (Cox 1988; Wieble et al. 1992; Ramlan et al. 1992; Poerwanto et al. 1995; Poerwanto 2000). Grafting method has been used on mangosteen in order to shorten the flowering period and improve its poor root system. However, the growth of grafted seedlings is worse even when using matured scions of mangosteen compared to non-grafted seedlings (Poerwanto 2000). Hence, micropropagation has become a suitable option to propagate mangosteen via in vitro organogenesis, using seeds as explant (Maadon et al. 2016).

Nevertheless, mangosteen needs to be improved to develop better traits. In order to do so, understanding the reproductive biology and the parental lineage is of utmost importance. Thus, this review discusses how different hypotheses of the origin of mangosteen have emerged, with the aim of obtaining information for further studies to aid future breeding initiatives.

Agamospermy in *Garcinia* species

In general, the involvement of pollinators is significant to ensure the pollen is able to reach and stick to the stigma of the flowers, especially for dioecious species. Usually, bees are known to be common pollinators in the tropical rain forest (Renner and Feil 1993). Some *Garcinia* species, such as mangosteen and *G. diospyrifolia* may have petals that have attractive colours which are considered as a contributing factor to attract pollinators. However, Nazre et al. (2018) emphasized that even without the help of pollinators, *Garcinia* species might actually survive and keep on reproducing due to its ability to generate fertile seeds without fertilization between male gametes and female gametes. This characteristic is known as agamospermy.

Theoretically, the progeny of any plant species considered as agamospermy will exhibit similar morphological characters to its mother plant; this is also known as seed clones (Richards 2003). Based on the observation made by Richards (1990b), five important conditions are listed to determine whether a species is considered as agamospermy or not: (1) the ability to produce viable seeds even without the presence of pollen, (2) the development of the embryo precedes anthesis, (3) the growth of the vegetative buds from nucellar, integument tissues or adventitious embryos, (4) the production of multiple seedlings from a single seed (polyembryony), and (5) the scarcity or absence of male plants, flowers or viable pollen. These five conditions are used to rule out the species in the genus of *Garcinia* that does not fulfil any one or more of these conditions (Table 1). According to Nazre et al. (2018), among the species accepted to be agamospermous based on the five conditions are mangosteen, *G. celebica* and *G. penangiana*.

Garcinia species can reproduce through sexual and asexual mechanisms. If male plants exist in the population, there is a high tendency of sexual seed formation through fertilization process. On the other

Table 1 Ten species of *Garcinia* that fulfil one of the agamospermy criteria Richards (1990b)

Criteria	Mangosteen	<i>parvifolia</i>	<i>G. cowa</i>	<i>G. treubii</i>	<i>G. livingstonei</i>	<i>G. penangiana</i>	<i>G. scorteichinii</i>	<i>G. celebica</i>	<i>G. atroviridis</i>	<i>G. nigrolineata</i>
Produce germinable seeds without male or bagging flowers	✓	✓			✓			✓	✓	
Precocious development of the embryo before anthesis		✓								
Occurrence of adventitious proembryos, budding vegetatively from nucellar or integumental tissues	✓		✓	✓				✓		
Multiple seedling production from a single seed	✓	✓			✓	✓		✓		
Rarity or absence of male plant	✓				✓	✓			✓	✓

hand, if a male plant is absent, some species can reproduce asexually Richards (1990b). These species are regarded as facultative agamospermy as they are able to switch between the two methods (sexual and asexual) depending on the situation. According to Malik et al. (2005), *G. indica* and *G. cambogia* are considered to be facultative agamospermy in *Garcinia*. Nonetheless, the species considered as obligate agamospermy are mangosteen Richards (1990b), *G. scortechinii* (Thomas 1997) and *G. xanthochymus* (Malik et al. 2005) due to the absence of a male plant. According to Idris and Rukayah (1987), the absence of male plant is closely related to the cultural practice of chopping down male plant since the cultivators assume that the plant has no benefits (produce no fruits). An obligate agamospermy is unable to switch between sexual and asexual reproduction modes.

