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Effects of cutting length, time and growing media on the sprouting of dormant semi-hardwood cuttings of pomegranate cv. Wonderful

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Scientific confirmation of the traditionally known health benefits of pomegranates caused a tremendous increase in the consumption and production of this highly valuable fruit tree. Reproduction of pomegranate plants with seeds shows great variability in tree vigour and fruit quality. Cuttings is one of the most successful and preferred methods for the propagation of pomegranates. It is easy, quick, economic and most the convenient method of obtaining true-to-type trees in considerably less time; however, it has a high rate of mortality. Therefore, the present study aimed to determine the effects of cutting length, collection time of cuttings and rooting media on the sprouting of dormant semi-hardwood cuttings collected from pomegranate cv. Wonderful. Studies conducted with the completely randomized block design with four replications, each replication containing 25 individual cuttings. According to the results, the highest sprouting percentage obtained from the 10 cm cuttings collected 43±3 days before sprouting (DBS) and grown in soil with 98%. Results showed that the sprouting percentage is higher at the shorter cuttings. It was also concluded from the results that, as time pass, the sprouting percentage of the cuttings decrease, and is important to collect cuttings about 40 days before sprouting. The growing media was found to significantly affect the sprouting percentage and soil was found to be better than perlite media.

Key words: Perlite, rooting, soil, sprouting, transplant.

INTRODUCTION

Pomegranate tree (*Punica granatum* L.) is among the first cultivated crops in the world. Although it is as long as history of cultivation, its production began to increase tremendously after 20th century, with the scientific confirmation of its health benefits (Jurenka, 2008). Furthermore, lots of research conducted about the health benefits of pomegranates and those research revealed the efficacy of pomegranate fruits, leaves, bark, juice etc. against cancer, heart disease, hypertension and some infectious diseases (Aviram et al., 2000; Lansky et al., 2005; Türk et al., 2008). Pomegranate tree originated in central Asia, but it is grown in variable geographic conditions due to its adaptability to wide range of climatic and soil conditions (Holland et al., 2009). The shrub-like

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trees of pomegranates are deciduous and known to be long-lived plants. Reproduction of pomegranate plants with seeds shows great variability in tree vigour and fruit quality. Therefore, the most desirable method for the propagation of pomegranate tree is vegetative propagation. In this respect, cuttings, air layering and tissue culture are successful (Karimi et al., 2012), but air layering is reported to adversely affect mother tree and is more expensive than cutting (Purohit, 1981). Reproduction of pomegranate plants from shoot cuttings is easy, quick, economic and most convenient method of obtaining true-to-type trees in considerably less time (Polat and Caliskan, 2006).

The most important problem for cuttings method is the high mortality rate (Sharma et al., 2009) which strongly varies among varieties (Owais, 2010), cutting length, time of the year (Sebastiania and Tognettib, 2004) and cultivation practices applied. It was previously reported that indole-3-butyric acid (IBA) (Sharma et al., 2009; Sarrou et al., 2014), ascorbic acid, gibberellic acid (GA3), hydrogen peroxide (H_2O_2) and melatonin (MEL) (Sarrou et al., 2014) have positive effects on the rooting, root number, root length and shoot length of pomegranate cuttings. The time of the year that shoot cuttings are being collected is so crucial for obtaining a high performance of sprouting in the cuttings. On the other hand, the length and diameter of the cutting is utmost important, in which it determined the physiological potential of the cuttings (Chandra and Babu, 2010). Generally, 6-12 mm thick stem cuttings provide optimum rooting (Dhillon and Sharma, 1992; Rajan and Markose, 2007) but no useful information exist for cutting length. The carbohydrate sink of the cuttings is important for sprouting, as it is required for the cuttings until it rooted (Chadha, 2001). Many studies have been conducted on the propagation of pomegranate (Polat and Caliskan, 2006; Sharma et al., 2009; Owais, 2010; Karimi et al., 2012; Sarrou et al., 2014; Chater et al., 2017), however no experiments are known to study the effects of cutting length, time and rooting media on the rooting of pomegranate cuttings. Thus, this present study aimed to determine the effects of cutting length, cuttings' collection time of the year and rooting media on the sprouting of dormant semi-hardwood cuttings collected from pomegranate (cv. Wonderful).

