Range Mgmt. & Agroforestry 34 (2) : 162-170, 2013
ISSN 0971-2070

Effect of seed collection time and pre-treatment on germination, identification and vegetative propagation of superior germplasm of *Terminalia chebula* Retz. - A multipurpose agroforestry tree

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Received: 4th May, 2012 Accepted: 1st July, 2013

Abstract

Interest in cultivation of multipurpose tree, *Terminalia chebula*, has been constrained by its poor regeneration, long gestation period, higher fruit set heights, unavailability of superior genotypes and lack of standardised agrotechnology. The present study investigates the effect of different harvesting time and seed treatments on seed germination, variation in fruit characteristics, and effect of sizes of root stock (diameter) and methods of grafting on sprouting success and growth of the seedlings. The seeds collected from mid January till mid March resulted in statistically significant higher germination than those collected from mid November to January beginning. Mechanical breaking of seed coat resulted in the highest germination (73.6%) which was significantly higher than all other pre-sowing treatments. The mother tree located in Mathwar in Indian province of Jammu and Kashmir, was statistically superior to other mother trees with respect to fresh and dry fruit weights, fresh pulp weight and fruit length and consequently, this mother tree was selected to obtain the scion for its further propagation using cleft grafting and patch budding. The cleft grafting resulted in significantly higher final sprouting success and growth of the seedlings. The thickness (collar diameter size) of the root stock did not influence the sprouting success but growth was significantly better in large sized rootstock.

Keywords: Cleft, Fruit, Grafting, Medicinal, Mother tree, Patch, Pulp

Abbreviation: DAGB:Dry aboveground biomass, **DBGB**: Dry below ground biomass, **DRW(P)**: Dry weight of primary roots, **DRW(S)**: Dry weight of secondary roots, **DSW:** Dry shoot weight, **DTB:** Total dry biomass, **FAGB:** Fresh above ground biomass, **FBGB**: Fresh below-ground biomass, **FLW:** Fresh leaf weight, **FRW(P)**: Fresh weight of primary roots, **FRW(S)**: Fresh weight of secondary roots, **FSW**: Fresh shoot weight, **FTB:** Fresh total biomass, **LL**: Leaf length, **LW**: Leaf width, **NL**: Number of leaves, **NSR**: Number of secondary roots, **RL(P)**: Length of primary root, **R:S (DW)**: Root: shoot ratio dry weight basis **SL**: Shoot length

Introduction

Trees and forests play an important role in the livelihood of the people in the various parts of world by providing goods and services (Sood and Mitchell, 2009). Increasing demand and shrinking habitats may lead to extinction of many plants and trees (Rana and Sood, 2012). The lack of knowledge on propagation techniques of a majority of these species and unavailability of their superior selections constrains their cultivation on farmersô fields and wastelands (Khan and Khanum, 2000). Further choice of many species for afforestation on farmersôland is limited by their long gestation period, longer harvesting heights and poor quality planting stock (Rana and Sood, 2012). This is also true for *Terminalia chebula*.

Terminalia chebula is of one of the multipurpose and medicinal agroforestry tree species occurring mainly in India, Sri Lanka, Myanmar, Thailand and China. It occurs in Indian forests and agroforestry systems extending from sub-temperate to tropical region (Singh, 1982). The fruits of the species are used locally in many medicines and are an important constituent of ôTriphalaô(a medicinal digestive stew) and commercially used in many Ayurvedic medicines to treat heart burn, flatulence, dyspepsia, liver and spleen disorders, asthama and constipation. Production in India is estimated to be 100000 tonnes of which 20% is exported to adjoining countries, Europe and U.S. (World Agroforestry Centre, n.d.). The tree is lopped for leaves to feed the livestock during winter when other green fodder is very scarce. The leaves contain 1.73% nitrogen (equivalent to 10.80% protein content) and 2.75%

Calcium (Singh, 1982). Large sized fruits fetch higher price in the national and international markets and thus fruit size is an important characteristic. The poor germination capacity, lack of natural regeneration and knowledge regarding its propagation are the limiting factors for its adoption in agroforestry systems. The juvenile period of the species is also longer as the full production of the fruits take place at about 20 years. Further its cultivation in agroforestry systems is constrained by lack of availability of superior selections. Thus, there is need of more productive planting stock with lower juvenile period and comparatively large fruit size. The studies on variability of desirable traits are useful to screen the available germplasm for higher productivity and future improvement (Todaria *et al*., 2003). Identification of mother trees with desirable trait(s) is, therefore, imperative for raising planting stock with desirable traits.

