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Effect of changing weather on success of wedge and veneer grafting and chlorophyll content in mango cv. Sufaid Chaunsa

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Abstract

Mango (*Mangifera indica* L.) is cross-pollinated and allopolyploid fruiting tree. It is necessary to keep such fruit true-to-type via asexual propagation (grafting). However, a significant decline in the success of grafting in mango seedlings under changing weathering conditions has become one of a major problem for the farming community. Therefore, current study was conducted with aim to synchronize the time and type of commonly used grafting techniques in mango. For experiment *Mangifera indica* L. cv. Sufaid Chaunsa seedlings were grafted by wedge (WG) and veneer grafting (VG) techniques in different seasons i.e. in months of August, September and October. The meteorological data (temperature and humidity) was collected to examine the effect of the changes in temperature and humidity. Results confirmed that WG was better and significant in August as compared to VG in the success of Sufaid Chaunsa grafting on *seedling mangoes*. However, VG remained better and significant in October as compared to WG. Likewise, similar kind of improvement was noted in 1st sprout of leaf, height of seedlings and number of leaves. A significant improvement in chlorophyll a (24.1%), total chlorophyll (24.7%) and anthocyanin (44.6%) validated that August season is the best for WG. It is concluded that the month of August with described temperature and humidity is the best time for WG, while October is better for VG of Sufaid Chaunsa on rootstocks of Desi mango.

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Introduction

In past for a long period of time, use of seed was the only method for the propagation of fruit trees (Sharma et al., 2003; Mir and Kumar, 2011). Later, it was identified that vegetative propagation of fruits trees through grafting of desired scion on rootstock is best and less expensive approach comparative to seed and it also require shorter time for orchard establishment (Alam et al., 2006; Bally, 2006). In the technique of grafting, a scion is detached from parent plant with the use of grafting knife and then attached on rootstock by applying any binding material (Sidahmed, 1992). The most desirable characteristic of a rootstock in grafting is vigorous, prolific bearing, disease resistance and make a good stionic combination for flow of nutrients from soil to the scion (Ouma, 2007).

It is a well-established fact that an alignment of scion and rootstock cambium tissues followed by rapid division validates the success of grafting (Pina and Errea, 2005). So far researchers documented many grafting techniques including side, cleft, bench and epicotyl grafting (Kiran, 2017; Alam *et al.*, 2006). However, among various techniques of grafting, the use of wedge and veneer grafting are considered the most appropriate methods for vegetative propagation of mango trees (Abu-bakre, 1999; Lynch, 1941; Pinto *et al.*, 2018; Yadav *et al.*, 2018).

In wedge method of grafting, just below the terminal bud, slanting cuts are given on both sides of scion that ultimately resulted in the formation of wedge (Agrawal, 2007).The rootstock that is usually preferred for wedge grafting is thick which is not considered suitable for other types of grafting techniques. However, the success rate of wedge grafting can be achieved upto 90-100% (Yadav *et al.*, 2018). For *in-situ* development of mango orchards, the use of veneer grafting is also considered as the most economical and successful grafting technique (Yadav *et al.*, 2018). For veneer grafting, it is suggested that a rootstock of large size should be used. In veneer grafting, only one side of the scion is sliced in a sloping manner comparative to wedge in which both sides are sliced (Yadav *et al.*, 2018). However, inappropriate time of grafting is a major cause of failure of grafting in mango under changing temperature and humidity conditions.

