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Study on the effect of height of rootstock and length of scion stick on success of epicotyl grafting in mango (*Mangifera indica* L.) cv. kesar

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Abstract
The investigation was carried out with twelve treatments comprising four heights (4, 6, 8 and 10 cm) of rootstocks and three lengths (10, 15 and 20 cm) of scion sticks. The maximum number of sprouted grafts, maximum sprouting percentage, minimum days for leaf emergence, maximum number of leaves per graft, girth (above the union), minimum mortality (%) and maximum survival (%) of grafts were recorded when grafts were made on 6 cm height of rootstock and 10 cm long of scion stick. Consequently, the maximum growth in terms of height and girth (below the union) were recorded in grafts made on 10 cm height of rootstock and 20 cm long scion stick of mango cv. Kesar.

Keywords: height of rootstock, length of scion stick, epicotyl grafting, mango and Kesar

Introduction
Mango (*Mangifera indica* L.) is considered as king of fruits. It is the national fruit of our country and seems to be under cultivation well over 400 years. Kesar can be the best variety for export in the form of fresh and processed product. Demand for this variety is increasing day by day, therefore land under Kesar cultivation is also increasing, which will certainly increase the production. However the pace of development is not very fast. The major hurdle in rapid expansion of mango industry is the shortage of quality mango grafts for new plantation. For the expansion of mango fruit industries, availability of true to type planting material in adequate quantity is a pre-requisite and for this standardization of propagation technique is also necessary.

In recent years, the epicotyl grafting, technique can be adopted for large scale multiplication of mango, being a faster method compare to inarching or veneer grafting. The plants produced by epicotyl grafting are found to be uniform, healthy and requiring less area for propagation and ultimately cheaper in cost. The cost of production of seedling rootstock is minimized to a great extent. Also it is simple, easy and rapid method. The research work carried out during the last few years have clearly shown that this technique can be adopted for large- scale multiplication of mango in this region. Epicotyl grafting method is being used on a commercial scale by Dr. B. S. Konkan Krishi Vidyapeeth, Dapoli, Ratnagiri and the farmers in Konkan region are also using this technique on wider scale.

Materials and Methods
Raising of seedlings for Rootstock
Seed stones of mango cv. Kesar were collected from processing factory. They were sown in line on the flat beds in flat position at 10 X 2.5 cm distance and were covered with 5 cm thick layer of farmyard manure and soil in the ratio of 1:1. Germinating stones from which the epicotyl was just emerged and straightened out were selected as rootstocks. The healthy vigorous seedlings with straight and stout epicotyl were uprooted along with seed stones without causing much injury to the roots.

Rootstocks
The healthy uniform mango seedlings free from pest and diseases were selected and sorted as per different heights i.e. H_1-4 cm, H_2-6 cm, H_3-8 cm and H_4-10 cm from the base of stock for propagation purpose.

Scion sticks

Defoliation was carried out to the selected scion sticks seven days prior to grafting process. These defoliated scion sticks were collected and stored with care until grafting. The healthy scion sticks were taken for further procedure and length of scion stick was 10, 15 and 20 cm. The cut was made according to the length and grafting was done on the same day.

Operation of epicotyl grafting

The epicotyl grafting was done by wedge technique of grafting, as described by Bhan *et al.* (1969). In this method, vertical cut of 3 cm length was given onto deheaded epicotyl of germinated seed so as to fit the wedge shape scion. Then the scion of comparative thickness was made like wedge by giving slanting cut of 3 cm length on opposite sides. The wedge shaped scion was inserted into the slit of the epicotyl of the stock. The joint was tied with polythene stripe of 200 gauges. The grafts were observed for initial success (sprouting) at 45, 60, 75 and 90 days of age of grafts. The observations on survival percentage were recorded at after 90 days of grafting. The height, girth (above and below the union) and total number of leaves were recorded one month interval.

Results and Discussion

Height of rootstock

There was significant difference in number of sprouted grafts and maximum sprouting percentage of grafts when they were made on different height of rootstocks and periodically observed at 45, 60, 75 and 90 days of grafts (Table.1). The higher number of sprouted grafts and maximum sprouting percentage of grafts were recorded at 45, 60, 75 and 90 days of grafting, respectively, when grafts made on 6 cm height of rootstock (H_2). While lower number of sprouted grafts and sprouting percentage of grafts were recorded when grafts made on 4 cm height of rootstock (H_1) at 45, 60, 75 and 90 days of grafting, respectively.

