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Quantification of Microclimate of Guava Wedge Grafting Success on Different Age of Rootstocks under Different Growing Environment

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ABSTRACT: An experiment was carried out to find the effect of wedge grafting success on different age of rootstock under different growing environment at CCS Haryana Agricultural University, Hisar during the year 2016-17 & 2017-18. The experiment was laid out in CRD with nine treatments and three replications to find out the suitable age for successful wedge grafting with polycap of guava rootstock cv. L-49 under different growing conditions (Open condition, Shade net 50% and Greenhouse) with different age of rootstock (8, 12 and 16 months). The results revealed that under greenhouse (temp. - 25±5ºC, Relative humidity - 70-80%) more than 12 month old rootstock takes minimum days were required for successful grafting, maximum successful grafting percentage, max. number of sprouts on rootstock, maximum number of leaves on scion and max. increase in scion length (cm), leaf area, fresh and dry weight of shoot-root followed by shade net at 8 months. This might be due to humid climatic condition played an important role in early contact of cambium layer of stock and scion resulting in easy callus formation and initiation of subsequent growth and less fluctuation between maximum and minimum temperature contributes to high success of graft union and further growth of grafts. In the experiment, the interaction between growing conditions and period after grafting was found significant. Present results suggest that highest number of spourts on rootstock, successful grafting percentage, number of sprouts on rootstock, number of leaves on scion, increase in scion length (cm), leaf area, fresh and dry weight were found best under greenhouse on 12 month old seedling followed by shade net 50%.

Keywords: Guava, Rootstock age, Greenhouse, shade net, Wedge grafting.

INTRODUCTION

Guava (*Psidium guajava* L.), which belongs to the family Myrtaceae, is one of the most important commercial fruit crops of India. Although it is native to tropical America, it is grown around the world in tropical and subtropical regions. It has been grown ever since the Portuguese introduced it in the 17th century (Sharma *et al.* 2022). The guava is rich in vitamins like vitamin A, pantothenic acid, and ascorbic acid as well as minerals like iron, calcium, and phosphorus. (Sharma *et al.* 2021). It's commonly referred to as "the apple of the tropics" due to its distinctive nutritional worth (Sharma *et al.* 2021). It is occupying fourth and fifth place, respectively in importance in term of area planted and of production. The plant is quite hardy, prolific bearer and highly remunerative, even without much care. Now a day, guava is getting popularity in international trade due to rich nutritional value and processed products.

Good planting material is crucial for guava production as it directly impacts the yield and quality of fruit. Using healthy and disease-free guava saplings ensures a strong start, faster growth, and higher resistance to pests and diseases, ultimately leading to a more successful and profitable guava orchard. But, multiplication of guava plant from unreliable source is one of the major drawbacks in guava farming. Non availability of quality planting material and use of poor quality seedling have negatively affected the guava production and productivity. These days Government of India has focused on establishing the model nurseries for providing planting material to Indian farmers in Mission for Integrated Development of Horticulture (MIDH) scheme (Sharma *et al*., 2020).

While choosing a particular propagation technique for guava, the impact of climate change mitigation strategies and management system for crop adaptation to climate change conditions should be considered (Eitzinger, 2009). The scenario is changing from traditional propagation with incorporation of science and technology in nursery management and trade. Success of each propagation method varies from region to region due to variation in agro-climatic region

(Kumar, 2000). The graft success varies with the environmental condition i.e. Temperature, humidity, rainfall etc. However, temperature and humidity appear to be the major limiting factors affecting the success of grafting. Sprouting and subsequent survival of successful grafts is influenced by the prevailing weather condition at the time of grafting (Singh *et al.*, 2011). Therefore, it has become imperative to standardize the method of wedge grafting for guava throughout the year by vegetative multiplication under greenhouse as well as in open conditions with polycap. As very little work has been done so far on Quantification of microclimate of guava wedge grafting under Haryana conditions. Therefore, an attempt was made to assess the influence growing condition and age of rootstock on day taken for Success and Growth of grafts in guava by using wedge grafting method under Hisar condition.

