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Standardization of softwood grafting techniques in sapota (*Manilkara achras* L.) on invigorated khirni rootstock under polyhouse and shade net conditions

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Abstract
Propagation studies in sapota with softwood grafting on invigorated khirni rootstocks were carried out with use of two length of scion (15 and 10 cm), under two propagation conditions (polyhouse and shade net) and in 4 different month of grafting (May, June, July and August). Growth of the grafted plant was recorded in 30 days intervals up to 120 days and there was increase in the growth parameters like number of leaves, height of the scion and number of sprouts per grafted plant had increased with the increase in days after grafting. In relevance to growth parameters like early sprouting, maximum number of leaves was recorded in grafts when they were made using 10 cm scion, propagated under polyhouse and grafted during the month of July. Maximum number of sprouts per grafted plant was observed in the month of May. The maximum growth in terms of height (above the union) were recorded in grafts made with use of 15 cm scion, propagated under polyhouse and grafted during the month of July.

Keywords: *Manilkara achras* L., khirni rootstock, polyhouse, shade net

Introduction
Sapota, (*Manilkara achras* L.) is one of the most important tropical fruit crop. It is native to South Mexico as well as North East Guatemala. It is also called by the name *Chickoo*, is an evergreen tree known for its fruits of delicate flavor, melting pulp with sweet taste. It produces crop continuously through out the year in warm and moist tropical climate. It is hardy, highly productive and generally free from major pests, diseases and physiological disorders. Hence, it has emerged as an important fruit crop and widely cultivated in India. Sapota is commonly propagated by approach method of grafting. Though it is hugely successful method but it is painstakingly laborious. However, their is an alternative method of propagation called softwood grafting method which is easier and successful. Khirni (*Manilkara hexandra*) is the rootstock used for grafting in sapota. This rootstock is not easily available. Depending on various factors. Success rate of graft varies. Quite a large number of rootstocks are wasted. Hence, this study aims at reusing of the khirni rootstocks of unsuccessful grafts. The scions would be grafted on to new flush of shoots borne on left over rootstock and these rootstocks are called as invigorated rootstocks.

Material and methods
The study was carried out at Department of Horticulture, College of Agriculture, University of Agricultural Science, GKVK., Bengaluru, Karnataka during 2016. The 3 year old invigorated khirni rootstocks were used for the experiment. The experiment was laid out in split-split plot design with moth(May, June, July and August) of grafting as main treatment, conditions (Polyhouse and Shade net) as sub treatment, length of scion(15 and 10cm) as sub sub treatment and with 3 number of replication. Days taken for first sprouting was recorded and different growth parameters like number of sprouts, number of leaves per graft and Height of the scion was observed at 30 days interval upto 120 days.

Results and discussion
Days taken for first sprouting: Among 4 months significantly lesser number of days for first sprouting was recorded in July (19.14 days), highest days were recorded in August (21.42 days). It is due to congenial climatic condition prevailing during July. The similar kind of findings were recorded by Prasanth *et al*. (2006) [8] on epicotyl grafting in mango results in

early sprouting in the first fortnight of July. Significantly lesser number of days for first sprouting was in the grafts under polyhouse (18.53 days) compared to shade net (21.54 days). It is obtained due to controlled condition prevailing the higher relative humidity and temperature inside the polyhouse when compared with the shade net condition. A similar kind of opinion was expressed by Gurjar and Singh (2012) [1] in aonla and by Patel *et al*. (2010) [6] in khasi mandarin under polyhouse for early sprouting. Significantly lesser number of days for first sprouting was recorded with the use of 10 cm scion (19.43 days) compared to 15 cm scion (20.64 days).In longer scions, late sprouting due to reduced chances of quick formation of callus from live parenchymatous cells (Table 1).

Number of sprouts per graft

Number of sprouts was significantly differed at 60, 90 and 120 DAG. At 60 DAG higher number of sprouts per graft were recorded in May (1.47), followed by July (1.11) and least in August (1.00). At 90 DAG higher number of sprouts per graft were observed in May (1.56), followed by July and June (1.14 in both the month) and lower in August (1.00). And at 120 DAG higher number of sprouts per graft were observed in May (1.56), followed by July (1.42) and least in August (1.00). This may be attributed to moderately high temperature coupled with high humidity, adequate supply of healthy and matured scion sticks because the mother trees resume the active growth phase at the onset of monsoon with adequate supply of moisture and nutrients, fast cambial activity and high accumulation of carbohydrates in scion shoots and the sprouting of dormant bud observed during May month which is contributed for the increased number of sprouts.

No significant differences were observed with growing condition and with use of different length of scion.