Keeping above in view this study aims at identifying the proper harvesting time and pre-sowing treatment so as to enhance the seed germination in this species. This study also investigates fruit size variation to identify mother tree with large sized fruits to obtain the scions for its further vegetative propagation. Grafting is a method of vegetative propagation that allows the production of the progeny of same genetic makeup. Thus, it helps in multiplication of trees of desirable genotype. Grafting can be used for multiplication of trees in reproductive phase (Hartmann *et al*., 2002; Zobel and Talbert, 1984). Different species have different success of vegetative propagation through different grafting methods (Awasthi and Shukla, 2003; Blada and Panea, 2011; Danthu *et al*., 2004). Grafting helps in reducing the juvenile phase of the trees (Hartmann *et al*., 2002) and results in tree dwarfing thereby making the harvesting of fruits more convenient. Therefore, study was also taken to know effect of different grafting methods on success and growth of seedlings of the species so obtained by grafting of scions obtained from identified superior tree to enable conservation of its superior germplasm.

Materials and Methods

Study area

The seed germination and grafting experiments were carried out in the nursery of Division of Agroforestry of Sher-e-Kashmir University of Agricultural Sciences and Technology, Chatha, Jammu (Jammu & Kashmir), India. This nursery is located at 32° 32ôN latitude and 74° 58ôE longitude at an elevation of 332 m above mean sea level. The average annual rainfall in this area is 1200 mm with biomodal distribution. A major part of the rainfall occurs in July-August followed by December-January (Rana and Sood, 2012). The mean maximum daily temperature is 38° C and mean of minimum daily temperature is 5° C. The soil of the nursery is mainly sandy clay loam (Rana, 2008). The site of collection of different mother trees is subtemperate and subtropical where the mean daily temperature ranged from 5-40°C and falls in Indian provinces of Jammu and Kashmir, and Himachal Pradesh.

Effect of time of seed collection on germination

Two trees at Mathwar (located in Jammu and Kashmir) were marked and fruits of these trees were collected from 15th Nov, 2005-18th March, 2006 at fortnightly interval. The seeds (without any treatment) were sown in the nursery in three replications of 100 seeds each during April and total germination percentage was recorded at 45 days after sowing of the seeds. Analysis of variance was carried out after square-root transformation of the percentage after Gomez and Gomez (1984).

Effect of pre-sowing treatments on seed germination

The seeds for conducting this experiment were collected in first week of March. The following treatments were given to seeds to enhance the germination: Dipping in concentrated sulphuric acid for 5, 10, 15, 20, 25 and 30 minutes respectively each followed by immediate washing of seeds with cold water, scarification in cowdung for 15 days, 30 days and 45 days respectively, mechanical breaking the seed coat and control (no treatment). Each treatment contained 100 seeds and 3 replications. Effect of treatments was analysed using one way ANOVA after arcsine transformation of the percentage. The sowing of the pre-treated seeds was done in April 2007.

Seed characteristic variability

To study the seed characteristic variability inter-personal and telephonic discussions, were held with forestry experts in Jammu & Kashmir and Himachal Pradesh to identify the locality with abundance of *Terminalia chebula* (Table 3). As it was very difficult to visit and to collect fruits from all trees in each respective locality, rapid rural appraisal (RRA) techniques were used to identify trees with large sized fruits in the respective locality. These localities were visited and trees bearing large sized fruits were identified by holding discussions with a group of key local informants as well as by locality transect walk and visual qualitative observations during the ripening period of fruits (November to March). These trees were then marked and sample of 20 fruits (three replications per tree) was collected randomly from each selected ontogenetically old tree to estimate the fruit size variation. The fresh weight of each sample was determined by weighing the samples using digital weighing balance. The pulp of fresh fruits in each sample was removed mechanically and weighed to obtain the fresh pulp weight of the respective sample. The pulp content and seeds (left after removing the pulp) of each sample were dried separately in oven at 80^{O}C for 48 hours (Raizada and Srivastava, 1989). The dry pulp weight and seeds for each sample was determined by weighing these dried samples. The dry pulp weight and dry seed weight of each sample was added to get the dry fruit weight of the respective sample. The fruit weight and pulp weight (fresh and dry) of each sample was divided by 20 to get the data per fruit basis for each replication. The mother tree with largest mean fruit size, fruit weight (fresh and dry) and pulp weight (fresh and dry) was identified.

