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EVALUATION OF ORGANIC AND INORGANIC PRETREATMENTS FOR BETTER SEED GERMINATION AND SEEDLING VIGOUR IN *SANTALUM ALBUM* L.

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Abstract

Santalum album also known as East Indian sandalwood is an internationally renounced timber species because of its scented heartwood and essential oil. Propagation through the seed is the cheapest and easiest method to propagate this species. The present investigation focuses on influence of various organic and inorganic seed pretreatment on germination and seedling vigour in sandal. The mature seeds of sandal were collected from Marayoor Nachivayal forest of Idukki district, Kerala. The seeds were subjected to 16 pretreatments, which included soaking in tap and boiling water, acid scarification and soaking in GA_3, cow dung slurry and cow urine in different concentration and duration. The results of the study indicated that the highest germination percentage (74.33) was observed in $\mathbf{T_8}$ (Soaking of seeds in 500 mg l^{-1} GA_3 for 24 h) and lowest (44.33) in $\mathbf{T_5}$ (Soaking of seeds in H_2SO_4 for 5 minutes). Acid scarification and boiling water treatments decreased the germination percentage of the seeds than control. With regards to germination energy, soaking the seeds in 1000 mg l^{-1} GA_3 for 24 hours ($\mathbf{T_9}$) recorded the highest germination energy (44.67%) followed by seeds soaked in 500 mg l^{-1} GA_3 ($\mathbf{T_8}$). Soaking in cow dung slurry for 24 hours ($\mathbf{T_{10}}$) also recorded comparable germination energy (38%). The treatment with GA_3 produced the highest germination index of the seeds and the lowest values were recorded in H_2SO_4 treatments. In the present investigation, pretreatment with GA_3 emerged as the best pretreatment as proven by earlier workers, but the second best treatment was soaking in cow dung slurry, especially 24 hours duration. This is a low cost pretreatment compared to GA_3 and hence can be widely used by farmers in nurseries. However, the no definite pattern of influence of pretreatments visible in seedling traits observed after one month.

Key words : Germination value, germination index, biomass production, *Santalum album*, vigour index.

Introduction

East Indian Sandalwood or *Santalum album* (*Santalaceae*) is an important tropical tree species with the fragrant heartwood, which attracted the global market from time immemorial. It is indigenous to India and the distribution is limited to an area of about 9600 km^2, mostly in the deciduous forests of the Deccan region of Peninsular India (Gairola *et al*., 2008) of which 8200 km^2 is in the states of Karnataka and Tamil Nadu (Srinivasan *et al*., 1992). Because of its scented heartwood and essential oil used in perfumery, medicine, cosmetics and also in incense stick industries, it has attracted the global market (Srinivasan *et al*., 1992). In genus Santalum, *Santalum album* has the highest oil content (about 6%) and 80-90% of sandal oil demand (80-90%) in the international market is met by Indian sandalwood for decades. Sandal is recognized worldwide as one of the most valuable commercial tree species with an estimated market volume of more than $1 billion (Viswanath *et al*., 2008). Sandal also occurs in Kerala, it occurs sporadically in the deciduous forest up to 900 m elevation; fairly common at Marayoor. Marayur sandal population has been considered as genetically superior with the highest oil content (Venkatesan *et al*., 1995).

Sandalwood tolerates a wide range climatic and edaphic conditions and grows naturally in a variety of localities in tropical areas (Sen-Sarma, 1977). Over-exploitation and illicit felling has resulted in a great decline in the population of sandal in natural habitats and it is considered a threatened species (Meera *et al*., 2000). Hence, in order to cater to global demand afforestration

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programmes has to be hastened for that quality planting stock production is of prime importance. The major problem than can be addressed during planting stock production in nurseries is the low seed germination. Sandal starts flowering at an early age of two to three years and flowering and fruiting season vary. Ananthapadmanabha *et al*. (1991) reported that sixty per cent of the trees flower and bear fruits twice a year (during March–May and September–December), 36% once a year (September–December) and 4% throughout the year. Fruit is a drupe, globose, 1.25 cm diameter, purplish-black, with hard-ribbed endocarp. Seeds are globose (Luna, 1996). After collection, the fruits are depulped by soaking in water and rubbing and depulped seeds are dried and stored. Sandal seeds are reported to have post drop dormancy of 50-60 days due to impermeable outer covering. Literature states that the germination of sandal is sporadic and takes 4-12 weeks time to complete germination (Srinivasan *et al*., 1992 and Srimathi *et al*., 1995). Various presowing treatments are tried in sandal to obtain quick and maximum germination. Scarification and soaking in gibberellic acid have been identified as the most successful methods (Nagaveni and Srimathi 1985; Rai, 1990; Tennakoon *et al*., 2000). However, there can be other treatments which could enhance the seed germination in order to provide fast and uniform germination. The organic pretreatments like soaking in cow dung slurry, cows urine etc are successfully employed to break dormancy in many plant species. In the present study, an effort is made to compare the efficacy of organic, inorganic and other soaking treatments to enhance the germination of sandal and to study their influence on early seedling development.