Although agamospermy is a desirable trait for crop improvement as it has the ability to retain genetic information of the progeny with extremely low variability, there are also hidden disadvantages. Richards (2003) mentioned that one of the main drawbacks of apomictic plants was the high risk of accumulating of disadvantageous recessive mutants. Due to this situation, plants with agamospermy traits are rarely diploid (Richards 2003). If a plant is diploid, the accumulated mutation can easily be expressed in its haploid products through the meiosis process. Being a polyploid enables the suppression of the expression of disadvantageous traits, causing agamospermous plants to be polyploid (Richards 2003). This was supported by Matra et al. (2016) who revealed evidence of multiple ploidy levels in mangosteen by investigating the number of alleles per locus in Ibaraki *Garcinia Mangostana* Puspahiang (IGMP) loci. Based on their analysis, the mangosteen originating from Puspahiang, Indonesia has more than two alleles per locus with maximum of five alleles per locus as polyploidy plants have more than two alleles per locus (Matra et al. 2016). Midin et al. (2017) also reported similar results, with high number of mangosteen chromosomes (ranged between 74 and 110) due to the possibility of aneuploidization occurring in the genome. Bayer (1996) asserted that agamospermy species with lower levels (triploid and tetraploid) might reproduce sexually due to the possibility of gene exchange activity. Furthermore, species that have higher ploidy levels tend to be obligate agamospermy as the species is reproductively isolated.

Surprisingly, as one of the obligate agamospermy, mangosteen exhibits morphological variation. According to Ramage et al. (2004), the mangosteen population exhibits unusual morphological characteristics, such as differences in yield, duration of juvenile period and fruit size, suggesting that variations do exist in the apomictic population of mangosteen. Ramage et al. (2004) studied the genetic diversity of mangosteen and its relatives (from various geographical locations in Indonesia, Malaysia, Thailand, Australia and East Africa) by using Randomly Amplified DNA Fingerprinting (RAF) method. The research suggested that one of the possible reasons for the variation was from multiple backcrossing events between mangosteen with other species in *Garcinia*. Unfortunately, according to Richards (1990a), backcrossing has a low possibility of success as shown by various experiments especially with *G. celebica* and *G. penangiana*. Another possibility pointed out by Ramage et al. (2004) is that the variation may be the product of genetic mutation accumulation. Even with that possibility, there is hardly enough evidence to confirm that such accumulation will lead to genetic diversity as compared to hybridization which definitely produces a significant level of variation in the population. The results, however, have indicated that the genetic variation within mangosteen was derived from at least three independent ancestors with different genotypes and phenotypes (Ramage et al. 2004). The results also showed that several accessions had identical genotypes and yet, showed different phenotype. Thus, it is important to bear in mind that environmental factors also play roles in determining how the plant will respond to the surroundings, appearing later in different physical characters.

Another experiment was conducted by Sobir et al. (2011) to assess the genetic variability of mangosteen using inter simple sequence repeat (ISSR) markers. The results showed that several closely related species to mangosteen, including *G. silygifolia*, *G. livingstonei*, *G. bancana*, *G. picrorhiza* and *G. porrecta* were completely separated from mangosteen accessions. In contrast, four species of *Garcinia*, *G. xanthochymus*, *G. mangostana* var. *malaccensis*, *G. celebica* and *G. dulcis*, were clustered together with mangosteen at 22% dissimilarity level (Sobir et al. 2011). In addition, two accessions of *G. celebica* were clustered separately from mangosteen while one accession of *G. celebica* was clustered with mangosteen. This proved that the genetic variability within *G. celebica* affected its

placement among the clusters. Some of the researchers' hypotheses that have been proposed over the years are discussed in the following subheadings.

Hypotheses of the origin of mangosteen

Since mangosteen is considered as one of the important cultivated crops because of its delicious taste, various studies were conducted to improve mangosteen cultivation. These studies used various methods to obtain comprehensive data on how mangosteen can survive as an apomictic species as shown in Table 2. It is crucial for researchers to study and identify the origin of mangosteen as the data can help produce a new variety of mangosteen with better traits.

Hypothesis of mangosteen origin based on morphological characteristics

According to Richards (1990a), mangosteen is an allotetraploid species, a hybrid that originated from

an interspecific crossing between *G. celebica* and *G. malaccensis*, making contrast among their morphological characteristics such as fruit colour, latex colour and presence of a sessile stigma. Thirteen main physical characters of mangosteen were compared with *G. celebica* and *G. malaccensis* as shown in Tables 3, 4 and 5. Five characters of mangosteen, flowering period, depth of stigma lobes, stigma diameter, and presence of staminodes in the female flower and fruit taste, were considered to be intermediate between *G. celebica* and *G. malaccensis*. Another four characters, fruit shape, stamen mass clearly lobed, stigma surface and fruit surface, resembled *G. celebica* while the remaining four characters, sessile stigma, latex colour, petal colour and fruit colour, were similar to *G. malaccensis*.