Effect of grafting methods on survival and growth of the grafts

To further propagate the identified superior mother tree vegetatively, scions from the said mother tree selected at Mathwar (Table 3) were grafted to 21 months old root stock of the species in 2007. Two types of root stock, small (collar diameter 8-15cm) and large (collar diameter 16-25cm) and two methods of grafting were (patch budding and cleft grafting) used to propagate the pre-said superior scion. Each diameter class contained 40 plants (20 for patch budding and 20 for cleft grafting) for each of four replications in the experiment. The experiment was laid in Factorial Randomised Block Design. The grafting was carried in April, 2007 at 10 cm height and length of the scion above grating height was 5 cm. Each scion and respective root stock was matched in size so that cambium tissues are juxta-posed. The data on scion sprouting success was recorded after 15, 30, 45, 90 and 120 days after grafting as the percentage of scions sprouted and growth parameters (fresh shoot weight, shoot length, fresh leaf weight, leaf length, leaf width, number of leaves, dry shoot weight, fresh root weight for primary and secondary roots, number of secondary roots, length of primary root, dry weights of primary and secondary roots, fresh and dry aboveground biomass, fresh and dry below ground biomass, total dry biomass and root: shoot ratio on dry weight basis) were recorded six months after grafting in the nursery conditions. Three sample grafted plants for each diameter and grafting method were selected using simple random sampling to record the growth parameters in each replication. The dry biomass of respective components was esti-mated by drying the samples (roots and above ground parts separately) in the oven at 80^{O}C for 48 hours and weighing the dried samples (Raizada and Srivastava, 1989). The data was analysed using Tukeyòs test and significance of difference ($p < 0.05$) between treatments (root stock size, grafting method and interaction root stock size x grafting method) was based on post-hoc comparisons of means. The seedlings obtained by cleft grafting of scion from Mathwar mother tree were planted in the same experimental area which started bearing fruits after 3 years of planting.

Results and Discussion

The seed collection time significantly ($p < 0.05$) influenced the germination per cent (Table 1). The germination was highest (10.7%) in case of the seeds collected in the February 16th collection which was significantly higher than Jan 16th collection but at par with February 1st, March 3rd and March 18th collections. No germination was observed in Nov, Dec and Jan 1st collections. This implies that maximum obtainable germination of untreated seeds is quite low (10.7%) but variable with time of the seed collection. The seeds for nursery rising can be collected February Ist onwards till mid of March. Significant effect of seed collection time on seed germination was also observed in *Terminalia sericea* in Malawi (Likoswe *et al*., 2008). The poor germination of untreated seeds could be attributed to hard seed coat of *Terminalia species* (Likoswe *et al*., 2008; MoEF, n.d.) The higher germination of seeds collected from February onwards may be due to their physical and physiological maturity.

Since germination with untreated seeds was quite low as evidenced in the above experiment, thus experiment to enhance seed germination using different pre-sowing treatments was carried out. There was significant ($p<0.05$) influence of seed treatment on germination (Table 2). Mechanical breaking of seed coat resulted in highest ger-mination percentage (73.6%) which was significantly higher than all other treatments. Similar findings have been reported in *Terminalia sericea* in Malawi (Likoswe *et al*., 2008). There were no significant differences between ger-mination per cent obtained with different sulphuric acid treatments except 25 minutes dip. However, the germina-tion under 5, 10, 15 and 20 minutes sulphuric acid treat-ments was significantly lower than that of cowdung scari-fication treatments for 15, 30 and 45 days (Table 2). Amongst cowdung scarification treatments, 30 days scari-fication had statistically higher germination (38.7%) than 15 days scarification (25.4%) but at par with 45 days scari-fication (37.7%). Under the field conditions of India where farmers have easy access to cowdung than sulphuric acid, cowdung scarification for 30 days can be used to enhance the germination of the species. The lowest germination in control than all other treatments could be due to higher impermeability of hard seed coat to water and air (Likoswe *et al*., 2008).