Materials and Methods

Seed collection

The mature seeds of sandal were collected from Nachivayal forest of Marayoor Sandal Division, Idukki District, Kerala during October to November 2012 and the seed Lot No. KFRI-142 was assigned. To extract the seeds, the fruits were water soaked overnight and the seeds were squeezed off from the pulp. The extracted seeds were dried in shade for one day and kept for storage. The evaluation of fruit lot at Kerala Forest Research Institute Seed Centre recorded moisture content 7.89% in fresh fruits and the average number of seeds per kg was 6000 and seed size ranged from 0.2 to 1.2 cm.

Pretreatment and germination evaluation

In order to hasten the germination of sandal seeds, sixteen pretreatments were selected which include soaking in tap and boiling water, acid scarification and soaking in GA_3, cow dung slurry and cow urine in different concentrations and duration (table 1).

Hundred seeds in four replications were prepared for each pretreatment. After subjecting to pretreatment, the seeds belonging to respective treatments were sown in plastic trays filled with vermiculite as germination medium. The medium was watered daily and the daily germination counts were made on the seeds until no further germination occurred. The following observations were also made. The imbibition period *i.e*. number of days from sowing to commencement of germination of the seeds, subjected to different pretreatments was recorded. The days taken for completion of germination also was noted. From the daily germination counts, the germination percentage, germination energy, mean daily germination (MDG), peak value of germination (PV) and germination value (GV) were worked out as suggested by Czabator (1962).Germination energy was calculated as the per cent, by number of seeds in a given sample which germinate up to the time of peak germination. Mean daily germination was calculated by using the following formula

$$MDG = \sum \frac{Cumulative\ \ germination\ \ percent}{Number\ \ of\ \ days\ after\ \ sowing}$$

Peak value is the maximum mean daily germination reached at any time during the period of germination test (Czabator, 1962). Germination value was calculated as GV= PV × MDG. The germination index (GI), which is expressed as speed of germination was calculated as suggested in the Association of Official Seed Analyst (AOSA, 1983).

$$GI = \frac{\text{No. of germinated seed}}{\text{Days of first count}} + \ldots + \frac{\text{No. of germinated seed}}{\text{Days of final count}}$$

Seedling vigour

The germinated seedlings belonging to different pretreatments were grown under similar environmental condition for 30 days. The seedlings were evaluated as described in Seedling Evaluation Handbook (AOSA, 1991). At the end of the experiment all seedlings were measured for total height and collar diameter. Total number of leaf in each seedling was also counted. Ten seedlings from each treatment were randomly selected for the destructive sampling. The seedling vigour index was determined by multiplying the average germination percentage and seedling length after 30 days; the seedling length included both root and shoot length. The seeds showing the higher seedling vigour index are considered to be more vigorous (Abdul-Baki and Anderson, 1973).

Data analysis

One way analysis of variance was carried out in SPSS 17 for windows to find the difference seedling attributes due to seed pretreatments. Duncan's Multiple Range Test was conducted to compare the treatment means.