Mango (Mangifera indica L.) is cultivated due to its nutritional and economic values, fruit elegance, exotic and flavour appeal (FAO, 2003; Anees et al., 2011). However, Pakistan is also one of the leading producers of mango in the world (Alexander, 1989). The optimum level of dietary fiber, high quality of taste, aroma and concentration and quality of carbohydrates made mango as king of fruits (Kumar et al., 2017). A lot of researchers documented different methods of grafting for the propagation of mango but little information is available regarding the influence of season and the appropriate method of grafting i.e. wedge and veneer under current changing climatic scenario in Pakistan. Therefore, the present study was designed with the aim to find the best grafting method i.e. wedge and veneer for propagation of mango cv. Sufaid Chaunsa compatible with changing weather conditions in semi-arid zone of south Punjab Pakistan. As mango cv. Sufaid Chaunsa is late bearing mango cultivars (August to September) so that it emerges vigorous and healthy scion due to fluctuation in temperature and humidity conditions. It is hypothesized that season of grafting could affect the success of wedge and veneer grafting in Sufaid Chaunsa.

Materials and methods

Experiment site

The experiment was conducted in the Experimental Nursery of Mango Research Institute, Old Shujabad Road, Multan. The GPS location of the site was 30°09'12.0"N and 71°26'43.1"E with 126 m elevation from sea level and 186.6 mm annual precipitation.

Design of experiment

There were three seasons for the grafting operations i.e. August, September and October while two grafting methods i.e. wedge grafting and veneer grafting. The experiment was framed into completely randomized design (CRD) with three replicates. Each replicate was comprised of 10 plants and then mean was computed.

Selection of rootstock and scion

For wedge and veneer grafting, rootstock of Desi Mango was taken from the nursery of Mango Research Institute. The age of each rootstock was approximately 12 months \pm 7 days. Uniform age and mature shoot of Sufaid Chaunsa was selected as scion almost equal in length (10-12 cm).

Grafting blade

Instead of a traditional knife, sharp surgical blade was used for grafting purpose as described by (Kiran *et al.,* 2018). No sterilization was done as the blade was already sterilized. However, a solution of 5% sodium hypochlorite was used to sterilize the grafting blade.

Grafting season

In order to evaluate the grafting season viz months of August, September and October were selected as a most appropriate time for grafting methods wedge and veneer in Multan, Punjab Pakistan as previously indicated by Kiran *et al.* (2018).

Climatic conditions

Average nocturnal temperature and humidity of August, September and October were recorded in Central Cotton Research Institute meteorological station (30° 12'N and 71° 28'E; 123 m above sea level) are presented below.

Irrigation schedule for rootstocks

Standard irrigation (every day in summer 450 ml of water) and plant protection measures were adopted as and whenever needed.

Success of grafting

For determination of grafting success, a criterion of one healthy flush was set and followed for the record of observations.

Height of grafted scion

The height of successfully grafted scion was taken by using measuring tape.

Chlorophyll contents in leaves

For determination of chlorophyll contents, 1st fresh and disease-free leaves were taken from the grafted scion of Sufaid Chaunsa. Initially, leaves were crushed in 80% acetone until it becomes paste. Finally, 20ml volume was made by using 80% acetone and absorbance was taken at different optical densities (OD) of 663, 645, 530, 503 and 480 nm on a spectrophotometer. Finally, chlorophyll content, carotenoids and anthocyanin calculations were made by using formulas of Arnon (1949):

Chlorophyll a (mg g-1) = $12.7(\text{OD } 663) \cdot 2.69 (\text{OD } 645) \text{V}/1000(W)$ Cholorophyll b (mg g-1) = $22.9 (\text{OD } 645) \cdot 4.68 (\text{OD } 663) \text{V}/1000(W)$ Total Chlorophyll (mg g-1) = $(20.2 \times \text{OD} 645) + (8.02 \times \text{OD } 663) \text{V}/1000(W)$ Carotenoids (mg g-1) = $\text{OD} 480 + (0.114 \times \text{OD} 663) - (0.638 \times \text{OD } 645) \text{V}/1000(W)$ Anthocyanin (µmol ml-1) = $(0.8173 \times \text{OD} 537) - (0.00697 \times \text{OD } 645) - (0.002228 \times \text{OD} 645) - (0.00228 \times \text{OD} 645) - (0.0028 \times \text{OD} 64$

Lycopene was calculated by using the equation of Ravelo-Pérez *et al.* (2008).