Table 1: Effect of seed collection time on seed germination

Date of collection	Mean Germination (%)
November 15th	0 (*0)[a]
December 1st	0 (0)[a]
Dec 16th	0 (0)[a]
Jan 1st	0 (0)[a]
Jan 16th	7.6 (15.9)[b]
Feb 1st	10.0 (18.0)[c]
Feb 16th	10.7 (18.9)[c]
March 3rd	10.3 (18.6)[c]
March 18th	10.3 (18.6)[c]
$CD_{0.05}$=2.97	

*Figures in brackets are transformed values
Means with different letters indicate p< 0.05 Tukey's honestly significant difference for transformed values

Table 2: Effect of different seed treatments on seed germination

Treatment	Mean Germination (%)
Control (No treatment)	9.5 (*17.91)[e]
Concentrated Sulphuric acid dip (minutes)	
5	12.4 (20.4)[d]
10	12.0 (20.2)[d]
15	15.6 (23.2)[d]
20	17.3 (24.3)[d]
25	20.3 (26.6)[c]
30	17.8 (24.8)[cd]
Cow dung scarification (days)	
15	25.4 (30.1)[c]
30	38.7 (38.4)[b]
45	37.7 (37.9)[b]
Seed coat breaking	73.6 (59.2)[a]
$CD_{0.05}$ =5.72	

*Figures in brackets are transformed values
Means with different letters indicate p< 0.05 Tukey's honestly significant difference for transformed values

There was significant variation in all the studied fruit characteristics from one mother tree to another in the current study (Table 3). This finding is in consonance with that of Sood *et. al*. (2009) who reported significant variations in the fruit characteristics of *Terminalia bellirica* in Jammu and Kahsmir. The highest value for all fruit characteristics was found in the mother tree located at Mathwar. The fresh fruit weight (44.6 g), dry fruit weight (21.75 g), fruit length (7.9 cm) and fresh pulp weight (34.78 g) of this mother tree was significantly different than all other mother trees. The fruit diameter of this mother tree of Mathwar was statistically at par with that of Rabta and Raya but significantly superior to remaining mother trees (Table 3). Due to superiority of a majority of fruit characteristics of this mother tree than other mother trees, the same was selected for taking scion material for grafting experiment. It is interesting to note that this mother tree is locally called "Raj Harad" *i.e.* "King of this species" owing to the superiority of its fruit characteristics which has been proved by this empirical work.

There was no effect of root stock size (diameter) on percentage of sprouting. This implies that root stock of both the sizes will result in similar sprouting success both in patch budding as well as cleft grafting. The method of grafting significantly influenced the percent of the graft sprouting ($p < 0.05$). The cleft grafting had significantly higher sprouting success than patch budding (Fig 1). At 120 days after grafting the overall sprouting in cleft grafting (72.5%) was significantly higher than patch budding (55.0%). In cleft grafting, a higher proportion of the grafts sprouted from the beginning onwards. This could be attributed to early establishment of union between root stock and scion in cleft grafting owing larger area of cambial contact between root stock and scion in this case. El-Dean and El-Rhman (2011) also observed higher sprouting success in cleft grafting in Pistachio in Egypt. The percent of the grafts sprouted varied significantly with the interval at which it was recorded (Fig 1) in both the methods. Mng'omba *et al*. (2010) also reported similar findings in *Mangifera indica*.