MATERIAL AND METHODS

The experiment was conducted during first fortnight of February 2016-2017 & 2018-2019 in research area of Department of Horticulture, CCS Haryana Agricultural University, and Hisar (29º 10C N latitude, 75 º 46´ E longitudes, 215.2 m altitude) Haryana. The experiment was laid out in CRD with three replications to find out the suitable conditions for successful wedge grafting with polycap of guava rootstock cv. L-49 under different environment conditions (open condition, greenhouse and shade net 50%) with different age of seedling (8,12 and 16 months) time of observation. Twenty plants were taken from one replication for recording the successful grafting percentage, days taken for successful grafting, diameter of rootstock (mm) at grafting, diameter of scion (mm) at grafting, diameter of union (mm) at grafting, number of sprouts on rootstock, number of leaves on scion, increase in scion length (cm) up to 95 DAG (days after grafting), leaf area, sprouts length, height of grafted plant, fresh and dry weight of shoot & root after 95 days after grafting (DAG) under different environments.

After selection of the scion material, the rootstock was headed back, leaving 12 to 15 cm long stem above the polyethylene bag. The beheaded rootstock was split open about 4.0 to 4.5 cm deep through the center from cut end of the rootstock with grafting knife. A grafting wedge shaped cut, slanting from both the sides 4.0-4.5 cm long was made on lower side of the scion shoot. The scion stick was inserted into split of the stock and pressed properly so that cambium tissues of rootstock and scion stick could come in contact of each other. The stock and scion combination was then tied with the help of 150 gauge, 2 cm wide and 25 to 30 cm long polyethylene strip and then covered with polycap data were analyzed using online statistical analysis package (OPSTAT, CCSHAU) and treatments means were compared by C.D. at 5% level of significance

RESULT AND DISCUSSION

Diameter of rootstock (mm) at time of grafting. Perusal of the data on diameter of rootstock is a reliable index of growth and development representing the growth over a period of time. The diameter of rootstock recorded at grafting time is presented in Table 1. Diameter of rootstock increased with age of rootstock. Maximum diameter of rootstock was recorded in 16 month age of rootstock whereas minimum diameter of rootstock was observed in 8 month old rootstock.

Diameter of scion (mm) at time of grafting. Data regarding diameter of scion is requiring for different age of rootstock for grafting. The diameter of scion recorded at grafting time is presented in Table 1. Maximum diameter of scion is requiring for 16 month age rootstock for grafting whereas, minimum diameter of scion was required for 8 month old rootstock.

Diameter of union (mm) after grafting. Data appertaining diameter of union after grafting varies significantly. The diameter of union recorded after grafting time is presented in Table 1. Maximum diameter of union was obtained after grafting with 16 month age rootstock whereas; minimum diameter of union was obtained after grafting with 8 month age rootstock.

Days taken for successful grafting. Data on days taken for successful grafting (Table 1) reveals that it was significantly influenced by age of seedlings and growing conditions. It indicates that the shortest period taken for successful grafting during both the year was observed under greenhouse conditions followed by shade net 50%.

Successful grafting percentage. Data on successful grafting percentage (Table 1) indicate that it was significantly influenced by growing conditions and age of seedlings. It indicates that highest successful grafting percentage during both the year was observed under greenhouse conditions. The maximum successful grafting percentage was observed on 12 months old guava rootstock followed by 8 months old sapling.

Number of sprouts on rootstock at fortnight interval. Data on number of sprouts on rootstock (Table 2) reveals that it was significantly influenced by growing conditions and age of seedlings. There were significantly increase in number of sprouts on rootstock at fortnightly interval, maximum increase in scion length will found after 95 days after successful grafting. It indicates that the highest number of sprouts on rootstock during 2016-17 (5.06), respectively during 2018-19 (5.16) was under greenhouse conditions. The maximum number of sprouts (5.06 & 5.16) rootstock on 12 months old guava seedlings was maximum followed by 8 months.

Increase in scion length at fortnight interval. Data on increment in scion length (Table 3) reveals that it was significantly influenced by growing conditions and age of seedlings. There were significantly increase in increment

in scion length at fortnightly interval, maximum increase in scion length will found after 95 days after successful grafting. It indicates that the highest increment in scion length during 2016-17 (11.3 cm), respectively during 2017-18 (12.83 cm) was under greenhouse followed by shade net conditions. The maximum increment in scion length on 12 months old guava seedling was maximum (11.3 & 12.83 cm) followed by 8 months and 16 months.