Lycopene content (mg g-1 f.wt.) = (0D503 - 0.0007) × 30.2 /g plant tissu

Where,

OD = Optical density (wavelength) V = Final volume made W = Fresh leaf weight (g)

Statistical analysis

For statistical analysis Statistix 8.1 software was used. Two factorial ANOVA was used to examine the significance of treatments. Tukey's test was applied for comparison of treatments at $p \le 0.05$.

Results

Success rate

Both main and interactive effects of season (S) and grafting techniques (GT) remained significant for success rate of the grafted scion of Sufaid Chaunsa on Desi rootstock. For success rate of the grafted scion of Sufaid Chaunsa on rootstock of Desi, wedge grafting (WG) remained significantly better as compared to veneer grafting (VG) in August and September

seasons. However, VG differed significantly better comparative to WG in October for success rate of the grafted scion of Sufaid Chaunsa on rootstock of Desi. Maximum increase of 72.3 and 171.4% in success rate was noted where WG was done as compared to VG in August and September respectively in grafted scion of Sufaid Chaunsa on rootstock of Desi. However, in October maximum increase of 150% in success rate was noted where VG was done as compared to WG in grafted scion of Sufaid Chaunsa on rootstock of Desi.

Month	Temperature (°C)		Humidity (%)		
	Maximum	Minimum	Maximum	Minimum	
August	40	28	78	60	
September	36	24	68	56	
October	30	20	65	51	

Table 1. Data of average temperature and humidity.

First leaf sprout

Both main and interactive effects of season (S) and grafting techniques (GT) differed significantly for 1st leaf sprout in grafted scion of Sufaid Chaunsa on rootstock of Desi. For 1st leaf sprout in grafted scion of Sufaid Chaunsa on rootstock of Desi, wedge grafting (WG) did not performed significantly better as compared to veneer grafting (VG) in August season. Both WG and VG remained statistically alike to each other in September for 1st leaf sprout in grafted scion of Sufaid Chaunsa on rootstock of Desi. However, VG remained significantly better comparative to WG in October for 1st leaf sprout in grafted scion of Sufaid Chaunsa on rootstock of Desi.

Table 2. Effect of wedge and veneer grafting in various season on chlorophyll contents and accessory pigments in scion of Sufaid Chaunsa on rootstock of Desi.

Grafting	Season of grafting							
	August	September	October	August	September	October		
	Chlorophyll a (mg g ⁻¹)			Chlorophyll b (mg g ⁻¹)				
Wedge	0.67±0.04 a	0.31±0.01 d	0.39±0.02 cd	0.29±0.02 a	0.12±0.02 d	0.13±0.01 cd		
Veneer	0.54±0.03 b	0.33±0.02 d	0.49±0.03 bc	0.22±0.01 ab	0.14±0.01 d	0.21±0.01 bc		
	Total Chlorophyll (mg g ⁻¹)			Carotenoids (mg g ⁻¹)				
Wedge	0.96±0.06 a	0.44±0.03 d	0.52±0.03 cd	0.039±0.002 a	0.018±0.001 d	0.021±0.001 cd		
Veneer	0.77±0.03 b	0.47±0.03 d	0.70±0.04 bc	0.031±0.001 ab	0.018±0.002 d	0.028±0.002 bc		
	Anthocyanin (µmol ml-1)			Lycopene (µg g ⁻¹)				
Wedge	0.025±0.002 a	0.011±0.001 c	0.013±0.001 c	1.66±0.07 bc	1.21±0.05 c	2.13±0.11 ab		
Veneer	0.022±0.001a	0.014±0.002 bc	0.021±0.002 ab	2.40±0.15a	1.25±0.12 c	1.22±0.12 c		

Means sharing different letters are significantly different ($p \le 0.05$).

