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Compatibility Studies of Different Rootstocks for Softwood Grafting in Abiu [*Pouteria caimito* (Ruiz & Pavon.) Radlk.]

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Abstract

This study evaluated the effect of six different rootstocks on the success of softwood grafting in abiu (*Pouteria caimito*), an exotic fruit crop. The different rootstocks used as treatments were *Pouteria caimito* (abiu), eggfruit (*Pouteria campechiana*), sapota (*Manilkara sapota*), khirni (*Manilkara hexandra*), star apple (*Chrysophyllum cainito*) and wild star apple (*Chrysophyllum oliviforme*). The results revealed that the scion growth parameters like number of leaves, number of branches and length of shoots of the resultant grafts was found to be maximum when abiu scion was grafted on abiu rootstock followed by sapota rootstock. Abiu rootstock exhibited the highest graft survival percentage (77.73%), followed by sapota (44.43%), while the other four rootstocks failed to survive till the end of experiment. To understand the anatomical basis of success of abiu on both abiu and sapota rootstocks, a histological study of the graft union was conducted for both abiu and sapota rootstocks. The study revealed well-developed vascular connections and strong callus formation in abiu rootstock, suggesting superior graft compatibility. Sapota rootstock also showed good union quality but was less efficient than abiu rootstock. Based on these results, abiu and sapota found to be suitable and compatible rootstock for softwood grafting in abiu.

***Key words*:** Sapotaceae, Propagation, Exotic, Graft union, Histology

Abiu [*Pouteria caimito* (Ruiz & Pavon.) Radlk.] is a tropical fruit tree belonging to Sapotaceae family originated from Amazon region of South America. Commonly called as 'Golden Star Apple,' this fruit has garnered the international market due to its distinct flavour and growing popularity. Abiu has been distributed to Venezuela, Peru, Ecuador, Trinidad, Brazil, and South Asian countries whereas in India, it is mainly found in the southern states of Kerala, Karnataka, and Tamil Nadu. The tree ranges from medium to large size with a pyramidal or oval canopy. The ripe fruits of abiu are unique with its bright yellow colour, round to oval shape and creamy white flesh, which is delicate, sweet, and tastes similar to tender coconut, custard apple and vanilla. Recently, exotic fruits are gaining popularity among the public with high market demand in India and abiu is one among them which is highly preferred by consumers due to its unique taste and flavour [1].

In India, though its cultivation and distribution are limited, it has potential for increased popularity due to its high consumer demand, as the fruits are very tasty with health benefits. It can be recommended as an intercrop as it needs only low maintenance, and can act as an ornamental or shade tree. As a highly profitable, adaptable, and low labour-intensive crop, abiu is well-suited for cultivation in Kerala.

Abiu is commonly propagated by seed which remains viable only for a few days and exhibits great genetic variability. The higher price of the fruits increases the cost of cultivation for raising rootstocks for their vegetative multiplication. Identifying a compatible, locally available rootstock or rootstocks is the pre-requisite to develop a cost-effective alternative. In line with this, a study was conducted to identify the best rootstock species from six different species of the Sapotaceae family that are compatible with abiu for developing grafts by adopting softwood grafting method.

MATERIALS AND METHODS

Experimental site

The present investigation entitled 'Compatibility studies of different rootstocks for softwood grafting in abiu [*Pouteria caimito* (Ruiz & Pavon.) Radlk.]' was carried out in the Department of Fruit Science, Kerala Agricultural University, Vellanikkara, Thrissur, Kerala. Vellanikkara is situated at 10^0 54'N latitude and 76^0 28'E longitude, and lying 22.25 m above MSL. This place experiences a humid tropical climate with heavy rainfall during monsoon and post-monsoon seasons.

Materials

The abiu scions for grafting was collected from a farmer's field at Thrissur. The different rootstocks were raised from the seeds taken from firm ripe fruits collected from the college orchard. The chemicals such as ethyl alcohol, acetone, xylene (rectified 98%), D.P.X. mountant and safranin required

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for staining and fixing microsections, were purchased from Nice Chemicals Pvt. Ltd. Edappally, Kochi, Kerala. The chemicals used were of analytical grade.