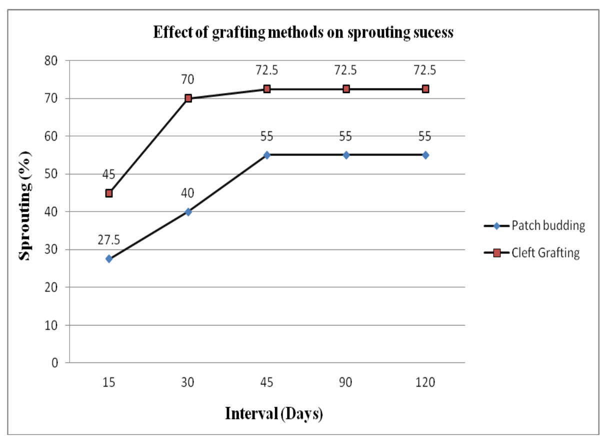

Fig 1. Effect of grafting methods on sprouting success at different intervals

There was significant effect of the grafting method and root stock size on above ground growth parameters, except in case of leaf length (LL) and leaf width (LW) where the effect of grafting method was non-significant (Table 4). The mean fresh shoot weight (FSW), shoot length (SL), total fresh leaf weight (FLW), number of leaves and dry shoot weight (DSW) were significantly higher in cleft grafting than in the patch budding, and in sized root stock than small sized root stock (Table 4). This could be attributed to early establishment of union between scion and stock which provides longer period of the growth for cleft grafts than in patch budding. This is evident as a higher sprouting success (45%) was obtained in cleft grafting than patch budding (27.5%) after 15 days of grafting (Fig 1). FSW, SL, FLW, NL and DSW was 34.75 g, 74.83 cm, 21.22 g, 24.00 and 15.60 g respectively in cleft grafting compared to 33.75 g, 72.98 cm, 19.93 g, 22.25 and 13.97 g respectively in patch budding (Table 4). The FSW, SL, FLW, LL, NL and DSW was 36.97 g, 82.97 cm, 55.91 g, 15.50 cm, 25.00 and 16.90 g respectively in large sized root stock compared to 31.51 g, 64.82 cm, 48.72 g, 14.19 cm, 21.25 and 12.66 g respectively in small sized root stock. However, the leaf width was significantly higher in small sized root stock than the large sized (Table 4). These findings are in consonance with that of Mannan *et al*. (2006) who reported higher sprouting success and better growth of plants in cleft grafting in Jack fruit. The interaction root stock size and grafting method also had significant influence on all the studied growth parameters except fresh shoot weight (Table 4).

There was significant effect of the grafting method, root stock size, and interaction of grafting method and root stock size on root parameters (Table 5). The fresh weight of primary FRW (P) and secondary roots-FRW (S), number of secondary roots-NSR, length of primary root-RL(P), dry weight of primary roots-DRW(P) and dry weight for secondary roots-DRW (S) were also significantly higher in cleft grafting than in the patch budding and also in large sized root than small sized root stock (Table 5).

The FRW (P), FRW (S), NSR, RL(P), DRW (P) and DRW (S) respectively were 34.28 g, 12.29 g, 58.50, 51.98 cm, 15.76 g and 4.52 g in cleft grafting compared to 33.01 g, 10.66 g, 56.00, 46.25 cm, 14.16 g and 3.34 g in patch budding (Table 5). The FRW (P), FRW (S), NSR, RL(P), DRW (P) and DRW (S) were 37.00 g, 12.14 g, 66.75, 52.67 cm, 16.18 g and 4.35 g respectively in large sized root stock in comparison to 30.29 g, 10.82 g, 47.75, 45.55 cm, 13.75 g and 3.51 g in small root stock (Table 5). Mng'omba et al. (2010) and Mng'omba and Toit (2013) also reported that thicker root stock resulted in better sprouting success and growth of seedlings in *Mangifera indica*, *Persia armeiaca* and *Prunus persica*.

All the studied biomass characteristics were significantly influenced by root stock size, grafting method and the interaction of root stock size and grafting method (Table 6). All the studied biomass characteristics were significantly higher in cleft grafting than patch budding, and also in large sized than small sized root stock except R:S ratio which was significantly higher in small sized root stock (Table 6). The early spouting in cleft grafting (Fig 1) and also in large sized root stock and consequently longer growth period led to better growth in all biomass parameters. Mannan *et al*. (2006) also reported similar findings in cleft grafting in Jack fruit.