It was also noted that WG in September and October remained statistically similar for 1st leaf sprout in grafted scion of Sufaid Chaunsa on rootstock of Desi. Maximum increase of 36.7% in 1st leaf sprout was noted where WG was done as compared to VG in August in grafted scion of Sufaid Chaunsa on rootstock of Desi. However, in October maximum increase of 83.9% in 1st leaf sprout was noted in VG was done as compared to WG in grafted scion of Sufaid Chaunsa on rootstock of Desi.

Number of leaves

Both main and interactive effects of season (S) and grafting techniques (GT) were significant for number of leaves in grafted scion of Sufaid Chaunsa on rootstock of Desi. For number of leaves in grafted

scion of Sufaid Chaunsa on rootstock of Desi, wedge grafting (WG) performed significantly better as compared to veneer grafting (VG) in August season. Both WG and VG remained statistically alike to each other in September for number of leaves in grafted scion of Sufaid Chaunsa on rootstock of Desi. However, VG remained significantly better comparative to WG in October for number of leaves in grafted scion of Sufaid Chaunsa on rootstock of Desi. It was also noted that WG in September and October while VG in August and September remained statistically similar for number of leaves in grafted scion of Sufaid Chaunsa on rootstock of Desi. Maximum increase of 1.89-fold in number of leaves was noted where WG was done as compared to VG in August in grafted scion of Sufaid Chaunsa on rootstock of Desi. However, in October maximum increase of 3.75-fold in number of leaves was noted in VG was done as compared to WG in grafted scion of Sufaid Chaunsa on rootstock of Desi.



Fig. 1. Effect of wedge and veneer grafting in various season on success rate of grafted scion of Sufaid Chaunsa on rootstock of Desi. Means sharing different letters are significantly different ($p \le 0.05$).

Scion height

Both main and interactive effects of season (S) and grafting techniques (GT) were significant for the height of grafted scion of Sufaid Chaunsa on rootstock of Desi. For the height of grafted scion of Sufaid Chaunsa on rootstock of Desi, wedge grafting (WG) performed significantly better as compared to veneer grafting (VG) in August and September seasons. However, VG remained significantly better comparative to WG in October for the height of grafted scion of Sufaid Chaunsa on rootstock of Desi.

It was also noted that VG in August and October remained statistically similar for the height of grafted scion of Sufaid Chaunsa on rootstock of Desi. Maximum increase of 50 and 253% in height was noted where WG was done as compared to VG in August and September respectively in grafted scion of Sufaid Chaunsa on rootstock of Desi. However, in October maximum increase of 155% in height was noted where VG was done as compared to WG in grafted scion of Sufaid Chaunsa on rootstock of Desi.

Chlorophyll contents

Both main and interactive effects of season (S) and grafting techniques (GT) were significant for chlorophyll a, chlorophyll b, total chlorophyll, carotenoids, anthocyanin and lycopene content in leaves of the grafted scion of Sufaid Chaunsa on rootstock of Desi. For chlorophyll a and total content in leaves of the grafted scion of Sufaid Chaunsa on rootstock of Desi, wedge grafting (WG) performed significantly better as compared to veneer grafting (VG) but remained statistically alike for chlorophyll b and carotenoids in August season. Both WG and VG remained statistically alike to each other in September and October for chlorophyll a, chlorophyll b, total chlorophyll and carotenoids content in leaves

of the grafted scion of Sufaid Chaunsa on rootstock of Desi. Maximum increase of 24.1and 24.7% in chlorophyll a and total chlorophyll content was noted where WG was done as compared to VG in August. In the case of anthocyanin, WG and VG remained statistically similar to each other in August and September. However, VG differed significantly better as compared to WG in October for anthocyanin in leaves of the grafted scion of Sufaid Chaunsa on rootstock of Desi. Maximum increase of 61.5% in anthocyanin in leaves of the grafted scion of Sufaid Chaunsa on rootstock of Desi was noted where VG was done as compared to WG in October.



Fig. 2. Effect of wedge and veneer grafting in various season on 1st leaf sprout of grafted scion of Sufaid Chaunsa on rootstock of Desi. Means sharing different letters are significantly different ($p \le 0.05$).