Number of leaves on scion at fortnight interval. Data on number of leaves on scion (Table 4) reveals that it was significantly influenced by growing conditions and age of seedlings. There was significantly number of leaves on scion at fortnightly interval, maximum number of leaves on scion was found after 95 days after successful grafting. It indicates that the highest number of leaves on scion during 2016-2017 (7.21), respectively during 2018-2019 (7.51) was under greenhouse conditions.

The maximum increment in scion length on 12 months old guava seedling was maximum (7.21& 7.51) followed by 8 months and 16 months.

Fresh weight of Shoot after 95 DAG. Data on Shoot weight of seedling after 95 DAG (Table 5) reveals that it was significantly influenced by different conditions and age of seedlings. There were significantly increase in Shoot weight of seedling will found after 95 days after successful grafting. It indicates that the highest Shoot weight of seedling (37.6 g) was under greenhouse conditions compare to open field conditions during both years. The Shoot weight of seedling progressively increases (due to growth and development) with age, in case of guava seedlings the diameter attained graftable size after 12 months age of seed sowing. The maximum number of Shoot weight on 12 months age guava seedlings was maximum (37.6 g) followed by 8 month age seedling whereas minimum Shoot weight (27.2 g)was observed in open condition on 16 month old rootstock.

Fresh weight of root after 95 DAG. Pooled data analysis on fresh root weight of seedling after 95 DAG (Table 5) reveals that it was significantly influenced by age of rootstock and growing conditions. It indicates that the highest fresh root weight of seedling during both the year was observed under greenhouse. The maximum fresh root weight of seedling on 12 months old guava seedlings was maximum (16.7 g) followed by 8 month age seedling, whereas minimum root weight(9.59 g)was observed in open condition on 16 month old rootstock.

Dry weight of shoot after 95 DAG. Perusal of the pooled data on dry weight of shoot (Table 5) reveals that it was significantly influenced by different growing conditions and age of seedlings. It indicates that maximum dry weight of shoot (13.7 g) was observed under greenhouse conditions followed by shade net. The maximum dry weight of shoot (13.7 g) was observed in 12 month old rootstocks followed by 8 month age rootstock, whereas minimum shoot weight (9.5g) was observed in open condition on 16 month old rootstock.

Dry weight of root after 95 DAG. Pooled data on dry weight of root after 95 DAG (Table 5) reveals that it was significantly influenced by age of rootstock and growing conditions. It indicates that the highest dry root weight of seedling during both the year was observed under greenhouse, followed by shade net 50%. The rootstock girth of the seedling increases with age; in case of guava seedlings the diameter attained suitable graftable size after 12 months of rootstock. The maximum dry root weight of seedling on 12 months old guava seedlings was maximum (8.26 g) followed by 8 month age seedling, whereas minimum shoot weight (5.0 g) was observed in open condition on 16 month old rootstock.

Leaf area after 95 DAG. Data on leaf area (Table 6) reveals that it was significantly influenced by growing conditions and different age of rootstock. There were significantly increase in leaf area after 95 days after successful grafting. It indicates that the highest increment in leaf area (27.3cm^2), during both the year was under greenhouse conditions on 12 month age rootstock followed by shade net condition. However minimum leaf area (16.8 cm^2) was observed in open condition on 16 month old rootstock.

Length of sprouted shoot (cm) 95 DAG. Pooled data analysis given in table 6 showed that in wedge grafting on different age of rootstock and growing condition significantly affect length of sprouted shoot. Maximum length of sprouted shoot was found under greenhouse conditions (8.29 cm) on 12 month old rootstock after 95 days of grafting and minimum length of sprouted shoot was found under open condition (5.03 cm).

Height of the grafted plant (cm) 95 DAG. The pooled data presented in table 6 showed that the age of rootstock and growing condition wedge grafting was significantly affect the height of sprouted shoot. Maximum height of the grafted plant was observed under (40.3 cm) under greenhouse conditions on 12 month old rootstock and minimum in open condition (32.7 cm) on 16 month old rootstock.