Results

Germination parameters

In general, the germination of seeds subjected to varying pretreatments ranged from 45 to 74%. Observations on germination pattern of the seeds indicated that the germination started 13 days after sowing and continued up to 152 days. Different pre-sowing treatments significantly affected various germination parameters of the seeds. The imbibition period of the seeds varied strikingly with pretreatments applied. The fastest germination *i.e.* the least imbibition period (13 days) was observed in seeds soaked in cow dung slurry for 48 hours ($\mathbf{T_{11}}$) followed by control (14 days) seeds. Whereas, the highest imbibition period (26 days) was recorded in $\mathbf{T_3}$ (water soaked for 48 hours, table 2). The time taken to complete germination also differed among the seeds subjected to various to pretreatments. The longest period to complete the germination (152 days) was observed in seeds scarified with H_2SO_4 for 1 minutes ($\mathbf{T_4}$) and the next highest was those soaked in water for 24 hours ($\mathbf{T_3}$). Whereas, the seeds soaked in cow dung slurry for 120 hours ($\mathbf{T_{12}}$) completed the germination within 103 days and those soaked in 75% cow urine ($\mathbf{T_{15}}$) recorded 104 days for completing the germination. The energy period and germination energy of the seeds also varied due to pretreatments. Germination energy is a measure of the speed of germination and hence, it is assumed, of the vigour of the seed and of the seedling which it produces. The lowest energy period (24 days) was recorded for the seeds subjected to $\mathbf{T_8}$ followed by $\mathbf{T_7}$ & $\mathbf{T_9}$ (25 days). Soaking of seeds in 1000 mg l^{-1} GA_3 for 24 hours ($\mathbf{T_9}$) recorded the highest germination energy (44.67%) followed by soaking in 500 mg l^{-1} GA_3 ($\mathbf{T_8}$). Soaking in cow dung slurry for 24 hours ($\mathbf{T_{10}}$) also recorded comparable germination energy (38%). The highest energy period was recorded for the seeds soaked in cow dung slurry for 48 hours ($\mathbf{T_{11}}$, 53 days). The lowest germination energy was observed in seeds treated with H_2SO_4 for 5 minutes ($\mathbf{T_5}$, 4.67%).

The germination pattern of the sandal seeds (cumulative germination over the entire germination period) subjected to different pretreatments is given in Fig. 1a-b. There was marked variation in germination of the seeds due to varying pretreatments. The variation germination percentage of the sandal seeds due to different pretreatments are given in fig. 2. With regard to germination percentage, the highest value (74.33) was observed in $\mathbf{T_8}$ (soaking of seeds in 500 mg l^{-1} GA_3 for 24 h) and lowest (17.00) was in $\mathbf{T_5}$ (soaking of seeds in H_2SO_4 for 5 minutes). Acid scarification and boiling water treatments resulted in lower germination percentage compared to control. Similarly soaking in tap water for 48 hours, also could not enhance the germination beyond the control seeds.

The MDG, PV and GV of the seeds subjected to different pretreatments are given in fig. 3. The highest MDG was observed in $\mathbf{T_8}$ (0.169) and $\mathbf{T_7}$ (0.161) and the lowest MDG was recorded in $\mathbf{T_4}$ and $\mathbf{T_5}$ (0.063). Similar to germination per cent, the highest MDG was recorded in seeds treated with GA_3 and the lowest was in H_2SO_4 treatment. Peak value of germination also showed similar trend, with seeds treated with 1000 mg l^{-1} GA_3 ($\mathbf{T_9}$) recording the highest value (0.583), followed by $\mathbf{T_8}$ (0.553) and the lowest was in $\mathbf{T_5}$ (0.096) and $\mathbf{T_4}$ (0.101). Germination value which is product of GV and MDG also showed similar trend, the highest germination value was recorded in $\mathbf{T_9}$ (0.093) followed by $\mathbf{T_8}$ (0.085) and the lowest was in $\mathbf{T_5}$ and $\mathbf{T_4}$ (0.006). The variation in germination index of the sandal seeds are given in the table 2. The treatment with GA_3 produced the highest germination index of the seeds and the lowest germination index was recorded in H_2SO_4 treatments.

Seedling growth and biomass production

The variation in seedling growth attributes as affected by pretreatments at the end of first month is presented in the table 3. Analysis of variance revealed significant difference in the growth attributes of the seedlings due to pretreatment at five per cent significance level. The mean shoot length of the seedlings under various treatments was the highest (18.32 cm) in $\mathbf{T_4}$ (H_2SO_4 treatment for 1 minute) and least was observed in $\mathbf{T_{11}}$ (soaking in cow dung slurry for 48 hours, 7.23 ± 0.39 cm). Highest collar diameter was recorded in $\mathbf{T_6}$ & $\mathbf{T_8}$ (2.44 ± 0.96 mm) and lowest in $\mathbf{T_3}$ (0.92 ± 0.04 mm). The highest number of leaves was observed in the seedling obtained from seeds soaked in 100 per cent cow urine for 24 hours (6.80c ± 0.51) and the lowest for the seeds soaked in cow dung slurry for 48 hours. Whereas, mean root length of the seedlings was the highest in $\mathbf{T_5}$ (15.90 ± 0.87 cm) and lowest in $\mathbf{T_3}$ (2.60 ± 0.16 cm). Hence, a definite pattern with regard to change in growth parameters due to seed pretreatments cannot be deduced.