Methodology

Preparation of grafts

Six fruit crops belonging to Sapotaceae family were taken as rootstocks (treatments) for softwood grafting in abiu viz., abiu *(Pouteria caimito)*, eggfruit *(Pouteria campechiana)*, sapota *(Manilkara sapota)*, khirni *(Manilkara hexandra)*, star apple (*Chrysophyllum cainito)* and wild star apple *(Chrysophyllum oliviforme)*. The rootstocks were raised from the seeds collected from firm ripe fruits which were sown in 5''x7'' sized polybags. Eight months old to one year old rootstocks were selected for the study. Healthy shoots of length 12-15cm having pencil thickness were collected from farmer's field, Thrissur and used as scion. Scion shoots were collected from mother trees in the early morning hours on the day of grafting and were wrapped in moist cloth and transported to the experimental site. To decontaminate the scion material from pests and diseases, the scion shoots were immersed in 0.1 percent Bavistin solution for 10 minutes. Grafting was performed on the same day of separation of scion from mother tree. The grafting process was carried out during the month of June and the resultant grafts were covered with narrow polythene covers after moistening and kept under shade to avoid desiccation.

Parameters observed

Growth parameters such as number of days taken for sprouting, number of leaves, number of branches, length of shoots and percentage of grafts survived were recorded at 90 days after grafting. The survival percentage of the grafts was calculated using the following equation.

$$\text{Survival percentage} = \frac{\text{Number of successful grafted plants}}{\text{Total number of grafted plants}} \times 100$$

Histological analysis

The anatomy of the graft union in softwood grafted plants were examined 90 days after grafting. Thin cross-sections of the graft union were prepared using a Leica sliding microtome, placed in water, and stained with safranin for 4-5 minutes. The sections were then dehydrated using a graded alcohol series (30% to 100%), followed by alcohol - xylene series (50:50) for 2 - 5 minutes and xylene (100 %) for 2- 5 minutes. It is then kept in xylene till the sections become clear and devoid of any air gaps. Finally, the sections were mounted on glass slides with DPX and observed under a stereo microscope at 4X magnification.

Statistical analysis

The data of different parameters recorded in the experiment were analyzed using analysis of variance (ANOVA) by Completely Randomized Design (CRD) tabulated at 5% level of significance. The statistical analysis was conducted using KAU GRAPES (General R based Analysis Platform Empowered by Statistics) software.

RESULTS AND DISCUSSION

Rootstocks play a crucial role in enabling the grafted plants to absorb water and nutrients, to produce hormones, and to store photosynthates. As a result of this, they can influence the overall growth of the tree and also the fruit yield both quantitatively and qualitatively [2]. Moreover, compatibility between the stock and scion is essential for forming a strong union and ensuring proper cambium formation. A histological study of grafts at various stages of grafting helps in understanding the physiology of graft union formation, as well as the compatibility reactions between the stock and scion [3].

Effect on scion growth parameters

The data on bud sprouting revealed that number of days to sprout ranged from 13.92 to 22.93 (Table 1). The rootstocks were found to significantly influence the number of days taken for initial sprouting. The earliest sprouting was recorded when rootstock of eggfruit (13.92) was used for grafting. This early sprouting observed in this treatment may be due to the food reserves present in the scion as these sprouts dried up after few weeks. The maximum number of days to sprout was shown when sapota was used as rootstock (22.93) followed by rootstock of star apple (21.77). When different rootstocks were used for grafting in sweet orange, the maximum number of days to sprout in pummelo was 16.5 days while minimum number of days to sprout recorded in rangpur lime were found to be 15.9 days [4]. The sprout survival is due to the rapid union between the xylem and cambium tissue of scion and rootstock [5]. The delayed sprouting may be due to inadequate flow of sap and low temperature conditions [6].

Growth parameters such as number of leaves, number of branches and length of shoots recorded after 90 days of grafting showed significant differences among the treatments. The treatment using abiu as rootstock was found to be significantly superior as the successful grafts recorded maximum number of leaves (18.03) as presented in (Table 1). Better leaf production (10.16) was observed in grafts when sapota was used as the rootstock, whereas all the other treatments failed to produce leaves as the sprouts dried off eventually. The production of more number of leaves can be attributed to good cambial contact between the stock and scion, positively influenced by ideal climate that promoted the cambial activity [7].