Table 1: Effect of growing conditions and seedlings age on days taken for successful grafting and Successful grafting percentage (pooled means of 2016-17 and 2017-2018).

Sr. No.	Conditions	Age of seedlings (months)	Diameter of rootstock (mm)	Diameter of scion (mm)	Diameter of union (mm)	Number of days taken for successful grafting	Successful grafting %
1.	Open nursery	8	6.57	8.1	12.5	35	71.7
		12	10.5	10.4	15.7	31.7	78.3
		16	14.0	15.9	26.8	39	60.0
2.	Greenhouse	8	6.02	7.2	11.7	28.5	83.3
		12	10.3	11.4	14.5	23.8	96.7
		16	14.2	13.9	25.7	25.2	78.3
3.	Shading-net (50%)	8	6.38	7.9	12.2	30.7	76.7
		12	10.3	10.0	15.5	28.7	85.0
		16	14.7	16.1	25.8	32.8	71.7
C.D. at 5% level of significance						1.08	5.30

Table 2: Effect of growing conditions and seedlings age on number of sprouts at rootstock fortnightly interval.

Sr. No.	Conditions	Age of seedlings (months)	2016-2017 Number of sprouts at rootstock (Days after grafting)				Mean	2017-2018 Number of sprouts at rootstock (Days after grafting)				Mean
			50	65	80	95		50	65	80	95	
1.	Open nursery	8	0.51	1.89	2.44	2.33	1.80	0.54	1.99	3.05	2.33	1.98
		12	0.33	2.44	3.56	4.00	2.58	0.54	3.57	2.57	3.12	2.45
		16	0.00	0.78	1.78	2.33	1.22	0.66	1.92	1.81	2.47	1.72
2.	Greenhouse	8	0.55	2.34	3.45	4.34	2.67	0.61	2.39	3.25	3.29	2.39
		12	1.78	4.56	6.33	7.56	5.06	0.91	4.88	6.53	8.33	5.16
		16	0.66	1.67	2.22	3.78	2.08	0.73	1.23	2.17	3.09	1.81
3.	Shading-net (50%)	8	0.22	2.56	3.44	3.78	2.50	0.62	2.45	3.56	4.66	2.82
		12	0.78	4.44	6.44	6.33	4.50	0.69	2.59	4.06	4.79	3.03
		16	0.33	1.78	3.00	3.22	2.08	0.35	1.57	2.23	2.67	1.70
Mean			0.58	2.49	3.63	4.19		0.63	2.51	3.25	3.86	
C.D. at 5% level of significance			Treatment=0.45 Time=0.30 Treatment× Time=0.89					Treatment=0.47 Time=0.31 Treatment× Time=0.94				

Table 3: Effect of growing conditions and seedlings age on increment in length of grafted plant at fortnightly interval.

Sr. No.	Conditions	Age of seedlings (months)	2016-2017 Increment in Length of grafted plant in cm (Days after grafting)				Mean	2018-2019 Increment in Length of grafted plant in cm (Days after grafting)				Mean
			50	65	80	95		50	65	80	95	
1.	Open nursery	8	3.4	5.92	11.2	17.1	9.39	2.57	5.62	10.70	16.12	8.75
		12	3.85	6.8	12.3	17.4	10.1	4.27	7.82	12.45	17.73	10.57
		16	2.65	5.36	10.6	16.4	8.77	1.95	4.93	9.62	16.03	8.13
2.	Greenhouse	8	4.47	6.83	12.1	17.7	10.3	4.40	7.00	13.93	17.97	10.83
		12	4.83	7.72	13.2	18.7	11.3	5.48	10.03	15.45	20.35	12.83
		16	3.78	6.4	11.3	17	9.62	3.78	6.72	11.72	17.37	9.90
3.	Shading-net (50%)	8	3.62	6.37	11.6	17.5	9.77	3.80	6.65	11.90	17.72	10.02
		12	4.51	7.38	12.8	18.4	10.8	4.87	8.60	13.42	19.48	11.59
		16	3.05	6.3	11.1	17	9.34	2.87	5.62	10.53	15.61	8.66
Mean			3.80	6.57	11.8	17.5		3.78	7.00	12.19	17.60	
C.D. at 5% level of significance			Treatment=0. 08 Time=0.05 Treatment × Time=0.15					Treatment=0.60 Time=0.40 Treatment × Time=0.11				

Table 4: Effect of growing conditions and seedlings age on number of leaves at fortnightly interval.

