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Impact of Pre-sowing Seed Treatments on Germination, Growth and Biomass Characteristics of *Embelia tsjeriam-Cottam*

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***Abstract*:** *Embelia tsjeriam-cottam* Burm f. is an important medicinal plant and threatened species in the Western Ghats. Due to small embryo and its abortive nature, natural regeneration through seeds is particularly challenging. Therefore, the experiment was conducted to assess the impact of different pre-sowing treatments on germination, growth and biomass characteristics. The experiment was performed in pro-trays arranged in a randomised block design replicated thrice. Among various treatments, the scarified seeds soaked in GA_3 at 750 ppm was found best treatment and improved all parameters. Parameters of germination were found to be significant however growth and biomass parameters were found to be non-significant.

Keywords: Scarification, Germination, Pro-trays, *Embelia tsjeriam-cottam*, Seed soaking

Embelia tsjeriam-cottam, also called as Malabar Emelia, is a significant, red-listed medicinal plant of India with moderate demand in the domestic and international markets according to IUCN, (Sudhakar Raja et al 2005). It is a member of the Myrsinaceae family. *E. tsjeram-cottam* is a diverse plant that can be a little shrub that grows 1 to 2 metres tall to a small tree or even a twining climbing plant in hilly regions of India. It is also found in the Himalayas, between Kashmir and Sikkim, at elevations of 400 to 1600 metres in deciduous to semi-evergreen forests. It is valued for its digestive, thermogenic, carminative, depurative, anthelmintic, and laxative properties since immemorial time and frequently (Bhattacharjee 2000). It is also used to treat malignant tumours, asthma, bronchitis, diabetes, heart-related issues, nervous system illnesses, and liver issues. A polyphenol called gallic acid and a benzoquinone called Embelin are main active ingredient of the plant which have antioxidant and anticancer effects. The National Medicinal Plants Board has prioritized 32 medicinal plants for extensive cultivation to bring about a medicinal plant revolution in our nation that will benefit people's health and prosperity; *E. tsjeram-cottam* is one of these plants due to its commercial worth. This plant is reportedly threatened in Kerala, Tamil Nadu, Karnataka, and the Western Ghats. This plant's greatest concern is its indiscriminate and unsustainable harvesting for commercial interests. Its population is declining due to several circumstances, including habitat degradation, Jhum cultivation, forest fires, and increased agricultural production. This plant has very poor regeneration. At present, *E. tsjeram-cottam* embryos are relatively tiny, and most of the seeds are sterile. Specific habitat requirements are necessary for its survival and growth. *E. tsjeram- cottam's regeneration* is sluggish and extremely poor. Due to its similar properties to those of *E. ribes,* it has grown in trade value and demand in the local market. The demand for *E. ribes* expanded significantly between 1990 and 2000, and the export volume increased to 250 t/year (Mhaskar et al 2011). Due to excessive demand, this species was also heavily wild-harvested in protected and conserved regions. Therefore, care must be taken to ensure the survival of this significant medicinal plant. The ideal optional plant for *E. Ribes* is *Embelia team-Cottam* because it shares the same qualities (Devaiah et al 2008). Due to overharvesting, overexploitation, dispersed populations result in inbreeding, the formation of abortive embryos and the sluggish germination of small, less viable seeds, its natural regeneration is limited. Due to poor seed viability and low germination rates, artificial regeneration of both species is challenging (Annapurna et al 2013). Therefore, the investigation was carried to determine the effects of different seed treatments to develop best pre sowing seed treatment.

MATERIAL AND METHODS

Experimental site and detail: The investigation was carried out at the College of Forestry Dapoli, Ratnagiri, Maharashtra

during 2017-18. The experimental site is situated at an altitude of 252 metres above mean sea level and 17°76'77" North latitude and 73°19'10" East longitude.

The required seed material was collected from the village Tulshi, Tahasil Khed, Dist. Ratnagiri. The outer mucilaginous covering present in the seeds was removed. The seeds were treated with mercuric chloride 0.1% for 10 min, washed with tap water and shade dried for 24 hours. The scarified seeds were subjected to pre-seed treatments viz., GA_3 750 ppm, Ethylene 50 ppm, KNO_3 1.0 %, H_2SO_4 2.0 %, HCL 2.0 %, H_2SO_4 2.0% + GA_3 750 ppm and HCL 2.0% + GA_3 750 ppm for 24 hours and compared with control i.e., no treatment (only scarification).

The treated seeds were sown in portrays containing coco peat media which was kept in the greenhouse under 50% shade and watering was done regularly. Observations recorded were days to first germination, germination per cent, germination rate index, mean daily germination, peak value germination, germination value, shoot height, root length, the number of leaves, diameter, shoot fresh weight, shoot dry weight, root fresh weight and root dry weight.

