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Assessment of grafting compatibility of various *Populus* species on *Populus deltoides* rootstock

Mir Adil[1], P A Khan[1], Aashfaq A Mir[1], Sabeena Nabi[1], M A Isam[2]
[1] Division of Forest Biology and Tree Improvement, Faculty of Forestry, SKUAST-K, Jammu and Kashmir, India
[2] Division of Natural Resource Management, Faculty of Forestry, SKUAST-K, Jammu and Kashmir, India

Abstract
An experiment was conducted at the Division of forest biology and tree improvement (FBT), Faculty of Forestry, SKUAST-K. Flowering shoots (Scions) of five *Populus* species (*P. deltoides*, *P. ciliata*, *P. alba*, *P. balsamifera* and *P. nigra*) were cleft grafted on rootstock of *Populus deltoides*. Days to shoot emergence, percentage of grafting success, number of sprouted buds per cutting, number of branches, and percentage of graft survival were recorded for each species. *P. alba* buds started bursting after 16.66 days of grafting followed by *P. ciliata* (14.66 days), *P. balsamifera* (13.00 days), *P. deltoides* (12.66 days) and *P. nigra* (11.66 days). *P. deltoides* (94.00%) showed highest grafting success while as *P. alba* (70.00%) showed lowest grafting success. Number of sprouted buds per cutting were highest in case of *P. deltoides* (5.24) while as lowest were in case of *P. alba* (2.32). Number of branches were highest in case of *P. deltoides* (3.92), and lowest in case of *P. alba* (2.04). The highest percentage of grafting survival was recorded for *P. Deltoides* (88.00 %) followed by *P. ciliata* (80.00 %), *P. alba* (68.00 %), *P. nigra* (56 .00%) and *P. balsamifera* (52.00 %). In nutshell it can be concluded that the homoplastic grafting between *P. deltoides* on the rootstock of *P. deltoides* showed better grafting success and survival as compared to heteroplastic grafting.

Keywords: Scions, *Populus*, cleft graft, rootstock

Introduction

Populus, a genus of 25–30 species of deciduous flowering plants in the family Salicaceae, native to most of the northern hemisphere. English names variously applied to different species include poplar, aspen, and cottonwood. These plants are usually dioecious, rarely monoecious. Poplar trees or clones may be tall and straight (excurrent), multiple branched (deliquescent), twisted and contorted or, rarely, shrub-like in extreme environments. Poplars usually are short-lived compared to other trees, in large measure because they are host to many diseases and insect pests. The rapid growth rate of poplars enables them to reach a large size (Zhuffa, 1993) [25]. All of the taxa in the genus are dioecious, individual trees bearing either staminate (male or pollen-bearing) or pistillate (female or seed producing) flowers lacking petals and sepals that elongate from axial reproductive buds. The small, inconspicuous flowers of both sexes – numbering 30–200 – are borne on pendant catkins. Occasionally, catkins will be hermaphroditic; producing both male and female flowers, or individual trees or clones will bear both male and female catkins (Stout and Schreiner 1993) [20]. Production of abnormal flowers can vary from year to year. Catkins typically appear before the leaves in early spring, but unseasonably warm winter temperatures can force them earlier. Flowers of both sexes are borne on cup-shaped disks lacking nectaries; bracts are rapidly deciduous. The number of stamens varies widely among species and ovaries can contain two to four carpels (IPC, 1996) [3].

The genetic improvement of tree species takes time. The breeding cycles are long and this makes the process difficult. However, the selection and recombination cycles can be reduced considerably by speeding up the recommendation of new clones using grafting. Grafting is the union of two plant organs, a scion, bud or plectrum (aerial part) and a rootstock (underground part), which continue to grow, thus generating a single plant (Gil *et al*., 1986) [9].

Propagating forest species through grafts enables the preservation of desirable genotypes of high commercial or ecological value; likewise, forest diversity can be maintained and loss of genetic variety can be prevented or reduced (Vargus *et al*., 2004) [24]. Pike *et al*., (2018) [17] pointed out the importance of identifying superior individuals that are resistant to attack by pests and diseases, and of propagating such individuals by grafting. Once the scions of the superior individuals are grafted, it is possible to establish asexual seed orchards (ASOs) (Koskela *et al*., 2014) [11]. ASOs can be used to preserve genotypes and produce high quality seed (Vargas *et al*., 2004) [25], thus enhancing the success of reforestation programs and commercial forest plantations, as successful establishment of plantations will help mitigate the degradation of natural forests (Thompson *et al*., 2014) [21].

Populus grafting may prove to be very promising since grafting can induce ultra-early flowering, making the use of the Genome-Wide Selection effective. Thus, the aim of this study was to establish a standard methodology for grafting to enable flowering in *Populus* species and to check the grafting success rate of different *Populus* species on the rootstock of *P. deltoides*. Success of grafting in *Populus* will help in making this technique quite effective in the forestry sector.

Materials and methods

The experimental site is located at the Faculty of Forestry, Benhama, Ganderbal is spread over 50 ha at an altitude of 1700 m above mean sea level. The site lies on the southern aspect at 34°16'N and 74°46'E longitude. The soil formation and soil deposits are essential pre-requisites for the growth and nature of plant life.