Sr. No.	Conditions		2016-2017 Number of leaves (Days after grafting)				Mean	2018-2019 Number of leaves (Days after grafting)				Mean
			50	65	80	95		50	65	80	95	
1.	Open nursery	8	0.8	2.33	4.23	5.23	3.15	0.93	2.50	4.13	5.17	3.18
		12	0.67	3.89	5.44	6.55	4.14	0.71	3.73	5.70	6.92	4.27
		16	0.30	1.56	3.89	4.66	2.60	0.28	1.62	3.99	4.54	2.61
2.	Greenhouse	8	1.44	3.44	5.89	8.54	4.83	1.17	3.70	5.91	8.47	4.81
		12	2	6.11	8.66	12.07	7.21	2.23	6.54	8.93	12.3	7.51
		16	2.33	4.67	6.55	8.44	5.50	2.59	4.73	6.98	8.38	5.67
3.	Shading-net (50%)	8	0.60	2.44	5.23	8.2	4.12	0.57	2.48	5.29	8.38	4.18
		12	0.97	2.89	6.56	8.87	4.82	1.20	3.12	6.92	9.12	5.09
		16	0.89	3.34	5.22	7.11	4.14	0.86	3.34	4.92	7.22	4.09
Mean			**1.11**	**3.41**	**5.74**	**7.74**		**1.17**	**3.53**	**5.86**	**7.84**	
C.D. at 5% level of significance			**Treatment=0.82 Time=0.54 Treatment × Time=1.63**					**Treatment=0.71Time=0.47Treatment × Time=1.42**				

Table 5: Effect of different growing conditions and seedlings age on growth parameter after 95 Days after grafting. (Pooled means of 2016-17 and 2017-2018).

Sr. No.	Conditions	Age of seedlings (months)	Fresh weight of shoot 95 DAG	Fresh weight of root 95 DAG	Dry weight of shoot 95 DAG	Dry weight of shoot 95 DAG
1.	Open nursery	8	27.2	9.59	10.3	5.00
		12	30.4	12.61	11.1	6.58
		16	28.6	10.89	9.5	5.78
2.	Greenhouse	8	35.1	15.05	12.9	7.65
		12	37.6	16.69	13.7	8.26
		16	34.9	14.59	12.1	7.03
3.	Shading-net (50%)	8	33.9	13.36	11.6	6.67
		12	34.8	14.98	12.5	7.12
		16	31.8	12.73	10.6	6.08
C.D. at 5% significance			**1.4**	**0.79**	**0.6**	**0.58**

Table 6: Effect of different growing conditions and seedlings age on length of leaf area sprouts and height of grafted plant after 95 Days after grafting. (Pooled means of 2016-17 and 2017-2018).

Sr. No.	Conditions	Age of seedlings (months)	Leaf area (cm^2) 95 DAG	Length of sprouted shoot (cm) 95 DAG	Height of the grafted plant(cm)95 DAG
1.	Open nursery	8	16.8	5.03	33.8
		12	20.7	6.43	34.5
		16	18.3	5.73	32.8
2.	Greenhouse	8	25.6	7.60	38.7
		12	27.3	8.29	40.3
		16	24.1	6.91	36.5
3.	Shading-net (50%)	8	22.9	6.60	35.4
		12	24.5	6.89	36.5
		16	21.0	6.06	34.9
C.D. at 5% significance			**1.2**	**0.30**	**2.16**