Table 3: Variability in fruit characteristics

Province	Location of mother tree	Fruit weight (g)		Fruit size (cm)		Pulp Weight (g)		Number of fresh fruits per kg
		Fresh	Dry	Length	Diameter	Fresh	Dry	
Jammu & Kashmir	Raya	15.59^{e}	8.10^{c}	3.72^{c}	3.29^{ab}	12.20^{f}	6.53^{a}	71^{a}
	Khadargal	20.16^{d}	7.65^{c}	5.59^{b}	2.66^{d}	17.22^{d}	5.54^{ab}	56^{b}
	Miran-Sahab	20.74^{d}	9.41^{c}	5.27^{b}	2.87^{b}	18.37^{cd}	7.97^{a}	51^{bc}
	Mangloor	18.34^{d}	7.05^{c}	3.38^{c}	2.24^{d}	15.06^{e}	5.16^{b}	55^{b}
	Keri	18.04^{d}	8.50^{c}	3.43^{c}	2.65^{cd}	14.27^{e}	6.77^{a}	56^{b}
	Dhoon	18.86^{d}	6.93^{c}	3.62^{c}	2.47^{d}	15.50^{de}	4.79^{b}	53^{b}
	Mathwar	44.60^{a}	21.75^{a}	7.90^{a}	3.38^{a}	34.78^{a}	10.79^{a}	24^{e}
	Rabta	35.80^{b}	18.00^{b}	5.67^{b}	3.55^{a}	27.21^{b}	8.71^{a}	29de
	Sandhi	26.46^{c}	13.36^{b}	5.69^{b}	3.00^{bc}	19.06^{c}	6.29^{a}	40^{cd}
	Sarore	27.00^{c}	14.85^{b}	5.49^{b}	3.03^{b}	21.33^{c}	6.39^{a}	39^{d}
	Nandini	28.00^{c}	14.28^{b}	5.40^{b}	3.06^{b}	21.34^{c}	6.42^{a}	37^{d}
	Joginder Nagar	19.91^{d}	8.93^{c}	5.08^{b}	2.66^{c}	15.81^{d}	6.91^{a}	52^{b}
Himachal Pradesh	Dhualakuan	19.42^{d}	6.21^{c}	3.56^{c}	2.24^{d}	16.19^{d}	4.42^{b}	52^{b}
	Panchrukhi-Palampur	18.86^{d}	5.94^{c}	3.59^{c}	2.31^{d}	15.98^{d}	4.45^{b}	53^{b}

$CD_{0.05}$

Fresh Fruit Weight 3.82; Dry Fruit Weight 5.41; Fruit Length 0.64; Fruit Mid-diameter 0.42; Fresh Pulp Weight 3.19 Dry Pulp Weight 5.25; Number of fruits 11.05

Means with different letters within each column indicate $p<0.05$ Tukey's significant difference for the respective characteristic

Table 4: Effect of rootstock size and grafting methods on growth of above ground parameters of elite grafts

Grafting method	Root stock size		Mean
	Small	Large	
Patch budding			
FSW	31.16^{aA}	36.31^{aA}	33.73^{A}
SL	63.75^{aA}	82.20^{bA}	72.98^{A}
FLW	46.23^{aA}	55.86^{bA}	19.93^{A}
LL	14.33^{aA}	15.55^{aA}	14.94^{A}
LW	8.75^{aA}	08.55^{aA}	08.63^{A}
NL	20.50^{aA}	24.00^{bA}	22.25^{A}
DSW	12.55^{aA}	15.38^{bA}	13.97^{A}
Cleft grafting			
FSW	31.86^{aA}	37.25^{aA}	34.75^{B}
SL	65.91^{aB}	83.75^{bB}	74.83^{B}
FLW	51.20^{aB}	55.96^{bB}	21.22^{B}
LL	14.05^{aB}	15.45^{aB}	14.75^{A}
LW	9.10^{aB}	08.51^{aB}	08.83^{A}
NL	22.00^{aB}	26.00^{bB}	24.00^{B}
DSW	12.76^{aB}	18.42^{bB}	15.60^{B}
Mean			
FSW	31.51^{a}	36.97^{b}	33.74
SL	64.82^{a}	82.97^{b}	73.91
FLW	48.72^{a}	55.91^{b}	20.58
LL	14.19^{a}	15.50^{b}	14.85
LW	08.93^{a}	08.53^{b}	8.73
NL	21.25^{a}	25.00^{b}	23.13
DSW	12.66^{a}	16.90^{b}	14.79