Despite the limited information on cytogenetics evidences, Richards (1990a) pointed out that mangosteen was derived from two different *Garcinia* species; a wild species of *G. penangiana* (as *G. malaccensis*) and a cultivated species of *G. celebica*.

Table 2 The previous researches related to the origin of mangosteen since 1990

Hypotheses of the origin of mangosteen	Tools	References
Mangosteen was the hybridisation product of <i>G. celebica</i> and <i>G. penangiana</i> (as <i>G. mangostana</i> var. <i>malaccensis</i>)	Morphological characters observation	Richards (1990a)
<i>G. mangostana</i> var. <i>malaccensis</i> was closer to mangosteen phylogenetically as compared to <i>G. celebica</i>	Comparing ITS sequence	Yapwattanaphun et al. (2004)
<i>G. opaca</i> was a closer relative to mangosteen and <i>G. penangiana</i> (as <i>G. mangostana</i> var. <i>malaccensis</i>) compared to <i>G. celebica</i>	Molecular markers from internal spacer, chloroplast sequencing and microsatellite	Abdullah et al. (2012)
(a) Mangosteen was a hybrid from different varieties of <i>G. mangostana</i> var. <i>malaccensis</i>	Morphological observation and ITS sequence	Nazre (2014)
(b) Superior selection from female trees of <i>G. malaccensis</i> in Peninsular Malaysia		

Table 3 Morphological characteristics of mangosteen that were considered to be intermediate between *G. celebica* and *G. penangiana* (as *G. mangostana* var. *malaccensis*) (Richards 1990a)

Morphological characteristics	<i>G. celebica</i>	Mangosteen	<i>G. penangiana</i> (as <i>G. mangostana</i> var. <i>malaccensis</i>)
Flowering period	January–March	March–April and July–September	April–July
Depth of stigma lobes	20% diameter	50% diameter	80% diameter
Stigma diameter	5–6 mm	8–12 mm	10–20 mm
Presence of staminodes	No staminodes	Small staminodes	Staminodes
Fruit taste	Astringent	Sweet, sour	Sweet, insipid

Table 4 Morphological characteristics of mangosteen that similar to *G. celebica* (Richards 1990a)

Morphological characteristics	<i>G. celebica</i>	Mangosteen	<i>G. penangiana</i> (as <i>G. mangostana</i> var. <i>malaccensis</i>)
Fruit shape	Globose	Globose	Depressed globose
Stamen mass	Lobed	Staminode mass lobed	Nearly unlobed
Stigma surface	Smooth	Smooth	Corrugated
Fruit surface	Smooth	Smooth	Wrinkled

Table 5 Morphological characteristics of mangosteen that resemble *G. penangiana* (as *G. mangostana* var. *malaccensis*) (Richards 1990a)

Morphological characteristics	<i>G. celebica</i>	Mangosteen	<i>G. penangiana</i> (as <i>G. mangostana</i> var. <i>malaccensis</i>)
Stigma	Stipitate	Sessile	Sessile
Latex colour	White	Yellow	Yellow
Petal colour	Cream	Red	White to pink
Fruit colour	Red	Purple	Maroon

Richards (1990a) also suggested in his study that the present cultivated mangosteen originally emerged from one asexual line which led back to the original hybrid between *G. celebica* and *G. penangiana* (as *G. mangostana* var. *malaccensis*).

Using molecular evidence to determine the parents of mangosteen

The research conducted by Abdullah et al. (2012) investigated the possible parents of mangosteen from several related species within the *Garcinia* section by comparing the genetic analysis of internal spacer, chloroplast and microsatellite regions. The study concluded that *G. opaca* was more likely to be one of the parents of mangosteen rather than *G. celebica*. This conclusion was drawn from the products amplified by using chloroplast and ITS markers, showing that *G. opaca* shared similar allele sizes with mangosteen at two different loci (GM5 and GM11), *G. opaca* and *G. penangiana* (as *G. mangostana* var. *malaccensis*) shared similar allele sizes at two loci (GM10 and GM11) and mangosteen shared similar allele sizes with *G. penangiana* (as *G. mangostana* var. *malaccensis*) at three loci (GM1, GM2 and GM11). On the other hand, *G. celebica* showed differences in allele sizes for three loci (GM8, GM10 and GM11)

as compared to the other *Garcinia* species. Therefore, after comparing with all of the available loci, Abdullah et al. (2012) suggested that *G. celebica* was not involved in the contribution in the alleles of mangosteen.