RESULTS AND DISCUSSION

The minimum days required for first germination (1.45 days) were recorded against seed treated with scarification + soaking seeds in HCL 2.0% + GA_3 750 ppm for 24 hours (Table 1) while, maximum % germination (10.63), germination rate index (0.29), mean daily germination (0.13), peak value germination (0.15) and germination value (8.75) were recorded in response to scarification + soaking seeds in GA_3 750 ppm for 24 hours.

Among the different germination-inducing treatments, the seeds treated with gibberellins responded well with a high rate of germination and vigorous seedling growth. Early germination was seen in seeds treated with GA_3. This may be due to the instigative action of GA_3 for the germination of seeds. GA_3 induces the *de-novo* synthesis of proteolytic enzymes like amylase and ribonuclease. Amylases in turn hydrolyse starch in the endosperm, providing the essential sugars for the initiation of the growth processes of an embryo (Weiss and Ori 2007). GA_3 treatment is also known to overrule photo dormancy, thermo-dormancy, dormancy imposed by the rudimentary embryos, mechanical barriers, and the presence of germination inhibitors (Kitchen and Meyer 1991).

Similar results of increased germination attributes such as % germination, germination rate index, mean daily germination, peak value germination and germination value were observed in *E. ribes* (Shruthi et al 2016). The rate of germination, mean daily germination, germination value and vigour were also higher due to GA_3 treatment (Masoodi et al 2000). (Gowda et al 2003) reported that GA_3 at 400 ppm considerably improved germination (48%) than the control (12%) in *Embelia tsjeram-cottam.* The findings are also supported by (Mawalagedera et al 2014) in *Phyllanthus Emblica* in which seeds were scarified and treated with 1.00% gibberellins and Pipinis et al (2012) who revealed that 30, 60 and 90 min scarified seeds of *Paliurus spina-christi* Mill. after GA_3 application (500, 1000 and 2000 ppm), resulting in higher germination per cent compared to non-

Table 1. Impact of pre-sowing seed treatments on seed germination parameters of *E. tsjeram-cottam*

Treatments	Days to first germination	(%) germination	Germination rate index	Mean daily germination	Peak value germination	Germination value
Scarification control (T1)	9.45	2.06	0.05	0.02	0.03	1.45
Scarification +soaking seeds in GA_3 750 ppm for 24 hours (T2)	3.06	10.63	0.29	0.13	0.15	8.75
Scarification +soaking seeds in Ethylene50 ppm for 24 hours (T3)	4.89	9.36	0.24	0.11	0.12	8.56
Scarification +soaking seeds in KNO_3 1.0% for 24 hours (T4)	7.06	7.94	0.22	0.09	0.11	7.13
Scarification +soaking seeds in H_2SO_4 2.0% for 24 hours (T5)	9.31	9.36	0.23	0.11	0.14	7.57
Scarification +soaking seeds in HCL 2.0% for 24 hours (T6)	1.70	4.76	0.12	0.06	0.06	4.21
Scarification + soaking seeds in H_2SO_4 2.0%+GA_3 750 ppm for 24hours (T7)	7.22	5.88	0.14	0.07	0.11	5.25
Scarification +soaking seeds in HCL 2.0% + GA_3 750 ppm for 24hours (T8)	1.45	5.40	0.16	0.06	0.08	4.53
Mean	5.52	6.92	0.32	0.15	0.10	5.93
$SE_{m(\pm)}$	0.05	1.17	0.04	0.01	0.01	1.18
CD (p=0.05)	0.14	3.55	0.13	0.04	0.04	3.58

Table 2. Impact of pre-sowing seed treatments on growth parameters of *E. tsjeriam-cottam*

Treatments	Growth parameters			
	Shoot height (cm)	Root length (cm)	Number of leaves	Diameter (mm)
Scarification (control)	3.11	3.15	1.75	1.12
Scarification + soaking seeds in GA_3 750 ppm for 24hours (T2)	4.54	5.10	2.49	1.53
Scarification + soaking seeds in Ethylene 50 ppm for 24 hours (T3)	3.72	3.51	2.27	1.39
Scarification + soaking seeds in KNO_3 1.0% for 24 hours (T4)	4.24	4.81	2.29	1.48
Scarification + soaking seeds in H_2SO_4 2.0% for 24 hours (T5)	3.28	3.51	2.16	1.37
Scarification + soaking seeds in HCL 2.0% for 24 hours (T6)	3.81	4.00	2.10	1.45
Scarification + soaking seeds in H_2SO_4 2.0% + GA_3 750 ppm for 24 hours(T7)	3.81	3.91	2.17	1.47
Scarification + soaking seeds in HCL 2.0% + GA_3 750 ppm for 24 hours (T8)	3.53	4.03	2.27	1.41
Mean	3.71	4.36	2.19	1.40
SE_m (±)	0.54	0.65	0.19	0.08
CD (p=0.05)	NS	NS	NS	NS

Table 3. Impact of pre-sowing seed treatments on biomass parameters of *E. tsjeriam-cottam*