	$CD_{0.05}$		
	Size (S)	Method (M)	S x M
FSW	1.22	1.22	4.43
SL	3.13	3.13	2.58
FLW	1.24	1.24	0.24
LL	0.17	NS	0.22
LW	0.16	NS	1.71
NL	1.21	1.21	4.16
DSW	2.94	2.94	0.83

- Lower case letters indicate results for means comparison between small and large sized root stock for respective characteristic.
- Upper case letters indicate results for means comparison between grafting methods for respective characteristic.
- Different letters for either comparison indicate $p<0.05$ Tukey's significant difference for the respective characteristics

Abbreviations: FSW-Fresh shoot weight (g), SL- Shoot length (cm), FLW- Fresh leaf weight (g), LL- Leaf length (cm), LW- Leaf width (cm), NL- Number of leaves, DSW- Dry shoot weight (g)

Further 3 years old grafted seedling of this superior germplam (Raj Harad) planted in the field has started bearing fruits and cursory look at fruits obtained from these seedlings reveals the same size as that of the mother tree. However, the empirical observations needed to be recorded to compare fruit size of the planted seedlings with that of mother tree.

Conclusion

The mechanical scarification of the seed is the best pre-treatment to enhance seed germination followed by cowdung scarification for 45 days. The seeds can be collected from February beginning to March-mid without affecting the germination levels. The cleft grafting should be used to propagate the superior germplasm as it resulted in better sprouting success and growth of the grafted seedlings than that of patch budding. The root stock of any size could be used for grafting without any effect on the success of the grafts, however, the large stock size should be used to obtain better growth of the grafted seedlings. The field performance of the seedlings raised through this method should be tested for its effect on rotation, yield (kg/plant), fruit set heights and maximum height of the progeny. There is also a need to carry empirical studies to compare the fruit characteristics of the clonal progeny obtained through grafting with that of the mother tree.

Table 5: Effect of rootstock size and grafting methods on growth of root characteristics of elite grafts

Grafting method	Root stock size		Mean
	Small	Large	
Patch budding			
FRW(P)	29.27^{aA}	36.76^{bA}	33.01^{A}
FRW(S)	09.44 ^{aA}	11.88 ^{bA}	10.66 ^{A}
NSR	46.50 ^{aA}	65.50 ^{bA}	56.00 ^{A}
RL(P)	40.90 ^{aA}	51.60 ^{bA}	46.25 ^{A}
DRW(P)	12.23 ^{aA}	16.08 ^{bA}	14.16 ^{A}
DRW(S)	2.51 ^{aA}	4.17 ^{bA}	3.34 ^{A}
Cleft grafting			
FRW(P)	31.31 ^{aB}	37.24 ^{bB}	34.28^{B}
FRW(S)	12.19 ^{aB}	12.40 ^{bB}	12.29 ^{B}
NSR	49.00 ^{aB}	68.00 ^{bB}	58.50 ^{B}
RL(P)	50.20 ^{aB}	53.75 ^{bB}	51.98 ^{B}
DRW(P)	15.24 ^{aB}	16.27 ^{bB}	15.76 ^{B}
DRW(S)	4.51 ^{aB}	4.53 ^{bB}	4.52 ^{B}
Mean			
FRW(P)	30.29 ^{a}	37.00 ^{b}	33.65
FRW(S)	10.82 ^{a}	12.14 ^{b}	11.48
NSR	47.75 ^{a}	66.75 ^{b}	57.25
RL(P)	45.55 ^{a}	52.67 ^{b}	49.12
DRW(P)	13.73 ^{a}	16.18 ^{b}	14.96
DRW(S)	3.51 ^{a}	4.35 ^{b}	3.93

	Size (S)	Method (M)	S x M
FRW(P)	0.89	0.89	1.26
FRW(S)	0.52	0.52	0.73
NSR	6.42	6.42	4.35
RL(P)	1.54	1.54	2.18
DRW(P)	0.59	0.59	0.83
DRW(S)	1.32	1.32	1.87

Abbreviations: FRW(P) - Fresh weight of primary roots (g), FRW(S)- Fresh weight of secondary roots (g), NSR- Number of secondary roots, RL(P)- Length of primary root (cm) , DRW(P)- Dry weight of primary roots (g), DRW(S)- Dry root weight of secondary roots (g)

- Lower case letters indicate results for means comparison between small and large sized root stock for respective characteristic.
- Upper case letters indicate results for means comparison between grafting methods for respective characteristic.
- Different letters for either comparison indicate $p<0.05$ Tukey's significant difference for the respective characteristics.