Number of leaves per graft: The maximum number of leaves was observed in July (3.53, 6.92, 11.75 and 15.53), closely followed by May (3.34, 6.89, 11.47 and 15.44) and minimum in August (1.58, 3.61, 6.81 and 9.94) at 30, 60, 90 and 120 DAG respectively. This may be due to the early sprouting of buds giving quick growth in the form of more number of leaves. Similar kind of results was reported by Patil *et al.* (2010) [7], in sapotaduring the month of July had shown maximum number of leaves on scion shoot on new khirni rootstock (15.08) and compared with the invigorated khirni rootstock (14.83).

Maximum number of leaves per graft was observed under polyhouse (3.13, 6.19, 10.60 and 14.17) compared to grafts under shade net (2.22, 4.97, 8.79 and 12.42) at 30, 60, 90 and 120 DAG respectively. This may be due to favourable condition prevailing inside the polyhouse stimulating rapid callusing and early contact of cambial layers, which enables the graft to heal quickly and make a strong union ultimately leading to better strength and faster growth of grafts by producing more number of leaves. Similarly, Raghavendra *et al*., (2011) [9] recorded in wood apple that, significantly more number of leaves under ploy mist house condition compared to open condition.

Use of 10 cm scion had shown maximum number of leaves (3.04, 6.35, 10.57 and 14.49) and the minimum with the use of 15 cm scion (2.31, 4.82, 8.82 and 12.10) at 30, 60, 90 and 120 DAG respectively. This may be due to long scion sticks are woody in nature and the union becomes difficult because of poor division of parenchymatous cells and thus poor callus formation. This might have led to poor translocation of water and nutrient. Similar kind of finding was reported by Nalage *et al*. (2010) [4] in epicotyl grafting of mango with the use of 10 cm scion.

Height of the scion: Height of the graft above the union (cm) was observed significantly higher in July (13.89, 15.19, 16.49 and 17.68) and the lower height was observed in August (13.03, 13.56, 14.37 and 15.24) at 30, 60, 90 and 120 DAG maximum number of leaves produced in the particular month had influenced the height of scion. Similar findings were reported by Patil *et al*. (2010) [7] in softwood grafting of sapota in the month of July, with new khirni rootstock (16.06 cm) and with invigorated khirni rootstock (17.47 cm). Pampanna and Sulikeri (2000) [5] observed maximum height of scion shoots in the month of May in softwood grafting of sapota.

Maximum height (cm) of graft was recorded under polyhouse (13.51, 14.45, 15.54 and 16.69) compared to shade net (13.32, 14.01, 14.99 and 15.89) at 30, 60, 90 and 120 DAG respectively. It is due to higher humidity and higher temperature inside the polyhouse leads to higher metabolic activity of cells and thus leads to extension of growth of scion. Similar kind of results were reported by Patel *et al*. (2010) [6], maximum height of grafts under polyhouse (32.05 cm) when compared to net house (27.35 cm) in softwood grafting of kashi mandarin.

Maximum graft height (cm) was recorded with the use of 15 cm scion (16.08, 17.09, 18.15 and 19.29) compared to 10 cm scion (10.75, 11.37, 12.39 and 13.34) at 30, 60, 90 and 120 DAG respectively. This is due to selection of different length of scion for our experimental study only directly impacted the height of scion. The longer scion sticks made linear growth of grafts. Similarly Nalage *et al.* (2010) [4] reported maximum growth in terms of height was recorded with the use of longer scions.

Table 1: Effect of different months, growing condition and length of scion on days taken for first sprouting, number of sprouts, number of leaves / graft and height of graft at 30, 60, 90 and 120 DAG.

Treatments		Days taken for first sprouting	Number of sprouts / graft				Number of leaves / graft at				Height of graft (cm) at			
			30 DAG	60 DAG	90 DAG	120 DAG	30 DAG	60 DAG	90 DAG	120 DAG	30 DAG	60 DAG	90 DAG	120 DAG
Months (M)														
M_1	May	19.28	1.19	1.47	1.56	1.56	3.34	6.89	11.47	15.44	13.45	14.36	15.49	16.49
M_2	June	20.31	1.06	1.08	1.14	1.14	2.25	4.92	8.75	12.25	13.28	13.82	14.79	15.73
M_3	July	19.14	1.06	1.11	1.14	1.42	3.53	6.92	11.75	15.53	13.89	15.19	16.49	17.68
M_4	August	21.42	1.00	1.00	1.00	1.00	1.58	3.61	6.81	9.94	13.03	13.56	14.37	15.24
F-test ($p \leq 0.05$)		*	NS	*	*	*	*	*	*	*	*	*	*	*
SEm±		0.277	0.053	0.062	0.068	0.098	0.166	0.313	0.34	0.477	0.041	0.152	0.247	0.358
C.D. at 5%		0.956	-	0.213	0.236	0.34	0.573	1.081	1.175	1.645	0.142	0.525	0.851	1.234
Conditions (C)														
C_1	Polyhouse	18.53	1.04	1.18	1.21	1.28	3.13	6.19	10.60	14.17	13.51	14.45	15.54	16.69
C_2	Shade net	21.54	1.11	1.15	1.21	1.28	2.22	4.97	8.79	12.42	13.32	14.01	14.99	15.89

