Seed Morphometry and Different Pre-sowing Treatments on Seed Germination of *Spondias pinnata* Linn. under Nursery Condition

Abstract

The present investigation was carried out during 2018–19 to evaluate the variation in seed dimension and most promising seed pre-treatments for accelerating germination of *Spondias pinnata* Linn. The various pre-sowing treatments were the Control (no treatment); soaking in cold water for 24 hours; soaking in cold water for 48 h; soaking in cold water for 72 h; soaking in 100 ppm thiourea for 24 h; soaking in 200 ppm thiourea for 24 h; soaking in 400 ppm thiourea for 24 h; soaking in 800ppm thiourea for 24 h; soaking in 1000 ppm thiourea for 24 h; soaking in 100 ppm KNO_3 for 12 h; soaking in 200 ppm KNO_3 for 12 h; soaking in 400 ppm KNO_3 for 12 h; soaking in 800 ppm KNO_3 for 12 h and soaking in 1000 ppm KNO_3 for 12 h. The experiment was laid out in randomised block design with 13 pre-sowing treatments and the control with three replications of 100 seeds. The highest (54.33%) germination percentage was observed from cold-water for 48 hrs followed by thiourea 1000 ppm for 24 hrs (50.00%) and potassium nitrate 200 ppm for 12 hrs (48.67%). Parameters like germination value, mean germination time, root length, shoot length and seedling vigour index were significantly different among all the pre-treatments.

Keywords: *Spondias pinnata*, germination value, seedling vigour index, thiourea

1. Introduction

Minor/ underutilized fruit crops are vital to fulfil the dietary requirement in daily life as a good supplement of major fruit crops because they possess nutritional values [1]. The minor fruit crops also essential for boosting socioeconomic conditions of the rural livelihood [2] but its potentiality and importance are lacking among the people. Among the minor fruit crops, the genus ‘Spondias’ is a small genus of tropical fast-growing trees [3]. The maximum diversification is found in the Indo-Malaysian region [4] where two indigenous and two exotic species are found in India. *Spondias pinnata* Linn. is one of the promising aromatic deciduous trees [5], locally called as ‘amara’ (Family: Anacardiaceae) found widely in moist and dry deciduous forest [6] and sporadically distributed in rural villages throughout India up to 1500 meters. This minor fruit crop plays an important role in northern parts of west Bengal [7] because of its multifarious values. The fruit is ovoid, one-seeded, yellow-skinned when ripe and appreciated for traditional food and medicinal values [8]. The flowers, raw and ripe fruits are eaten as both raw and curry [9]. It is also used as a condiment, pickles, stews, jellies, sauces and

chutneys. It is a rich source of different minerals and vitamin C [10]. Its fruit contains crude fat 12.23–12.54%, carbohydrate 16.30–23.54%, sodium 0.96–1.38%, iron 1.3–1.5%, calcium 0.15–0.93% and food energy 189–203 kcal g^{-1} [11]. The pulp contains sucrose (2.9%), glucose (1.7%), fructose (1.8%), iodine (0.45–0.61 mg kg^{-1} dry weight), riboflavin (0.02 mg $100g^{-1}$), vitamin-C (21 mg $100g^{-1}$) and vitamin-A (450 I.U. $100g^{-1}$) [12]. The fruit is astringent and antiscorbutic. The bark is useful in dysentery, diarrhoea and prevent vomiting and its paste is used as an embrocation for both articular and muscular rheumatism. Root is useful in regulating menstruation. The gum is employed as an adhesive. The leaf juice is applied in earache. The plant is reported to have anti-tubercular properties [13]. It is highly exploited for food, timber, wood products and other services like shade and boundary barriers. Despite of this importance, lack of domestication is evidenced from great morphological diversity. Its propagation is not sustainable in wild because of the biotic interference and seed borne pathogens [14–16]. If the regeneration and conservation approach will not be carried out which cause irreparable loss of genetic resources leading to extinction. It is conventionally propagated through seeds and little success obtained through vegetative propagation. A few workers/ scientists viz., Dey et al. [7], Dey et al. [17], Dey et al. [18] and Sundriyal and Sundriyal [19] has worked out on the propagation practices of *S. pinnata* and *S. axillaris*, respectively. Though, all necessary conditions for germination are available but the germination is erratic, poor and delayed due to thick seed coat that inhibits water absorption, type of seeds and size in addition to the problem of seed dormancy and longevity.