For this study 40 cuttings (Flowering shoots) of each species of 05 *Populus* species i.e; *Populus* alba, *Populus* ciliata, *Populus* nigra, *Populus* balsamifera and *Populus deltoides* were cleft grafted on the rootstock of *Populus deltoides* present in the experimental field of FBT Division at FOF, Benhama. From each species 5 terminal cuttings were selected for studying grafting success. The grafting was performed at the beginning of growing season i.e; in the last week of Febuary before bud burst on the rootstocks that were already present in the nursery of FOF, Benhama, Ganderbal. The experiment was laid out in Randomized Complete Block Design (RCBD) with 5 treatments and 3 replications for each treatment.

For grafting study, cleft grafting method was used as suggested Hartmann *et al* (2002) [10] and Chong *et al* (2005) [5]. Cleft grafting is a method of grafting in which the scion is placed in a cleft or slit in the stock or stump made by sawing off a branch, usually in such a manner that its bark evenly joins that of the stock. Grafting was carried out as follows: tops of plants to be used as rootstocks were removed and the wedge-shaped scions (flowering shoot) were inserted into the cleft stems immediately after separation of the scions from the mother tree and bound tightly with two pieces of wetted raffia. Preparation of the scions as well as cleaving the stocks was done with new grafting knife. A high air-humidity around the scion was maintained with the aid of polyethylene bags (0.05 mm thick) which are fixed around the stem below the graft union with a metal ring. The sprouts that emerged from rootstocks below the graft portion were removed manually as when they appear in rootstocks.

Observations recorded

1. **Days to shoot emergence or first flush (days)**
   Days to first bud break or first shoot emergence were recorded by counting the days elapsed from days to grafting until to the first bud break from the scion in each treatment and the mean values were computed and used for further analysis.

2. **Percentage of grafting success (%)**
   Success percentage of grafting, the number of successful grafts in each treatment were counted at 30 days interval up to 90 days after grafting. Emergence of shoots from the terminal buds of scions were considered as success of grafting. Grafted scions which produce shoots were counted and expressed in percentage using the formula below:

$$Percentage\ of\ grafting\ success = \frac{Number\ of\ successful\ grafts}{Total\ No.of\ grafted\ rootstocks} \times 100$$

3. **Number of sprouted buds per cutting**
   Time and duration of bud burst was recorded by selecting three trees per species and five branches per tree. Date of first and last bud burst was recorded.

4. **Number of branches**
   Number of branches per plant of five randomly selected plants was counted in each treatment and then their mean calculated.

5. **Percentage of graft survival**
   After 40 days of initial success, the grafts were observed for next four months. The number of grafts survival was recorded and percentage of survival was calculated by using the formula below:

$$Percentage\ of\ grafting\ survival\ = \frac{Total\ number\ of\ grafts - Total\ \ number\ of\ death\ scion\ after\ grafting}{Total\ No.of\ graft} \times 100$$

Results and discussion

Days to shoot emergence or first flush (days): Significant differences were observed for the numbers of days to shoot emergence among five *Populus* species. Days to shoot emergence was recorded by counting the days elapsed from days to grafting until to the first shoot emergence from the scion in each treatment and the mean values were computed. *P. alba* graft started producing shoots after 16.66 days of grafting followed by *P. ciliata* (14.66 days), *P. balsamifera* (13.00 days), *P. deltoides* (12.66 days) and *P. nigra* (11.66 days).

Percentage of grafting success: Data in Table 1 revealed significant difference in grafting success among different *Populus* species. The number of successful grafts in each treatment was counted at 30 days interval up to 90 days after grafting. Emergence of shoots from the terminal buds of scions were considered as success of grafting. Grafted scions which produce shoots were counted and expressed in percentage. *P. deltoides* (94.00 %) showed highest grafting success followed by *P. nigra* (88.00 %), *P. ciliata* (86.00 %), *P. balsamifera* (86.00 %) and while as *P. alba* showed lowest grafting success of 70.00%.

Table 1: Grafting success of *Populus* species on the root stock of *Populus deltoides*

S. No	Species	Shoot emergence or 1st flush (days)	Grafting success (%)	No. of sprouted buds per cutting	No. of branches per graft	Graft survival (%)
1.	*P. deltoides*	12.66	94.00 (6.69)	5.24	3.92	88.00 (9.38)
2.	P.balsamifera	13.00	86.00 (9.27)	4.00	3.80	52. 00 (7.21)
3.	*P. ciliata*	14.66	86.00 (9.27)	4.28	5.40	80.00 (9.27)
4.	*P. nigra*	11.66	88.00 (9.38)	4.60	3.12	56.00 (7.48)
5.	*P. alba*	16.66	70.00 (8.36)	2.32	2.04	68.00 (8.24)
	CD 0.05	1.15	0.88	0.74	0.76	0.65

*Figures in parenthesis are square root transformed values

Number of sprouted buds per cutting: The cursory glance at data in Table 1 disclosed the significant difference in number of sprouted buds per graft among five *Populus* species. Five grafts from each replication were selected and number of sprouted buds counted. Number of sprouted buds per cutting were highest in case of *P. deltoides* (5.24) followed by *P. nigra* (4.60) and *P. ciliata* (4.28) while as lowest were in case of *P. balsamifera* (4.00) and *P. alba* (2.32).