For lycopene content, VG remained significantly (44.6%) as compared to WG in August for lycopene in leaves of the grafted scion of Sufaid Chaunsa on rootstock of Desi. However, WG differed significantly (90.9%) as compared to VG in October for lycopene in leaves of the grafted scion of Sufaid Chaunsa on rootstock of Desi.

Discussion

In current study, mango cv. Sufaid Chaunsa scions which were used as WG in August manifested significantly better success rate comparative to VG. However, success rate was significantly better in VG done in October as compared to WG. This improvement in success rate of grafted Sufaid Chaunsa might be due to more active alignment of Sufaid Chaunsa scion's cambium tissues with rootstock of Desi due to availability of optimum temperature and humidity required for respective grafting technique. Moreover, high temperature facilitates the free sap flow in the scion of mango cv. Sufaid Chaunsa and this free sap serve as cementing agent for the firm stionic combination of mango scion to its desi or seedling mangoes rootstock tissues. The presence of this free sap also promotes the actively dividing cambium tissue as well as callus development and bridging the flow of assimilates from rootstock to scion for the proliferation of new buds. A direct relationship was found between the aerial temperature and humidity with the activity of sap flow in the tissue mangoes. Islam et al. (2004) argued that optimum environmental conditions facilitates cell division that improved growth in grafted seedlings. Rapid cell division helped a lot in the formation of callus and intermingling of vascular bundles in rootstock as well as in scion. Sadhu (2005) documented that ideal temperature and humidity range in environment enhanced the success of grafting in mango. Furthermore, results of current study also validated that chlorophyll a, chlorophyll b, total chlorophyll, carotenoids and anthocyanin were also improved where success rate, 1st leaf sprout,

height of seedling and number of leaves were improved significantly in WG (August) and VG production (October). Higher of lycopene (antioxidant) validated the presence of any stresses condition might be due to wrong selection of grafting technique according to season requirement. Gagandeep and Malhi (2006) that continuity of sap as a result of proper translocation of nutrients solutes played an imperative role in improvement of chlorophyll content in leaves of grafted Sufaid Chaunsa (Sivudu *et al.,* 2014).

According to Sivudu *et al.* (2014) better availability of water under optimum level of temperature improved rate of photosynthesis in grafted seedlings. Similar trend of results were also noted by Joshi and Syamal (2014) and Joshi *et al.* (2014).



Fig. 3. Effect of wedge and veneer grafting in various season on number of leaves of grafted scion of Sufaid Chaunsa on rootstock of Desi. Means sharing different letters are significantly different ($p \le 0.05$).



Fig. 4. Effect of wedge and veneer grafting in various season on height of grafted scion of Sufaid Chaunsa on rootstock of Desi. Means sharing different letters are significantly different ($p \le 0.05$).

However, Singh *et al.* (2005) suggested significant role of optimum temperature in the sprouting of bud and development of 1st leaf on seedlings. Higher level of humidity decreased the desiccation area of bud in scion that facilitate the active tissue to grow and resulted in leaf sprouting (Visen *et al.*, 2010). The aerial environmental conditions support results of the sap flow and activity of cambium in mango cv. Sufaid

Chaunsa for its success. On the other hand, photosynthetic machinery like pigment synthesis i.e. chlorophyll a, b, total chlorophyll, carotenoids, lycopene and anthocyanin enhanced in warm weather conditions and decline as the temperature of the environment decline. Inversely, veneer graft success might be due to less surface exposure in grafting while wedge grafting poses large surface area in terms of invert cut into rootstock and further a long slanting cut into both sides of scion needs to be longer span of time and energy to heal up and make firm joint between rootstock and scion tissues.

Conclusion

It is concluded that the month of August with described temperature and humidity is the best time for WG of Sufaid Chaunsa mango on Desi rootstock. However, month of October is better for VG of Sufaid Chaunsa on rootstocks of Desi mangoes.

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