Analysis of variance also indicated that the biomass production, root shoot ratio and vigour index of the

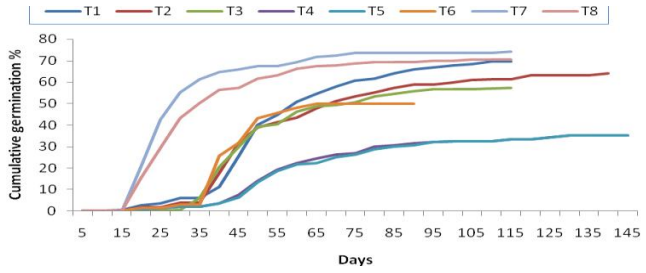

Fig. 1a : Cumulative germination per cent of the sandal seedlings under various presowing treatments.

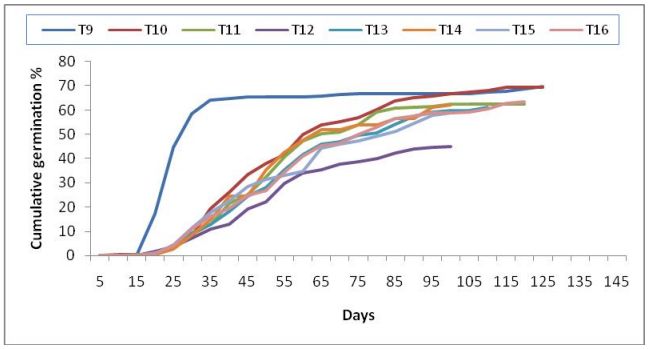

Fig. 1b : Cumulative germination per cent of the sandal seedlings under various presowing treatments.

Table 1 : The various pretreatments adopted prior to sowing of *Santalum album* seeds.

T_1	Control
T_2	Soaking in tap water for 24 hours
T_3	Soaking in tap water for 48 hours
T_4	Sulphuric acid scarification for 1minute
T_5	Sulphuric acid scarification for 5 minutes
T_6	Soaking in boiling water for 5 min
T_7	Soaking in GA_3 100 mg l^{-1} for 24h
T_8	Soaking in GA_3 500 mg l^{-1} for 24h
T_9	Soaking in GA_3 1000 mg l^{-1} for 24h
T_{10}	Soaking in cow dung slurry for 24h
T_{11}	Soaking cow dung slurry for 48h
T_{12}	Soaking in cow dung slurry for 120 h
T_{13}	Soaking in 25% cow urine for 24h
T_{14}	Soaking in 50% cow urine for 24h
T_{15}	Soaking in 75% cow urine for 24h
T_{16}	Soaking in 100% cow urine for 24h

seedlings varied significantly (p=0.05) due to pretreatments (table 4). However, a definite trend in biomass attributes could not be delineated for variation due to different pretreatments. With regards to shoot biomass production, the highest value was observed in seeds soaked in cow dung slurry for 24 hours (T_{10}, 0.156 ± 0.09 g) followed by those scarified with acid for 1 minutes (T_4, 0.074±0.01) and the lowest in Soaking GA_3

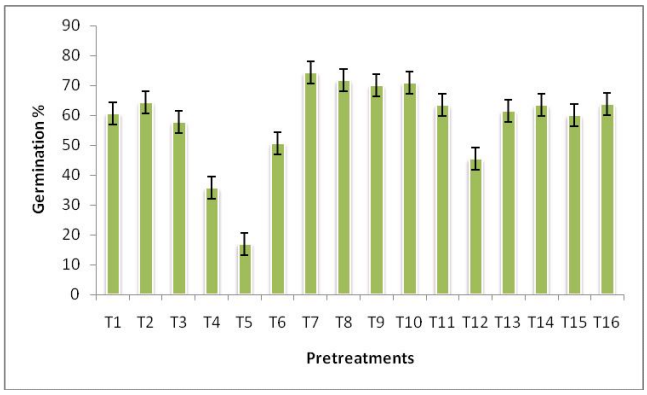

Fig. 2 : The effect of various pretreatments on germination percentage of *Santalum ablum* seeds.