Number of branches: Five grafts from each replication were selected, number of branches counted and mean computed. Number of branches were highest in case of *P. ciliata* (5.40) followed by P.deltoides (3.92), *P. balsamifera* (3.80), *P. nigra* (3.12) while as lowest in case of *P. alba* (2.04).

Percentage of graft survival: After 40 days of initial success, the grafts were observed for next four months. The number of grafts survived was recorded and percentage of survival was calculated. The highest percentage of grafting survival was recorded for *P. Deltoides* (88.00%) followed by *P. ciliata* (80.00%), *P. alba* (68%), *P. nigra* (56.00%) and *P. balsamifera* (52.00%).

The above findings are in agreement with Mehta (1969) [14] who observed the superiority of cleft grafting in his study on efficacy of various methods in propagation of walnut. A good graft success in walnut was recorded by Dwivedi *et al*., (2000) [7]. The increment in success is directly related to increase in temperature and humidity. These observations are in conformity to the findings of Ozkam and Gumus (2001) [16], who found higher success from grafts operated during February in walnut. Sharma *et al*. (2006) [18] noted that the suitable grafting time in order to achieve the maximum success in walnut is ranged from 4th week of February to 1st week of March under zero energy polyhouse condition. The variation in graft success in respect to time of walnut grafting was recorded by Tshering *et al*. (2005) [22]. The results are also in conformity with the findings of the Mudge *et al*. (2009) [12] who reported that nearly all inter-specific grafts of different citrus species are compatible. Anjarwalla *et al*. (2016) [1] also reported different grafting methods for vegetative propagation of baobab (Adansonia digitata L.) in Kenya to assist its domestication and promote cultivation. The overall grafting survival rate was 63.00%. Length of new shoots growing from the scions of the successful grafts did not significantly differ among the treatments which align with the results mentioned above.

The potential reason for variations in grafting success and survival may be taxonomic affinity, vigour and asepsis, age of scion and rootstock, environmental effects, graft compatibility-incompatibility etc.

The taxonomic affinity of the species used in heteroplastic grafting is an important factor to take into consideration, as the yield from this method tends to be lower than in homoplastic grafting (Climent *et al*., 1997) [6]. Variable results have been obtained with heteroplastic grafting, with compatibility, semicompatibility and incompatibility observed (Climent *et al*., 1997) [6], as with grafting buds of the genus Pinus onto rootstocks of Abies balsamea (L.) Mill. (Ahlgren & Wilderness, 1972) [2].

An important factor to consider in grafting is the sanitary state and vigour of the buds used, because the grafted plants are more likely to survive if the material used is in optimal condition (Upchurch, 2009) [23]. Similarly, it is essential to use rootstocks with healthy and robust roots (White *et al*., 1983). The characteristics of the scion and the rootstock are important for grafting success, and it is therefore necessary to use vegetative material of good phenotypic quality (Melchior, 1984) [13].

The scions used for grafting must be obtained from superior trees, so that the grafted trees inherit high genetic gain in the future production of seeds in ASOs (Venturini & Lopez, 2010) [24]. Superior conifer trees are considered to be those of dominant height, straight stem, desirable natural pruning, small crown occupying less than one third of the total height, insertion of branches at an angle close to 90°, and with no damage caused by pests and diseases (Prieto & Lopez, 2006) [18]. These types of characteristics are detected when the trees reach maturity, once the stems and branches clearly show their definitive physiognomy. In the processes of grafting specimens of the genus Pinus, some environmental factors cause low rates of sprouting and survival, and some intrinsic factors in the species can lead to incompatibility between the grafted organs (Cuevas, 2014) [4].

In several studies, the clones died two to three years after grafting and even after establishment in the field. The genetic, technical and cultivation factors that cause this apparent late incompatibility in grafts must be determined (Guclu, 2019) [8]. Graft incompatibility can be divided into localized and translocated incompatibility. Localized incompatibility refers to the lack of coincidence (in size or taxonomy) between the grafted organs, while translocated incompatibility depends on factors not related to the characteristics of the scion or the rootstock and that are mainly caused by inadequate application of the grafting techniques. Localized incompatibility in Pinus radiata grafts in New Zealand were observed, with no coincidence between the cambium of the scion and the cambium of the rootstock during grafting, thus hindering graft healing (Guclu,2019) [8]. On the other hand, translocated incompatibility has been observed in several trials, due to phloem damage when the grafted organs are cut, causing their degeneration and short- and long-term graft mortality (Hartmann *et al*., 2002) [10].

Conclusion

It can be concluded that *P. deltoides* showed significantly high grafting success, maximum number of sprouted buds per cutting and excellent percentage of graft survival. Whereas, *P. alba* showed lowest grafting success, minimum number of sprouted buds per cutting and minimum number of branches per graft. Lowest grafting survival was recorded for *P. balsamifera* and *P. nigra*.

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