MATERIALS AND METHODS

Plant materials, experiments and growth conditions

Semi-hardwood cuttings from different time of the year (1st research parameter: time) with different lengths (2nd research parameter: cutting length) were planted in different growing media (3rd research parameter: rooting substrate). Dormant semi-hardwood cuttings of the present study were collected on 1st of February and 1st of March 2017, from 8 years old pomegranate tree (cv. Wonderful). Cuttings were taken from the above branches of the trees, not from the basal suckers. The thickness of the collected plant materials was uniform, equalling to 10±1 mm diameter as suggested by Rajan and Markose (2007). Since cutting length was a research parameter in present study, semi-hardwood cuttings were cut as 10, 20, 30 and 40 cm lengths, respectively. The other research parameter of present study was the growing media, which are soil (sandy loam) and perlite. Growing media was filled in plastic containers (30 × 50 cm), and one plastic container was used a replication for each treatment (cutting length). The experiments were designed according to the completely randomized block design with four replications, each replication containing 25 individual cuttings. Minimum two nodes from each cutting length were planted below ground (3 to 5 cm deep), where minimum two nodes were also left above ground. The plastic containers with shoot cuttings were then placed in a greenhouse with a temperature of 25 to 30°C. Cuttings were hand-watered every day for 4 weeks and then irrigated as needed. Sprouting of the cuttings began in Mid-March and studies continued until the end of May (119 days for cuttings was collected on 1st of February, and 91 days for cuttings collected on 1st of March). A foliar application of a nutrient solution including 200 ppm nitrogen, 80 ppm phosphorus and 60 ppm potassium was performed on 1st of April and 1st of May.

Data collection and statistical analysis

During the experiments, sprouting of the plants were checked regularly. At the end of the experiments, number of sprouted plants and length of the highest shoot of each cutting was recorded. The data was summarized using Office Excel and SPSS 20. Data was analysed with analysis of variance (ANOVA) and mean separations were done by Tukey's multiple range test at $P < 0.05$. Comparison of the different time of cutting collections was done with independent samples t-test at 1 and 5% level respectively.

RESULTS AND DISCUSSION

First sprouting of cuttings began on 16th of March 2017 (43±3 days after 1st of February and 15±3 days after 1st of March). Mean sprouting results of present study showed that since the cutting length increase, the sprouting percentage of the cuttings decrease (Table 1.). The highest sprouting percentage was measured as 98%, obtained from the 10 cm cuttings, collected 43±3 days before sprouting (DBS) and grown in soil. The same length of cuttings (each 10, each 20, each 30 or each 40 cm, respectively) from the same time (1st of February or 1st of March 2017), but grown in perlite showed 95% sprouting, and no statistical difference was obtained between them. The sprouting percentage of 40 cm cuttings collected 43±3 DBS was recorded to be 69% in soil and 59% in perlite. The lowest sprouting percentage (19%) was measured from the 40 cm cuttings collected 15±3 DBS and grown in perlite. The results showed that as time pass, the sprouting percentage of the cuttings decrease, and is important to collect cuttings about 40 days before sprouting. These results are consistent with other studies who reported that cutting length (Chadha, 2001; Sebastiania and Tognettib, 2004; Chater et al., 2017) and time of the year (Sebastiania and Tognettib, 2004; Chandra and Babu, 2010) are important factors for propagation of plants from cuttings. On the other hand, results of present study are differing from the results of

Table 1. Mean sprouting percentage of semi-hardwood pomegranate cuttings of different length obtained at different time of the year and planted in different growing media.

Treatments (cutting length cm)	Sprouting percentage					
	43±3 DBS		15±3 DBS		Average	
	Soil	Perlite	Soil	Perlite	43±3 DBS	15±3 DBS
10	98.00^{aA}	95.00^{aA}	95.00^{aA}	79.00^{aB}	96.50^{a}**	87.00^{a}**
20	94.00^{aA}	88.00^{aA}	87.00^{aA}	60.00^{bB}	91.00^{a}**	73.50^{a}**
30	80.00^{bA}	70.00^{bAB}	65.00^{bB}	30.00^{cC}	75.00^{b}**	47.50^{b}**
40	69.00^{cA}	59.00^{cB}	41.00^{cC}	19.00^{cD}	64.00^{c}**	30.00^{c}**

Values followed by the same small letter or letters 'a, b, c' within the same column; and same CAPITAL letter or letters 'A, B, C' within the same row are not significantly different at 5% level (Tukey's HSD multiple range test). Average data were compared with independent samples t-test; and * used to show significant differences at 5% level, ** to show significant differences at 1% level, and ns represents non-significant at 5% level. DBS: days before sprouting.