Mangosteen origin theory based on morphology and ITS sequence analysis

Nazre (2014) stated that the identification of *G. mangostana* var. *malaccensis* is involved in a classical problem which has led to the misidentification of the species. The *G. penangiana* obtained from Pasoh Forest Reserve, Malaysia was mistreated as *G. mangostana* var. *malaccensis* and was used by Ha et al. (1988) in their cytogenetics studies. Later, this inaccurate information was used by Richards (1990a) who concluded that mangosteen originated from a hybrid between *G. celebica* and *G. mangostana* var. *malaccensis*. Abdullah et al. (2012) also drew the conclusion on the origin of mangosteen by using inaccurate information. Besides that, Nazre (2014) emphasized that by analysing the morphological characters, e.g. petal colour, presence and shape of pistillodes, fruit, leaf colour and glandular line patterns, of mangosteen, it was clear that mangosteen was more closely related to *G. mangostana* var. *malaccensis* rather than *G. penangiana*.

The results of ITS sequence similarity of mangosteen and *G. mangostana* var. *malaccensis* (grouped into the same monophyletic clade based on Nazre 2014) and the similar morphological resemblance suggested that these two species should be treated as one single species which might have diverged later into different botanical varieties. Interestingly, Nazre (2014) proposed two possibilities if mangosteen and *G. malaccensis* were grouped together as single species. First, the hybridization process of mangosteen might occur in the past between the varieties of *G. mangostana* var. *malaccensis*. Then, a phenomenon

that is known as concerted evolution may cause the ITS sequence to homogenize multiple loci in the multigene family. It was not a surprise that later in Nazre et al. (2018) taxonomical study, they decided to reduce *G. malaccensis* into a variety of mangosteen (*G. mangostana* var. *malaccensis*) based on the morphological comparison (the fruit) and ITS sequence that were similar to mangosteen.

Nevertheless, the first possibility only works when the chances of hybridization between varieties are high (Nazre 2014). The difficulty of finding a male tree of *G. mangostana* var. *malaccensis* makes it extremely rare for the first possibility to occur. The second possibility involves the human selection of *G. mangostana* var. *malaccensis* whereby cultivators only prefer to retain the female plants which can bear fruits. Then, the varieties of mangosteen arose from a long and extensive selection process. This possibility may be true based on the morphological and molecular data which pointed out that variations exist within the population of *G. mangostana* var. *malaccensis* and they can be easily distinguished by geographical location (Nazre 2014).

Potential origin of mangosteen

G. mangostana var. *malaccensis* was initially observed by Ha et al. (1988) in Pasoh Forest Reserve, Negeri Sembilan, Malaysia. The observation was made to study agamospermy that existed in *Garcinia* species. Later, a direct connection between *G. malaccensis* and mangosteen was made by Richards (1990a) to find the parents of mangosteen by narrowing down the *Garcinia* species using morphological comparison. Later, Yapwattanaphun et al. (2004) further studied *G. mangostana* var. *malaccensis* using samples obtained from Bogor, Indonesia via ITS sequence analysis, and revealed that both species, mangosteen and *G. mangostana* var. *malaccensis* were phylogenetically closer than other *Garcinia* species. This result strengthens the hypothesis of *G. mangostana* var. *malaccensis* being the parent of mangosteen. In addition, Yapwattanaphun et al. (2004) also stated that there was a possibility that allopolyploidization might occur due to hybridization based on their observations on nucleotide additivity in the ITS sequence in both mangosteen and *G. mangostana* var. *malaccensis*. Then, by using the information provided by Richards (1990a) on *G.*

mangostana var. *malaccensis*, Abdullah et al. (2012) similarly concluded after examining the molecular evidences obtained from their research, confirming that *G. mangostana* var. *malaccensis*, which was collected in Pasoh Forest Reserve, was highly plausible to be the parent of mangosteen.