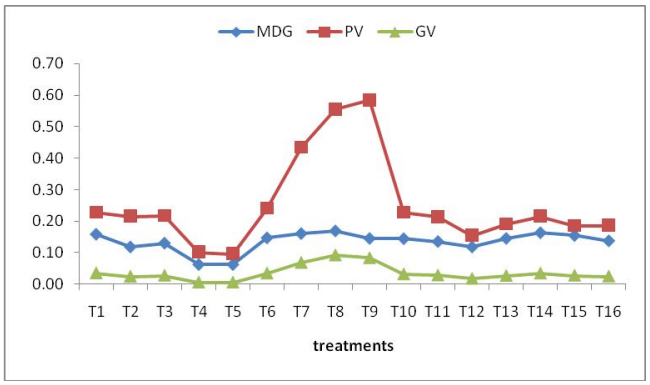

Fig. 3 : The MDG, peak value and germination value of *Santalum ablum* seeds.

1000 mg l^{-1} for 24h (T_9, 0.060 g). Whereas, Soaking in 100% cow urine for 24h recorded the highest root biomass (T_{16}, 0.080±0.01 g) and it was followed by T_{15} *i.e.* soaking in 75% cows urine (0.019±0.01 g). The lowest root biomass (0.009a±0.00 g) was recorded in control (T_1). Meanwhile, the total biomass was the highest in T_4 *i.e.* H_2SO_4 acid scarification for one minute (0.23±0.10 g) followed by T_9 (0.19±0.08 g) and the least value was recorded in the control (0.07a±0.01) seedlings. The root shoot ratio also varied significantly among different pretreatments and the highest root shoot ratio was recorded in T_{16} *i.e.* seeds soaked in 100% cow urine for 24h (0.31±0.14) and the least in T_{10} (0.13±0.01) and control (0.13±0.02). With regards to seedling vigour index, the highest vigour index was recorded in T_{16} (2053.36±71.9) and the lowest in T_5 (561.68±23.62) *i.e.* scarifying the seeds in H_2SO_4 for 5 minutes. So, we could not find a definite trend in variation of growth and biomass attributes due to pretreatments.

Discussion

Seed dormancy is the major riddle in the planting stock production of tree species. Various presowing treatments are tried on seeds all over the world to get

Table 2 : The variation in time, germination energy and germination index of *Santalum album* seeds due to pretreatments.

Pretreatment	Imbibition period	Total germination period	Energy period	Germination Energy (%)	Germination Index
	(Days)				
T_1	14	118	49	32.33	4.784
T_2	19	143	40	17.33	4.531
T_3	26	144	38	14.00	3.980
T_4	16	152	52	18.33	2.125
T_5	20	123	41	4.67	1.293
T_6	19	109	40	25.00	2.817
T_7	20	118	25	29.33	7.856
T_8	20	115	24	42.67	8.458
T_9	20	128	25	44.67	8.408
T_{10}	15	127	49	38.00	5.841
T_{11}	13	124	53	35.00	4.641
T_{12}	19	103	45	19.30	4.148
T_{13}	19	113	45	23.33	6.269
T_{14}	19	125	44	29.33	5.316
T_{15}	19	104	45	28.33	4.470
T_{16}	19	122	45	23.67	5.780

Table 3 : Growth attributes of the *Santalum album* seedlings as affected by different pretreatments.