Early callus formation occurred mainly from the rootstock with cells produced in definite rows, often in fan-like array. The establishment of the cambial bridge between the stock and scion was followed by the formation of a protective layer, the periderm across the callus edges (Asante and Barnett, 1997) [1]. Success in grafting depends upon good cambial bridge between stock and scion may result into better callus formation and union. Hence, maximum numbers of sprouted grafts were recorded in mango cv. Kesar, have been recorded in this study. The results obtained by Kanwar and Bajwa (1974) [5], Patel and Amin (1976) [7], Chakrabarti and Sadhu (1984) [2], Patil *et al*. (1984) [8], Ratan *et al*. (1987) [12] and Reddy and Kohli (1989) [13] found the same trend while working on mango as well as other fruit crops. The results are in conformity with the present findings.

Data presented in Table 2 clearly revealed that total numbers of leaves per graft were significantly influenced by various heights of rootstocks at periodical interval of growth stages of grafts. Mango grafts made on 6 cm height of rootstock (H_2) had the maximum number of leaves per graft consistently with 10.88, 17.02, 24.45 and 28.94 at 1, 2, 3 and 4 months interval, respectively.

The rapid division of parenchymatous cells of cambium of rootstock within 2-3 days of grafting, which later on interlocked with parenchyma cells of scion and partly produced the limited passage within short period for translocation of water and nutrients, which might have resulted into production of new leaves. Further, the total number of leaves increased periodically indicated good functional activity of xylem and phloem. Patil *et al*. (1984) [8] and Patil *et al*. (1994) [8] also found the similar findings in mango and jackfruit which are in agreement of the results of present study.

The data pertaining to the height of epicotyl graft and girth (above and below the union) as influenced by the use of different height of rootstocks presented in Table 2 and table 3. Significantly more height and girth (below the union) of grafts were recorded when grafts were made on 10 cm height of rootstock. The minimum height and girth (above and below the union) was recorded when grafts were made on 4 cm height of rootstock. The data clearly revealed that the height of graft was significantly influenced when grafts were made on 10 cm height of rootstocks (H_4) at all the age of grafts i.e. 1, 2, 3 and 4 months recording 25.65, 27.68, 29.73 and 33.00 cm, respectively. On the other hand, minimum height of graft was recorded as 20.25, 22.25, 24.43 and 27.72 cm at one month interval respectively, when grafts were made on 4 cm height of rootstocks (H_1). The data of present research work are similar to those of Chakrabarti and Sadhu (1984) [2] and Patil *et al.* (1984) [8], while working on mango.

The temperature and humidity during July-August were seemed to be congenial for growth and also sap flow condition might be higher during these periods which led faster growth of scion shoots. The temperature and humidity during later part were low due to which, minimum growth might have taken by grafts. The gradual increase in girth periodically i.e. at one month interval because of the increase in leaf number and leaf area resulting into synthesis of photosynthates, which translocates towords the union. However, poor translocation may have occurred because of the narrow passage in newly differentiated vascular tissues at union portion. This might have resulted into increase in girth above the union than that of below the union.

The data presented in Table 4 revealed that there were significant differences in days required for leaf emergence in grafts, survival percentage of grafts and mortality percentage of grafts due to different height of rootstock. When the grafts made on 6 cm height of rootstocks (H_2), they took significantly the minimum days (15.65) for leaf emergence, high survival percentage (55.00) and minimum mortality percentage (19.17) per grafts.

Callus formation is pre-requisite for successful formation of graft union. New parenchymatous callus proliferates in one to seven days from both the rootstock and scion. However, the stocks produced most of the callus. This perhaps may due to absorption of water and nutrient by rootstock initiating, involvement in rapid cell division of parenchymatous cells. These parenchyma cells comprising the spongy callus tissue, penetrates the thin necrotic layer within two to three days and fill the space between the two components of the grafts (scion and stock), becoming initially interlocked and providing some mechanical support as well as allows for limited passage of water and nutrient between the stock and scion (Hartman and Kester, 2002) [4] due to which early sprouting may have taken place in epicotyl grafting. Patel and Amin (1976) [7], Ratan *et.al.* (1987) [12] and Radha and Aravindakshan (1998) [10].

The data presented in Table 4 showed that height of rootstock did not exert any significant effect on leaf area.

The present findings are consonance with Epicotyl grafting is generally done by uprooting seedlings and after grafting, grafted seedling is again planted in polythene bags. The well-established root system get disturbed at the time of uprooting

and transplanting, such uprooted seedlings required more time for establishment, such type of injury or damage to root system might be responsible for lower survival percentage. While in grafted plant at 6 cm height of rootstock helped to minimize the shock and ensure better survival and minimum mortality of the epicotyl grafts.