It was evident from (Table 1), after 90 days of grafting, the number of branches was observed to be the highest (5.18) in abiu rootstock. However other treatments did not show any branches as the sprouts got dried off after few days of growth except in sapota rootstock (3.14). In a study, when kalamansi (*Citrofotunella macrocarpa*) was used as the rootstock, it resulted in the maximum number of branches in mandarin and sweet orange, but when pummelo was used as the rootstock for pummelo itself, it produced the highest number of branches [8]. These findings further demonstrated the impact of different rootstocks on production of number of branches also.

Similarly, abiu rootstock recorded maximum length of shoot (8.29cm) likely due to greater number of leaves and branches indicating a successful union and growth. Sapota as rootstock also showed a shoot length of 4.73cm at 90 DAG while the rest of the treatments did not survive. The increased length of the shoot can be due to the early production of leaves which helped in more photosynthate production and the vigour of the rootstock species used [9]. The poor or slow growth rate of shoots might be due to less reserved food causing low physiological activity of the growing rootstock and scion.

Different rootstocks were found to have significant influence on graft survival percentage and the data related to survival percentage is presented in Table 1. The maximum percent survival was recorded for the abiu rootstock at 77.73%. However, sapota as a rootstock also demonstrated a substantial survival percentage, achieving 44.43%. Further studies to improve the success percentage of sapota rootstock has to be taken up by considering various factors such as age of scion and rootstock, season of grafting, growing conditions and so on.

Other rootstocks, other than abiu and sapota, though sprouted early, did not survive until the end of the experiment, indicating that these rootstocks are incompatible with abiu and therefore cannot be used as rootstocks for production of grafts in abiu. These findings are in line with the study by [10], in which the effect of various rootstocks on custard apple was examined and found that the highest successful grafts were produced using the chandsili rootstock. Similarly, an investigation on inter and intra-specific grafting in different *Annona* species reported highest survival percentages for the combinations *A. muricata* onto *A. muricata* (100%), *A. squamosa* onto *A. squamosa* (99.46%), and *A. reticulata* onto *A. muricata* (80.90%) [11].

Table 1 Number of days to sprout, number of leaves, number of branches, length of shoots and survival percentage as influenced by different rootstocks

Treatments (rootstocks)	Number of days to sprout	Number of leaves	Number of branches	Length of shoots (cm)	Survival percentage (%)
T_1: Abiu	19.02^{c}	18.03^{a}	5.18^{a}	8.29^{a}	77.73^{a}
T_2: Eggfruit	13.92^{e}	-	-	-	-
T_3: Sapota	22.93^{a}	10.16^{b}	3.4^{b}	4.73^{b}	44.43^{b}
T_4: Khirni	14.17^{e}	-	-	-	-
T_5: Star apple	21.77^{b}	-	-	-	-
T_6: Wild star apple	15.73^{d}	-	-	-	-
SE (m)	0.27	0.51	0.13	0.22	3.21
CD @ 5%	0.83	1.57	0.39	0.69	9.90

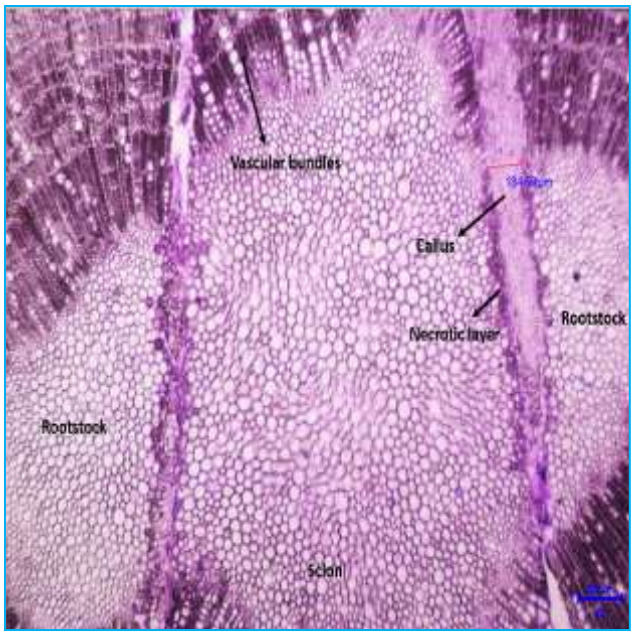

Fig 1 Horizontal sections from 90 days old samples of abiu grafted on abiu rootstock