Pretre-atment	Shoot length (cm)	Collar diameter (mm)	Leaf numbers	Root length (cm)
T_1	$14.89^{bc} \pm 0.82$	$1.18a \pm 0.11$	$5.20^{b} \pm 0.42$	$11.54^{cde} \pm 1.13$
T_2	$14.90^{bc} \pm 0.99$	$1.19^{a} \pm 0.06$	$5.20^{b} \pm 0.42$	$11.45^{cde} \pm 1.06$
T_3	$7.77^{a} \pm 0.29$	$0.92^{a} \pm 0.04$	$2.0^{a} \pm 0.00$	$2.60^{a} \pm 0.16$
T_4	$18.32^{e} \pm 0.61$	$1.47^{ab} \pm 0.05$	$6.40^{bc} \pm 0.58$	$14.51^{ef} \pm 1.44$
T_5	$17.14^{de} \pm 0.66$	$1.33^{ab} \pm 0.05$	$5.80^{bc} \pm 0.57$	$15.90^{f} \pm 0.87$
T_6	$15.40^{cd} \pm 0.57$	$2.44^{b} \pm 0.96$	$5.80^{bc} \pm 0.55$	$13.78^{def} \pm 1.44$
T_7	$13.10^{b} \pm 0.70$	$1.47^{ab} \pm 0.05$	$5.90^{bc} \pm 0.38$	$9.80^{c} \pm 0.87$
T_8	$15.40^{cd} \pm 0.57$	$2.44^{b} \pm 0.96$	$5.80^{bc} \pm 0.55$	$13.78^{def} \pm 1.44$
T_9	$8.58^{a} \pm 0.70$	$1.42^{ab} \pm 0.06$	$5.50^{bc} \pm 0.40$	$5.80^{b} \pm 0.44$
T_{10}	$14.50^{bc} \pm 0.67$	$1.28^{ab} \pm 0.07$	$5.50^{bc} \pm 0.34$	$10.83^{cd} \pm 0.91$
T_{11}	$7.23^{a} \pm 0.39$	$1.48^{ab} \pm 0.07$	$1.90^{a} \pm 0.10$	$3.55^{ab} \pm 0.23$
T_{12}	$15.53^{cd} \pm 0.81$	$1.70^{ab} \pm 0.11$	$5.40^{bc} \pm 0.50$	$15.25^{f} \pm 0.89$
T_{13}	$17.19^{de} \pm 0.69$	$1.45^{ab} \pm 0.09$	$5.30^{b} \pm 0.47$	$13.99^{def} \pm 0.54$
T_{14}	$16.48^{cde} \pm 0.70$	$1.29^{ab} \pm 0.06$	$6.60^{bca} \pm 0.40$	$13.97^{def} \pm 0.96$
T_{15}	$16.11^{cd} \pm 0.75$	$1.42^{ab} \pm 0.06$	$5.20^{b} \pm 0.33$	$11.88^{cde} \pm 1.19$
T_{16}	$17.24^{de} \pm 0.78$	$1.48^{ab} \pm 0.04$	$6.80^{c} \pm 0.51$	$15.01^{f} \pm 0.71$

Data are mean ± standard error, values with the same alphabet in the superscripts within a column are not significantly different at 0.05 level.

quick and uniform germination. There are many reports on seed dormancy and pre-sowing treatments on sandal. In sandal, the stony endocarp, although not to be called seed coat, is referred to as seed coat literally, though it is a false seed coat causes dormancy. Sandal normally takes 4-12 weeks time to complete germination (Srinivasan *et al*., 1992 and Srimathi *et al*., 1995). In the present study, the germination commenced from second week onwards and was continued up to 22 weeks in poor treatments. The earliest completion of germination took nearly 15 weeks in those seeds soaked in cow dung slurry for longer duration. Nagaveni *et al*. (1989) tried different pre-treatment like (1) soaking in water (control), 1% $ZnCl_2$, 0.5% NaOH, 100 p.p.m. IBA, 5% cytozyme, 0.5% thiourea, 100 p.p.m. IAA, 0.5% HCl or a methanolic extract of fresh sandal leaves for 4 h; (2) soaking in 1% H_2O_2 for 2 h; and (3) soaking in 10 p.p.m. kinetin for 3 h. They reported that all treatments increased speed of germination over that of the control, reducing the time for first germinants to appear from 60 to 15-45 days. In the present study, the highest time taken for the emergence of germinants was 26 days in seeds soaked in tap water for 24 hours.

Table 4 : Biomass production, root: shoot ratio and vigour index of *S. album* seedling as affected by pretreatments.