Grafting at 4 cm height of rootstock resulted into good initial sprouting, but later on failed to survive inspite of necessary cares taken at nursery level. Higher humidity during the monsoon months of heavy rain in this south Gujarat agro-climatic condition might have initiated the incidence of infection either at union or collar level because of delicate tissues.

Table 1: Effect of height of rootstock and length of scion on number of sprouted grafts and sprouting percentage per treatment at 45, 60, 75 and 90 days

Treatments	Number of sprouted grafts per treatment at 15 days interval				sprouting percentage of grafts at 15 days interval			
	45	60	75	90	45	60	75	90
Height of rootstock (H)								
H_1	12.00	10.08	8.92	7.92	60.00 (50.75)	50.42 (45.22)	44.58 (41.87)	39.58 (38.97)
H_2	14.83	12.83	11.67	11.00	74.17 (59.43)	64.17 (53.21)	58.33 (49.77)	55.00 (47.85)
H_3	14.08	12.25	11.00	10.17	70.42 (57.03)	61.25 (51.48)	55.00 (47.85)	50.83 (45.46)
H_4	13.17	11.00	9.83	9.17	65.83 (54.20)	55.00 (47.85)	49.17 (44.50)	45.83 (42.59)
S.Em.±	0.26	0.28	0.29	0.24	0.41	0.41	0.41	0.41
C.D. at 5%	0.75	0.79	0.82	0.70	1.18	1.18	1.18	1.18
Length of scion (L)								
L_1	15.56	13.75	12.56	11.88	77.81 (61.87)	68.75 (55.59)	62.81 (52.40)	59.38 (50.38)
L_2	14.38	12.56	11.38	10.50	71.88 (57.95)	62.81 (52.40)	56.88 (48.93)	52.50 (46.41)
L_3	10.63	8.31	7.13	6.31	53.13 (46.77)	41.56 (40.12)	35.63 (36.63)	31.56 (34.16)
S.Em.±	0.23	0.24	0.25	0.21	0.36	0.36	0.36	0.36
C.D. at 5%	0.65	0.69	0.71	0.60	1.02	1.02	1.02	1.02
Interaction effect (H X L)								
S.Em.±	0.45	0.48	0.50	0.42	0.71	0.71	0.71	0.71
C.D. at 5%	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
C.V. %	6.72	8.33	9.62	8.82	6.63	7.22	7.66	7.99

***** Values in parenthesis are arcsine transformed values

Table 2: Effect of height of rootstock and length of scion on total number of leaves per graft and height of grafts at one month interval

Treatments	Total number of leaves at one month interval				Height (cm) of grafts at one month interval			
	1 month	2 month	3 month	4 month	1 month	2 month	3 month	4 month
Height of rootstock (H)								
H_1	8.93	14.79	21.00	25.24	20.25	22.25	24.43	27.72
H_2	10.88	17.02	24.45	28.94	21.82	23.63	25.93	29.37
H_3	9.97	16.09	23.10	27.53	23.53	25.62	27.77	31.18
H_4	9.52	15.67	22.05	26.35	25.65	27.68	29.73	33.00
S.Em.±	0.08	0.14	0.12	0.17	0.17	0.20	0.17	0.15
C.D. at 5%	0.23	0.41	0.35	0.48	0.49	0.56	0.48	0.42
Length of scion (L)								
L_1	10.10	16.22	23.07	27.46	18.14	20.14	22.34	25.76
L_2	9.83	15.86	22.61	26.95	22.69	24.64	26.94	30.36
L_3	9.55	15.59	22.27	26.63	27.61	29.61	31.78	34.83
S.Em.±	0.07	0.12	0.11	0.14	0.15	0.17	0.15	0.13
C.D. at 5%	0.21	0.35	0.30	0.41	0.42	0.48	0.42	0.37
Interaction effect (H X L)								
S.Em.±	0.14	0.25	0.21	0.29	0.30	0.34	0.29	0.26
C.D. at 5%	N.S.	N.S.	N.S.	N.S.	0.85	0.97	0.83	0.73
C.V. %	2.95	3.11	1.87	2.15	2.60	2.72	2.17	1.69

Table 3: Effect of height of rootstock and length of scion on girth of grafts at one month interval