Table 6: Effect of rootstock size and grafting methods on above and below ground biomass of elite grafts

Grafting method	Root stock size		Mean
	Small	Large	
Patch budding			
FAGB	77.39 aA	92.17^{bA}	84.78^{A}
FBGB	38.71 aA	48.64^{bA}	43.68^{A}
FTB	116.1 aA	140.81^{bA}	128.46^{A}
DSW	12.55 aA	15.38^{bA}	13.97^{A}
DRW(P)	12.23 aA	16.08^{bA}	14.16^{A}
DRW(S)	2.51 aA	4.17^{bA}	3.34^{A}
DAGB	29.73 aA	38.07^{bA}	33.90^{A}
DBGB	14.74 aA	20.25^{bA}	38.07^{A}
DTB	44.47 aA	58.31^{bA}	51.39^{A}
R:S(DW)	0.49 aA		
Cleft grafting		0.53^{bA}	0.51^{A}
FAGB	83.07 aB	93.61^{bB}	88.34^{B}
FBGB	43.51 aB	49.64^{bB}	46.57^{B}
FTB	126.58 aB	143.25^{bB}	134.91^{B}
DSW	12.76 aB	18.42^{bB}	15.60^{B}
DRW(P)	15.24 aB	16.27^{bB}	15.76^{B}
DRW(S)	4.51 aB	4.53^{bB}	4.52^{B}
DAGB	31.68 aB	41.94^{bB}	36.81^{B}
DBGB	19.75 aB	20.81^{bB}	41.94^{B}
DTB	51.43 aB	62.75^{bB}	57.09^{B}
R:S(DW)	0.62 aB		
Mean		0.51^{bB}	0.56^{B}
FAGB	80.23 a	92.89^{b}	86.56
FBGB	41.11 a	49.14^{b}	45.22
FTB	121.34 a	121.34^{b}	131.69
DSW	12.66 a	16.90^{b}	14.79
DRW(P)	13.73 a	16.18^{b}	14.96
DRW(S)	3.51 a	4.35^{b}	3.93
DAGB	30.70 a	40.01^{b}	35.36
DBGB	17.24 a	20.53^{b}	40.01
DTB	47.95 a	60.53^{b}	52.24
R:S(DW)	0.57 a	0.52^{b}	0.53

	$CD_{0.05}$		
	Size (S)	Method (M)	S x M
FAGB	2.25	2.25	3.18
FBGB	1.38	1.38	1.96
FTB	3.34	3.34	4.72
DSW	2.94	2.94	4.16
DRW(P)	0.59	0.59	0.83
DRW(S)	1.32	1.32	1.87
DAGB	4.22	4.22	5.97
DBGB	0.48	0.48	0.68
DTB	4.61	4.61	6.52

Abbreviations: FAGB-Fresh above ground biomass (g), FBGB-Fresh below-ground biomass (g), FTB-Fresh total biomass (g), DSW-Dry shoot weight (g), DRW(P)- Dry weight of primary roots (g), DRW(S)- Dry weight of secondary roots (g), DAGB-Dry aboveground biomass (g), DBGB- Dry below ground biomass (g), DTB- Total dry biomass (g), R:S (DW)- Root: shoot ratio dry weight basis

- Lower case letters indicate results for means comparison between small and large sized root stock for respective characteristic
- Upper case letters indicate results for means comparison between grafting methods for respective characteristic
- Different letters for either comparison indicate p<0.05 Tukey's significant difference for the respective characteristics

Acknowledgements

Authors thank SKUAST-J, Jammu and ICAR for providing financial assistance to carry out the current work through RCM and HTMM 1.18 projects respectively. The farmers of study area need special mention for their cooperation during survey work for the current work.

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