Treatments	Biomass parameters			
	Shoot fresh Wt. (gm)	Root fresh Wt. (gm)	Shoot dry Wt. (gm)	Root dry Wt. (gm)
Scarification (control) (T1)	3.11	3.51	2.16	1.37
Scarification + soaking seeds in GA_3 750 ppm for 24hours (T2)	4.54	5.10	2.49	1.53
Scarification + soaking seeds in Ethylene 50 ppm for 24 hours (T3)	3.72	3.51	2.27	1.39
Scarification + soaking seeds in KNO_3 1.0% for 24hours (T4)	4.24	4.81	2.29	1.48
Scarification + soaking seeds in H_2SO_4 2.0% for 24 hours (T5)	3.28	3.15	1.75	1.12
Scarification + soaking seeds in HCL 2.0% for 24 hours (T6)	3.81	4.00	2.10	1.45
Scarification + soaking seeds in H_2SO_4 2.0%+GA_3 750 ppm for 24 hours (T7)	3.81	3.91	2.17	1.47
Scarification + soaking seeds in HCL 2.0% + GA_3 750 ppm for 24 hours (T8)	3.53	4.03	2.27	1.41
Mean	3.71	4.36	2.19	1.40
SE_m(±)	0.54	0.65	0.19	0.08
CD (p=0.05)	NS	NS	NS	NS

Fig. 1. Pro-trays under greenhouse

Fig. 2. Scarified seeds of *Embelia tsjeriam-cottam*

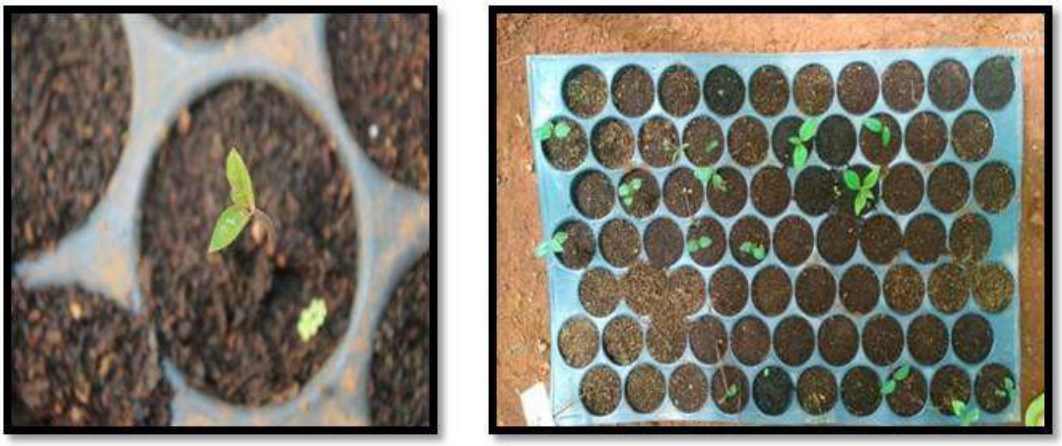

Fig. 3. Germination of *Embelia tsjeram-cottam* after 32 days

scarified seeds with GA_3. Similar results have been noticed by Maharana Rashmiprava et al (2018) in *Gmelina arborea.*

The data on the impact of scarification and seed-soaking treatments on growth parameters is presented in Table 2 and were found non-significant. The parameters related to growth such as shoot height, root length, number of leaves and diameter enhanced and recorded the highest values when scarified seeds were treated with GA_3 at 750 ppm for 24 hours (T2) and treatment T4 i.e., scarification +soaking seeds in KNO_3 1.0% for 24 hours (T4) was found to be the second better treatment. While, treatment T1 i.e., control i.e., only scarification recorded minimum values in all growth parameters.

The data recorded on the impact of scarification and seed soaking treatments on biomass parameters is found to be non-significant and given in Table 3. It has resulted that treatment T2 i.e., scarification + GA_3 at 750 ppm for 24 hours followed by treatment T4 i.e., scarification + soaking seeds in KNO_3 1.0% for 24hours increased all the parameters related to biomass such as shoot fresh weight, root fresh weight, shoot dry weight and root dry weight while, treatment T1 i.e., control (scarification) resulted in the least values.

CONCLUSION

From the investigations, it is concluded that scarified seeds of Malabar Embelia when pre-soaked with growth regulators, acids and their combinations recorded maximum values over control i.e., only scarification. All germination attributes except days to the first germination were higher when scarified seeds were treated with treatment T2 i.e., GA_3 750 ppm for 24 hours while the number of days to first germination were minimum with T8 i.e., scarification + soaking seeds in HCL 2.0 % + GA_3 750 ppm for 24 hours. Growth and biomass parameters were found to be non-significant but responded well with GA_3 and $KN0_3$.

AUTHORS CONTRIBUTION

A.D. Rane, S.S. Narkhede and V.K. Patil designed the study; Vaibhav R Jumale and Akshay Kailas Pingale collected data and developed draft of manuscript; Ankush Moran and Tapan Adhikari added additional data inputs and helped in laboratory; Pratik Santosh Kharat- Software.

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