Pretreatment	Biomass (g)			Root: shoot ratio	Seedling Vigour Index
	Shoot	Root	Total		
T_1	0.063a±0.00	0.009a±0.00	0.07a±0.01	0.13a±0.02	1601.66cde±100.38
T_2	0.065a±0.01	0.011a±0.00	0.08a±0.01	0.19ab±0.01	1695.10de±97.14
T_3	0.071a±0.00	0.011a±0.00	0.08a±0.01	0.16ab±0.02	598.04a±20.09
T_4	0.074a±0.01	0.012a±0.00	0.23b±0.10	0.25ab±0.07	1171.05b±49.12
T_5	0.067a±0.000	0.012a±0.00	0.09a±0.01	0.14a±0.03	561.68a±23.62
T_6	0.062a±0.01	0.012a±0.00	0.07a±0.01	0.21ab±0.05	1478.55cd±92.91
T_7	0.073a±0.00	0.016a±0.00	0.08a±0.01	0.22ab±0.02	1641.24de±90.78
T_8	0.062a±0.01	0.012a±0.00	0.07a±0.01	0.21ab±0.05	1478.55cd±92.91
T_9	0.060a±0.00	0.010a±0.00	0.19b±0.08	0.17ab±0.02	1006.60b±44.82
T_{10}	0.156b±0.09	0.010a±0.00	0.08a±0.01	0.13a±0.01	1798.43ef±82.04
T_{11}	0.070a±0.01	0.010a±0.00	0.08a±0.01	0.15ab±0.01	682.70a±28.74
T_{12}	0.066a±0.00	0.014a±0.00	0.08a±0.00	0.21ab±0.03	1395.26c±66.53
T_{13}	0.064a±0.00	0.010a±0.00	0.07a±0.00	0.16ab±0.01	1912.27fg±61.45
T_{14}	0.065a±0.00	0.012a±0.00	0.08a±0.00	0.19ab±0.01	1928.40fg±89.11
T_{15}	0.062a±0.00	0.019a±0.01	0.07a±0.00	0.18ab±0.01	1679.40de±76.02
T_{16}	0.043c±0.002	0.080b±0.01	0.09a±0.01	0.31b±0.14	2053.36g±71.9

Data are mean ± standard error, values with the same alphabet in the superscripts within a column are not significantly different at 0.05 level.

In sandal, freshly collected seeds show dormancy for 2 month period. It is likely that the enforced dormancy of seed is due to the presence of chemical substances in the seed coat which are impervious to water and gases (Das and Tah, 2013). Ananthapadmanabha *et al*. (1988) reported that treating with dilute sodium hydroxide or dilute hydrochloric acid or gibberellic acid can remove the dormancy principle from the seed. Srimathi and Rao (1969) had reported early and quick germination in a short time of 15 days by breaking the false seed coat, indicating the presence of inhibitory principles in the seed coat. Pretreatment of seeds with GA_3 500 mg l^{-1} for 16 h resulted in 60% germination under field conditions (Nagaveni *et al*., 1989). In the present investigation, the treatment with pretreatment with GA_3 emerged as the best one as proven by earlier workers. Seed germination is promoted by gibberellin (GA) in many plant species. Abscisic Acid (ABA) is an example of a hormone (endogenous), which inhibits seed germination, while gibberellic acid (GA_3) is known to promote seed germination (Rehman and Park, 2000). Several GA signalling factors are known to induce the expression of genes encoding enzymes that mobilise food reserves, including starches, proteins and lipids, stored in the endosperm during seed germination (Peng and Harberd, 2002).

The second best treatment in the present study was soaking in cow dung slurry, especially 24 hours duration. This is a low cost pretreatment compared to GA_3 and hence can be widely used in nurseries. In *Meliadubia* seeds, the best seed treatment was treating the seeds with cow dung solution (Parthiban, 2009). Anand *et al* (2012) also reported that the *M. dubia* seed soaked in cow dung slurry for seven days increased germination speed, germination percentage and seedling growth and biomass production in comparison to the control treatments. The third best pretreatment was soaking in cow urine at different concentration. Organic presowingtreatment are found to be effective in most of the species. Sankaranarayan (1994) had reported in tamarind that soaking the seeds in 10% cow urine or in cowdung solution for 24 h increased the germination percentage from 37% (untreated controls) to 72.6 and 82.8%, respectively. The treatment with sulphuric acid and boiling water reduced the germination percentage compared to control. Hence, these treatments can be avoided.

Other than enhancing the germination many authors have reported the enhanced seedling growth due to presowing treatments (Koirala *et al*., 2000; Khan *et al*., 2001; Alamgir and Hossain, 2005; Hossain *et al*., 2005; RafiqulHaider *et al*., 2014). However, in the present study, no such definite trend was observed in seedling growth, biomass production and vigour index due to pretreatments.

Conclusion

The summarization of the present study indicates that the presowing treatments affected the germination parameter of the sandal seeds and seedling growth also. The treatment with GA_3 once again proved to be the best pretreatment, however, the organic pretreatments like soaking in cow dung slurry and cow urine also produced good germination. The sulphuric acid and boiling water treatments reduced the germination of the seeds below the control. However, the significant influence of pretreatments on seed germination was not visible in seedling traits observed after one month.

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