Table 2. Mean shoot length of semi-hardwood pomegranate cuttings of different length obtained at different time of the year and planted in different growing media.

Treatments (cutting length cm)	Length of highest shoot (cm)					
	43±3 DBS		15±3 DBS		Average	
	Soil	Perlite	Soil	Perlite	43±3 DBS	15±3 DBS
10	11.74^{aAB}	10.42^{bC}	12.48^{aA}	11.36^{aB}	11.08^{b}ns	11.92^{a}ns
20	13.44^{bA}	11.80^{aB}	9.59^{bC}	9.41^{bC}	12.61^{a}**	9.50^{b}**
30	15.65^{aA}	11.69^{aB}	9.52^{bC}	5.82^{cD}	13.67^{a}**	7.67^{c}**
40	12.75^{bA}	11.92^{aA}	6.68^{cB}	5.42^{cC}	12.33^{ab}**	6.05^{d}**

Values followed by the same small letter or letters 'a, b, c' within the same column; and same CAPITAL letter or letters 'A, B, C' within the same row are not significantly different at 5% level (Tukey's HSD multiple range test). Average data were compared with independent samples t-test; and ** used to show significant differences at 1% level, where ns represents non-significant difference at 5% level. DBS: days before sprouting.

Owais (2010) who recommended 20 cm cutting length for optimum propagation of pomegranate. Significantly, similar performance of 10 cm length cuttings with the cuttings of 20 cm length is a valuable result in which number of plants might be doubled by dividing the 20 cm cuttings into two; while at the same time the sprouting percentage of the cuttings would be better. Previously, Mehta et al. (2018) conducted a similar study with pomegranate cuttings collected in different periods of the year, which is end of December, mid-January and end of January. They reported that the rooting and sprouting percentage of the pomegranate cuttings are higher at the end of January. These results are supporting our findings, where highest sprouting percentage obtained from the cuttings collected in February. Kaur et al. (2016) conducted a similar study by collecting the pomegranate cuttings in August and in January; and they reported that the rooting percentages are higher in August. This study also supports present results in a case that: collecting time of the cuttings is crucial for success.

Contrary to the sprouting percentages, highest shoot length of the cuttings recorded from the 30 cm cuttings collected 43±3 DBS and grown in soil with 15.65 cm (Table 2). It was followed by 20 cm and 40 cm cutting lengths. Similar results were obtained at perlite growing media. The energy required for the cuttings is obtained from the carbohydrate source of the cuttings (Chadha, 2001) and these results are not surprising. The shooting length of the cuttings with 30 cm and 40 cm length obtained 15±3 DBS and grown in perlite were found to be shorter than the other shorter cuttings. These results are in agreement with the other results of present study and suggest that the cuttings should be collected at least 40 days before sprouting to reach success. The results of the present study are in agreement with the findings of Rajkumar et al. (2016) who reported that the rooting percentage of pomegranate cuttings is decreasing in perlite growing media while increasing in soil, or in the combination of soil with cocopeat. The low percentage or sprouting (and rooting) in perlite media seems to be due to its low water retention capacity. Previously many studies conducted to improve the rooting and sprouting percentage of the pomegranate cuttings, especially with indole-3-butyric acid (IBA) and gibberellic acid (GA3) (Ghosh et al., 1988; Sandhu et al., 1991; Rajan and Markose, 2007; Sharma et al., 2009; Sarrou et al., 2014;

Rajkumar et al., 2016; Mehta et al., 2018; Hakim et al., 2018). Auxins plays an important role in rooting and IBA promotes root development via promoting cell enlargement by affecting the synthesis of enzymes (Damar et al., 2014). However, the results of the present study are opposing to the idea that, pomegranate cuttings require auxins for initiations of rooting and sprouting (Singh, 2017); suggesting that correct time of collection of cuttings and correct length (mainly shorter lengths, average: 10 cm) provide high efficacy in rooting and sprouting without needing any rooting hormone.

Conclusion

The greatest percentage in sprouting was recorded in cuttings with short length (10 cm) which was collected 43±3 DBS. The growing media (soil or perlite) was also found to significantly affect the sprouting percentage and the soil was found to be better than perlite media. The results suggest that using shorter cutting lengths and collecting the propagation materials at the right time might double, even triple the number of rooted plants and improve economic performance of reproduction.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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