Treatments	Girth (cm) of grafts at one month interval							
	Above the union				Below the union			
	1 month	2 month	3 month	4 month	1 month	2 month	3 month	4 month
Height of rootstock (H)								
H_1	2.48	2.50	2.56	2.60	1.54	1.56	1.62	1.67
H_2	2.63	2.65	2.71	2.76	1.58	1.61	1.66	1.71
H_3	2.58	2.61	2.66	2.72	1.62	1.64	1.71	1.78
H_4	2.55	2.58	2.63	2.70	1.66	1.69	1.76	1.82
S.Em.±	0.02	0.02	0.02	0.01	0.01	0.01	0.01	0.01
C.D. at 5%	0.05	0.05	0.04	0.04	0.03	0.03	0.03	0.02
Length of scion (L)								
L_1	2.53	2.55	2.61	2.66	1.58	1.61	1.67	1.73
L_2	2.56	2.59	2.64	2.69	1.60	1.62	1.69	1.74
L_3	2.58	2.61	2.67	2.73	1.61	1.64	1.70	1.76
S.Em.±	0.02	0.01	0.01	0.01	0.01	0.01	0.01	0.01

C.D. at 5%	0.04	0.04	0.03	0.04	N.S.	N.S.	N.S.	N.S.
Interaction effect (H X L)								
S.Em.±	0.03	0.03	0.03	0.02	0.02	0.02	0.02	0.02
C.D. at 5%	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
C.V. %	2.44	2.28	2.04	1.82	2.38	2.03	1.99	1.73

Table 4: Effect of height of rootstock and length of scion on days required for leaf emergence, leaf area (cm^2), survival and mortality percentage of grafts.

Treatments	days required for leaf emergence	leaf area (cm^2)	Survival (%) of grafts per treatments	Mortality(%) of grafts per treatments before 45 days	Mortality (%) of grafts per treatments after 90 days
Height of rootstock (H)					
H_1	16.43	254.16	39.58 (38.97)	40.00 (39.21)	20.42 (26.85)
H_2	15.65	261.50	55.00 (47.85)	25.83 (30.53)	19.17 (25.95)
H_3	15.86	267.93	50.83 (45.46)	29.59 (32.94)	19.58 (26.25)
H_4	16.22	275.19	45.83 (42.59)	34.17 (35.76)	20.00 (26.55)
S.Em.±	0.09	5.37	0.41	0.41	0.41
C.D. at 5%	0.25	N.S.	1.18	1.18	1.18
Length of scion (L)					
L_1	15.54	262.21	59.38 (50.38)	22.18 (28.08)	18.44 (25.42)
L_2	15.81	264.50	52.50 (46.41)	28.22 (32.07)	19.38 (26.11)
L_3	16.78	267.38	31.56 (34.16)	46.88 (43.19)	21.56 (27.65)
S.Em.±	0.08	4.65	0.36	0.36	0.36
C.D. at 5%	0.22	N.S.	1.02	1.02	1.02
Interaction effect (H X L)					
S.Em.±	0.15	9.31	0.71	0.71	0.71
C.D. at 5%	N.S.	N.S.	N.S.	N.S.	N.S.
C.V. %	1.89	7.03	7.99	8.34	12.45

***** Values in parenthesis are arcsine transformed values

Length of scion stick

From the same table 1, it is seen that the highest numbers of sprouted grafts and maximum sprouting percentage of grafts were recorded at 45, 60, 75 and 90 days after grafting when scion sticks of 10 cm length (L_1) were used for grafting. The lowest numbers of sprouted grafts and sprouting percentage of grafts were recorded at 45, 60, 75 and 90 days of grafts, respectively, when scion sticks of 20 cm length (L_3) were used for grafting.

Shorter scion sticks proved very effective to obtain maximum sprouting as compared to longer scion sticks. Precured scion sticks (bud wood) and smaller area of transpiration as well as thin cell wall of bud sticks which may have involved in better callus formation and graft take. Early callus formation occurs from the rootstock component producing good union (Asante and Barnett, 1997) [1] which supports the present findings. Thus it may have resulted into higher sprouting percentage of grafting. The results obtained by Kanwar and Bajwa (1974) [5], Patel and Amin (1976) [7], Chakrabarti and Sadhu (1984) [2], Patil *et al*. (1984) [8], and Reddy and Kohli (1989) [13] found the same trend while working on mango as well as other fruit crops. The results are in conformity with the present findings. Likewise, longer bud sticks may contain thick cell walls and more woody portion resulting into poor establishment of cambial bridge and there by poor graft-take and sprouting percentage.