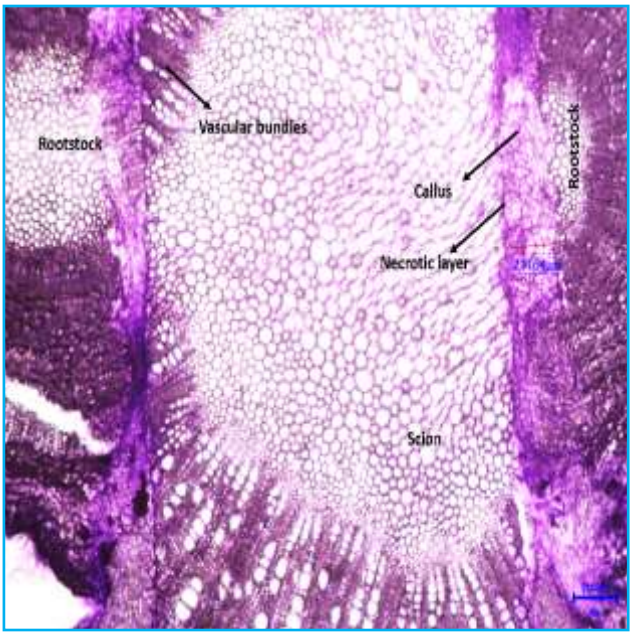

Fig 2 Horizontal sections from 90 days old samples of abiu grafted on sapota rootstock

Histological analysis

In the present study, abiu grafted onto abiu rootstock demonstrated well-established callus formation at the graft union. The thickness of the callus formed was observed to be 184.59 µm (Fig 1). The success of the abiu-abiu graft combination (homograft) may be attributed to the proper alignment of vascular bundles, sufficient callus formation at the graft junction, uniform thickness of both the scion and rootstock, and favourable biochemical, physiological, enzymatic and genetic factors. After three months of grafting, the graft union was predominantly filled with callus, with minor gaps that were expected to close over time. As the graft matured, the callus differentiated into vascular bundles, facilitating complete connectivity. In chestnut grafts, the callus tissue fully occupied the space between the grafted parts 60 days after grafting [12]. Although the adhesion line remains visible, it may disappear at a later stage. In unsuccessful grafts, a persistent necrotic layer may develop, hindering tissue continuity. This could be attributed to factors such as heat shock, adverse climatic conditions, or errors during the grafting process. Graft compatibility is more likely to occur between genetically similar (homogenetic) combinations, rather than genetically different (heterogenetic) ones [13] but lignification was completed earlier in homogenetic grafts compared to heterogenetic ones [14].

In abiu-sapota (heterografts), callus growth was observed from the rootstock but not from the scion, with the callus thickness measuring to 214.64µm (Fig 2). It was observed that the rootstock was in its active growth phase, characterized by a high number of meristematic cells, which promoted callus formation. But as the scion was more mature and thicker in size due to the onset of secondary growth, it led to the misalignment of the cambial ring hindering the rate of grafting success. This misalignment could be a key reason for the lesser success rate of abiu grafts using sapota rootstock in this study. This can be overcome by selecting scion and rootstock of similar age, size and thickness and ensuring proper cambial contact during the grafting process.

The physiological, anatomical, genetical and enzymatic compatibility between scion and rootstock determines the ultimate success and survival of the grafts. Endogenous plant hormones and long-distance signals (from shoot to root and root to shoot) play a critical role in regulating and synchronizing the growth of both above-ground organs and the root system.

Therefore, using different rootstocks which can influence the endogenous hormone signaling can lead to changes in scion growth parameters.

CONCLUSION

The findings of the present study clearly indicated that abiu and sapota can be effectively used as rootstocks for softwood grafting in abiu, as they demonstrated better survival and growth parameters compared to other rootstock species within the same family. The adoption of sapota as a rootstock for grafting in abiu can significantly reduce cost of cultivation, as raising sapota rootstocks is an economically viable option for farmers. Additionally, to improve the success rate of these rootstocks, further research is needed on factors such as the age and size of the scion and rootstock, the season and method of grafting, as well as growing conditions.

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