F-test ($p \leq 0.05$)		*	NS	NS	NS	NS	*	*	*	*	*	*	*	*
SEm±		0.27	0.034	0.05	0.064	0.125	0.109	0.176	0.276	0.399	0.034	0.068	0.089	0.154
C.D. at 5%		0.88	-	-	-	-	0.356	0.572	0.898	1.3	0.11	0.221	0.29	0.501
Length of Scion (L)														
L_1	15cm	20.64	1.10	1.21	1.25	1.28	2.31	4.82	8.82	12.10	16.08	17.09	18.15	19.29
L_2	10 cm	19.43	1.06	1.13	1.17	1.28	3.04	6.35	10.57	14.49	10.75	11.37	12.39	13.34
F-test ($p \leq 0.05$)		*	NS	NS	NS	NS	*	*	*	*	*	*	*	*
SEm±		0.194	0.046	0.064	0.072	0.072	0.101	0.158	0.245	0.261	0.034	0.127	0.226	0.295
C.D. at 5%		0.581	-	-	-	-	0.304	0.474	0.735	0.783	0.101	0.379	0.677	0.884

Table 2: Effect of different months, growing condition and length of scion on initial graft success (%) at 30 DAG and number of days taken for first sprouting

Treatments		Graft success (%)	Graft survival (%)
Months (M)			
M_1	May	75.83	67.50
M_2	June	50.00	35.83
M_3	July	80.00	70.00
M_4	August	41.67	28.33
F-test ($p \leq 0.05$)		*	*
SEm±		2.152	0.867
C.D. at 5%		7.426	2.994
Conditions (C)			
C_1	Polyhouse	74.58	61.25
C_2	Shade net	49.17	39.58
F-test ($p \leq 0.05$)		*	*
SEm±		3.062	1.863
C.D. at 5%		9.973	6.069
Length of Scion (L)			
L_1	15cm	57.50	46.25
L_2	10 cm	66.25	54.58
F-test ($p \leq 0.05$)		*	*
SEm±		2.465	2.517
C.D. at 5%		7.391	7.548

DAG= Days after grafting. NS = Non-significant. *= Significant.

It is seen that the initial graft success rate was highest in the month of July (80.00%), under polyhose (74.58%) and with the use of 10 cm scion (66.25%). It is due to the optimum temperature and higher relative humidity, which are the two major factors which plays a greater role in the graft union formation. Higher humidity prevent scion from desiccation and thus helps in maintaining cell turgidity and help in quick callus formation between stock and scion during July month and also under the polyhouse. Similarly Ghosh *et al.* (2010) [2] noticed, highest graft success during July (72%) in softwood grafting of sapota, Sulikeri *et al.* (1997) [11] with higher graft success (80%) in sapota under mist house condition. In 10 cm scion sticks, smaller area of transpiration as well as thin cell wall of scion sticks which may have involved in better callus formation and graft take compared to 15 cm scions. Similar kind of result was reported by Nalage *et al*. (2010) [4] in mango (Table 2).

It is seen that highest graft survival was seen in the month of July (70.00%), under polyhouse (61.25%) and with the use of 10 cm scion (54.58%). During July month and under polyhouse, the moderate temperature and higher relative humidity helps in higher metabolic activity of the cells thus activates the cambium cell development. The new callus tissue arising out of the cambial region is composed of thin walled turgid cells which can easily desiccate and die off and relative humidity can protect such cells in the cambial region of the graft union (Hartmann *et al*. 2007) [3]. Similarly, Singh and Bons (2016) [10] reported maximum graft survival (62.8%) during the month of July in sapota. In 10 cm scions higher survival is due to production of new xylem and phloem permits the vascular connection between the scion and the rootstock. The enlarging leaf surfaces on the scion shoots has little or no water to offset that lost by transpiration and the scion quickly become desiccated and die and thus causing low survival (Hartmann *et al.*, 2007) [3]. Similarly Nalage *et al.* (2010) [4] reported higher survival with the use of 10 cm long scion sticks in mango.

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