It is seen from the Table 2 that the significantly more number of leaves per graft were recorded as 10.10, 16.22, 23.07 and 27.46 at 1, 2, 3 and 4 month age of graft, respectively, when 10 cm long scion sticks (L_1) were used for grafting. On the other hand, the minimum numbers of leaves per grafts were recorded as 9.55, 15.59, 22.27 and 26.63 at monthly interval when scion sticks of 20 cm length (L_3) were used for grafting. Long scion sticks are woody in nature and the union becomes difficult because of poor division of parenchymatous cells and callus formation. This might have led to poor translocation of water and nutrient through very narrow passage. The similar result were obtained by Seshadri and Rao (1985) [14] and Gurudutta *et al.* (2004) [3] in cashew and mango, respectively.

According to Zimmerman (1958) [16] maximum number of leaves were recorded when fresh scion sticks were used for epicotyl grafting which may be due to more stored carbohydrates and other food substances available in the scion sticks after defoliation leads to more growth in terms of number of leaves. The length of scion sticks also showed significant effect on height of grafts in all stages of growth. Significantly the maximum height of grafts were recorded as 27.61, 29.61, 31.78 and 34.83 cm of epicotyl grafts when scion sticks 20 cm length (L_3) were used for grafting, while minimum height of epicotyl grafts were noted periodically at 1, 2, 3 and 4 month age as 18.14, 20.14, 22.34 and 27.56 cm, respectively, when 10 cm length of scion stick (L_1) was used.

Looking to the data presented in Table 3, it is revealed that the maximum girth (above and below the union) of graft was noted when scion sticks of 20 cm length was used for grafting at 1, 2, 3 and 4 month old grafts, respectively. This treatment was at par with L_2 treatment at all the age of grafts. The minimum girth was recorded at periodical interval when 10 cm long scion sticks (L_1) were used for epicotyl grafting. The similar results were obtained by Kanwar and Bajwa (1974) [5], Chakrabarti and Sadhu (1984) [2], Seshadri and Rao (1985) [14] and Gurudutta *et al.* (2004) [3].

The height and girth of grafts increased significantly with an increase in the length of the scion sticks in grafts. The longer scion sticks made linear growth of grafts.

The temperature and humidity during July-August were seemed to be congenial for growth and also sap flow condition might be higher during these periods which led faster growth of scion shoots. The temperature and humidity during later part were low due to which, minimum growth might have taken by grafts.

The data presented in Table 4 revealed that there were significant differences in days required for leaf emergence, survival percentage and mortality percentage of grafts when different length of scion sticks were used during the course of

study. The minimum days (15.54) for leaf emergences, maximum survival percentage (59.38) and minimum mortality percentage (18.44) were significantly recorded when 10 cm length of scion sticks (L_1) were used for epicotyl grafting. The least time consumed in the sprouting may be due to abundant supply of carbohydrate and defoliation which initiates bud activation and they are in a position to sprout early (Zimmerman, 1958) [16]. The results obtained by Purbiati *et al.* (1991), Ratan *et al.* (1987) [12], Radhamony *et al.* (1989) and Radha and Aravindakshan (1998) [10] and Gurudutta *et al.* (2004) [3] are in consonance to the present study.

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 and Kester, 2002) [4]. The gradual decline in the survival of epicotyl grafts of mango cv. Kesar is reported with the passage of time, i.e. at 15 days interval. High humidity accompanied by high temperature during the months of August end and September might have caused tissue injury and death of callus cells. In longer scions and rootstocks reduced the chances of quick formation of callus from live parenchymatous cells. Singh and Shrivastava (1981) also reported the decrease in survival when the age of seedling increased. Kulwal and Tayde (1989) and Asante and Barnett (1998) [1] also reported the similar findings which are in agreement with the present findings.

The effect of length of scion stick on leaf area of graft was found non-significant.

Summary and Conclusion

Based on the present investigation on, "Effect of height of rootstock and length of scion stick on success of epicotyl grafting in mango (*Mangifera indica* L.) cv. Kesar", it can be concluded that epicotyl grafts can be made successfully on 6 cm height of rootstock and 10 cm long scion stick. The maximum number of sprouted grafts, maximum sprouting percentage, minimum days taken for leaf emergence, maximum number of leaves per graft, minimum mortality and maximum survival (55 % after 90 days of grafting). Whereas, height and girth (below the union) of grafts were recorded when grafts were made on 10 cm height of rootstock with use of 20 cm long scion sticks in epicotyl grafts of mango cv. Kesar.

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