DISCUSSION

Age of seedling influenced the success of grafting. The success of wedge grafting also dependent upon the environment conditions and thus may vary from place to place within a season. The seasonal aspects could be ascribed to the influence of prevailing humidity, temperature rainfall and to some extent light. Relative humidity is influenced mainly by rainfall, especially in the interior parts of country. The various factors associated with seedling age have been studied and their effects on

grafting union were thoroughly assessed by many researchers in different fruit crops. Magielse and Ochse (1930) and Sawke (1992) founded that the success percentage of grafting varies with age at which the rootstocks. Use of rootstocks at very younger age is bound to reduce the cost of planting material in nursery and will be short period, prone to enable less damage in transport as well as better survival on transplanting. It is known that the physiological maturity of seedling play an important role in the success, growth and development of grafts in different fruit crops as reported by several researcher Sawke (1992) and Vishnuvardhan (2002). Days taken for successful grafting (Table 1) reveals that shortest period was required for successful grafting under greenhouse conditions. The successful graft depends upon callus formation between scion and rootstock and rate of callus was probably higher under greenhouse condition due to better microclimate conditions compared to open condition. This trend was also observed by Visen *et al.* (2010), Singh *et al.* (2011), Syamal *et al.* (2012), Beer *et al.* (2013) and Joshi *et al.* (2014) in guava.

The minimum days taken for successful grafting were on 12 months age guava seedling. Similar results were founded in tamarind by Kumar (2001). At 12 months stage, the maturity of the rootstock might be the most appropriate for quick and successful grafting avoiding the pest-diseases and desiccation because of appropriate level of plant growth hormones. Successful grafting percentage (Table 1) reveals that highest successful grafting percentage was noticed under greenhouse growing conditions. This might be due to humid climatic environment played an important role in early and quick contact of cambium layer of scion and rootstock resulting in easy tissue formation and initiation of subsequent growth and less variation between maximum and minimum temperature contributes to high success of grafting union and further growth of grafts. This similar trend was also obtained by Singh *et al.* (2007), Visen *et al.* (2010), Singh *et al.* (2011) Syamal *et al.* (2012), Beer *et al.* (2013), Joshi *et al.* (2014), and Jadia *et al.* (2015). Highest number of sprouts on rootstock was under greenhouse conditions. This may be due to higher relative humidity in greenhouse and inside the polythene cap. It might be due to maintaining of high humidity level around the graft scion decrease the desiccation of active tissue of scion buds as compare to open conditions. Similar results werealso reported by Joshi *et al.* (2014) in guava. The highest increment in scion length (11.3) was under greenhouse conditions. It might be due to favorable high humidity, temperature, lux hours and low evaporation rate like weather condition, which are congenial for growth and development of sapling. Similar results were recorded by Patel, (2010) in mandarin; Singh *et al.* (2007) and Joshi *et al.* (2014) in guava. According to Hartman *et al.* (1997), the age of rootstock has a relationship with the regenerating ability of plant parts, which is found to be higher in young rootstock, and this is because of higher ability of meristematic tissue resulting in faster callus formation and quick healing of grafting union. Number of leaves on scion highest was recorded under greenhouse conditions. Similar results were reported Joshi *et al.* (2014) in guava.

The highest increment in leaf area ($27.3cm^2$) was under greenhouse conditions on 12 month age rootstock. It might be due to optimum temperature increases the rate of photosynthesis in leaves and leads to formation of more food material and development of graft inside greenhouse. The maximum fresh and dry weight of root -shoot was observed under greenhouse conditions on 12 month old rootstock. It might be due to increases the rate of photosynthesis in leaves and leads to formation of appropriate food material and development. The maximum length of spourt and height of grafted plant after 95 DAG founded under greenhouse on 12 month old seedlings. This might be due to appropriate temperature in protected condition in comparison of open conditions and early initiation of sprouting and fast movement of food material Singh *et al.* (1978).

CONCLUSION

Based on the results of present investigation can be concluded that the relative humidity is major factor, which along with temperature, wind speed and lux hour influence successful wedge grafting in guava. With proper monitoring of such weather factors, it may be possible to predict that protected condition for guava propagation on 12 month age rootstock can be utilized for further improvement in the technique for wedge grafting under greenhouse followed by shadenet -50% with polycap.

Declarations

Ethics approval: Not Applicable.

Consent to participate: All authors consent to participate in this manuscript.

Consent for publication: All authors consent to publish this manuscript in

Availability of data and material: Data will be available on request to corresponding or first author.

Code availability: Not Applicable

Author contribution. NS and AK drafted the experimental design and performed the experiments. NS, AS,and AG helped in data collection and data analysis. NS, AK and AGwrote initial draft of manuscript and revised the manuscript to present form. All authors read the manuscript before communication.

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