But, Nazre (2014) proved that *G. mangostana* var. *malaccensis* was not the species used by Ha et al. (1988), Richards (1990a) and Abdullah et al. (2012), but instead was the species collected in Pasoh Reserve Forest identified as *G. penangiana*. Nazre (2014) also stated that there were insufficient data resulting from previous authors who had not completely checked the identity of *G. mangostana* var. *malaccensis*. Such conclusion was crucial as the data of *G. penangiana* (mistaken for *G. mangostana* var. *malaccensis*) by Ha et al. (1988), Richards (1990a) and Abdullah et al. (2012) might be used in later studies if not properly corrected. Nevertheless, this situation creates new opportunities to properly identify and obtain the real data of *G. mangostana* var. *malaccensis*. Because of the misidentification, Nazre (2014) concluded that *G. mangostana* var. *malaccensis* has never been studied by previous authors in Malaysia, and he confirmed that *G. mangostana* var. *malaccensis* to be the origin of mangosteen.

Interestingly, due to the misidentification, we can assume that other species, such as *G. penangiana*, can be considered as one of the potential origins of mangosteen. In the previous study, Abdullah et al. (2012) found that *G. penangiana* (misidentified as *G. mangostana* var. *malaccensis*) was the closer relative to mangosteen in the phylogenetic analysis. This species is interesting to be investigated in the future. The study on its chromosome and genome structure is still lacking. *G. penangiana* is a widely distributed species in Peninsular Malaysia (Nazre et al. 2007) and locally, it is known as ‘Kandis Burung’. Morphologically, its bark is dark brown, its leaves are bright reddish when dry (Nazre 2014), and its fruit shape is ovoid or globose with fewer stamen bundles that are widely spaced and displaying a nodule-like surface.

One of the aspects that can be further studied is the cytogenetics of *Garcinia* species since a lot of research conducted proposed that the actual number of chromosomes in *Garcinia* species has not been clearly defined, except for mangosteen. The cytogenetics data of *Garcinia* species such as chromosome number and genome size could help determine the

actual ancestors of mangosteen and later, be used in the breeding program to improve its cultivation and production quality. The chromosome number of other *Garcinia* species could lead to the possibility of other species to be the possible parents of mangosteen. As for now, the cytogenetics data of *G. mangostana* var. *malaccensis* (potential mangosteen origin) such as chromosome number and genome size are not available. Physical mapping techniques such as genomic in situ hybridisation (GISH) can be implemented in order to determine the origin of mangosteen by using the genomic DNA from possible ancestors as DNA probes, and mapping the blocking DNA on the mangosteen chromosome. GISH technique was first demonstrated by Schwarzacher et al. (1989) on *Hordeum chilense* x *Secale africanum* hybrid plants. GISH has also been employed on natural hybrids or allopolyploid species such as *Millium montianum*, *Trifolium repens*, and *Carassius auratus* (Bennett et al. 1992; Zhu et al. 2006; Williams et al. 2012). In addition, flow cytometry analysis can be performed to estimate the genome size and ploidy level of potential mangosteen ancestors. These data can be used as an important marker to indicate the occurrence of hybridisation among species (Ellul et al. 2002; Madon et al. 2008; Mandák et al. 2016). Mandák et al. (2016) revealed the origin of *Alnus glutinosa* by using flow cytometry and microsatellite approach. As for now, the genome size data of mangosteen has already been reported by Midin et al. (2017). Besides that, the leaf anatomy and micromorphology analyses can be used to ascertain mangosteen ancestors. The data obtained from these analyses can be utilised to construct a dichotomous identification key to study the relationship among *Garcinia* species.

Conclusion

Several hypotheses have been proposed by previous studies to determine the origin of mangosteen as discussed above. Various methodologies have been used from the most conventional taxonomy identification to more advanced molecular analysis. Cytogenetics tools such as GISH and flow cytometry should be applied in the future researches to determine the mangosteen origin. The gDNA of

mangosteen potential parents as discussed above should be utilised as probe and blocking DNA in GISH. The genome size of possible parents of mangosteen can be determined by flow cytometry as it can be used to detect the occurrence of species hybridisation. This information will provide ideas for crop improvement and further development in the cultivation of mangosteen in the future. Determination of mangosteen origin will also help the production of new varieties with superior traits as compared to the existing mangosteen.

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