Considering its importance and demand, it is essential to develop economical methods for mass multiplication and cultivation through seed particularly in culturable wastelands under agroforestry system. Pre-sowing treatments like soaking in cold water, hot water, acid scarification, use of chemicals and growth regulators etc. are necessary to improve uniform and synchronize germination for obtaining large scale of planting materials. Therefore, the present paper was an attempt to study the seed morphometry and the different pre-sowing treatments for enhancing the seed germination.

2. Materials and Methods

2.1 Description of the study site

The experiment was carried out in the Central Forest Nursery, Department of Forestry, Uttar Banga Krishi Viswavidyalaya, Pundibari, West Bengal, in India during 2018–19. The experimental site is located at 26^0 23’ 45.8” NL, 89^0 23’ 16.7” EL and altitude of 43m where the climate is sub-tropical humid.. The average minimum and maximum temperature varied from

21.84^{0}C to 33.51^{0}C with relative humidity from 64 to 98%. The annual rainfall ranges from 2300 to 2500mm because of the pre-monsoon and monsoon periods.

2.2 Experimental design and procedure

Based on the physiological maturity, fresh fruits were collected from the surrounding areas of the university. These fruits were de-pulped manually to extract seeds and quantify the content of pulp per fruit in grams. The seeds were washed in running tap water and dried under shade at room temperature for 3 days. Seed dimension as length, width and seed weight were recorded as the ISTA [20] Rules. The experiment was laid out in randomised block design with 13 pre-sowing treatments and one control with three replications of 100 seeds. Various pre-sowing treatments were used for breaking the seed dormancy: T_1 - Control (no treatment); T_2 - soaking in cold water for 24 h; T_3 - soaking in cold water for 48 h; T_4 - soaking in cold water for 72 h; T_5 - soaking in 100 ppm thiourea for 24 h; T_6 - soaking in 200 ppm thiourea for 24 h; T_7 - soaking in 400 ppm thiourea for 24 h; T_8 - soaking in 800 ppm thiourea for 24 h; T_9 - soaking in 1000 ppm thiourea for 24 h; T_{10} - soaking in 100 ppm KNO_3 for 12 h; T_{11} - soaking in 200 ppm KNO_3 for 12 h; T_{12} - soaking in 400 ppm KNO_3 for 12 h; T_{13} - soaking in 800 ppm KNO_3 for 12 h and T_{14} - soaking in 1000 ppm KNO_3 for 12 h. The seeds were sown in polybags of 5’’x7’’ with well pulverized soil, FYM and sand in the ratio of 1:1:1. A total of 5,600 seeds were sown and watering done regularly to avoid desiccation of the seeds. Weeding was carried out as soon as required.

2.3 Method of data collection

Observation on seed germination was recorded from the date of sowing up to 4 weeks and the following parameters were recorded: Germination percent (%) = (Number of seeds germinated/ Total number of seeds sown) X 100. Germination value (GV) was calculated as per the method given by Czabator [21]. Mean Germination Time (MGT) was determined according to Dey et al. [18] as MGT= $\sum$(Daily Germination X Days)/ Number of seed sown (Un-germinated seeds at the end of the test were given values of n+1, where n is the number of days in the test). After 45 days of seed sowing five seedlings were selected and uprooted randomly from each replication and root length and shoot length was measured by digital Vernier calliper; seedling vigour index (SVI) was calculated as Final Germination Percent (%) X seedling length (cm) as per the method given by Abdul-baki and Anderson [22]. One Way ANOVA for each parameter was performed using statistical tools and mean separation for different treatment under different parameter was

performed using Critical difference (CD) test ($P \leq 0.05$). Angular transformation was done following Gomez and Gomez [23].

3. Results and Discussion

3.1 Seed dimension

The fruits of *S. pinnata* were oval shape and formed in clusters at the distal end of the branches. The fruits were light green at premature stages which turned yellow brown after ripening. The individual seed (Fig. 1) length was varied from 4.49 ±0.06 to 4.70±0.04 cm and width 3.37±0.03 to 3.52±0.03 cm. The individual fruit weight was 27.10±0.50 g which consist of 17.97±0.67 g pulp and 8.88 ±0.12 g seed weight. The moisture content in the pulp and seed was 77.91 ±0.25% and 60.23± 0.55%, respectively. The number of fruits and seeds per kilogram was 36.95 ±0.66 and 112.74±1.48, respectively. Our results are similar to studies in *S. pinnata* [16], *Pongamia pinnata* [24], *Azadirachta indica* [25] and *Elaeocarpus serratus* [26]. The variation in seed and fruit dimensions may be due to the environmental influences during the development combined with genetic variability [16].

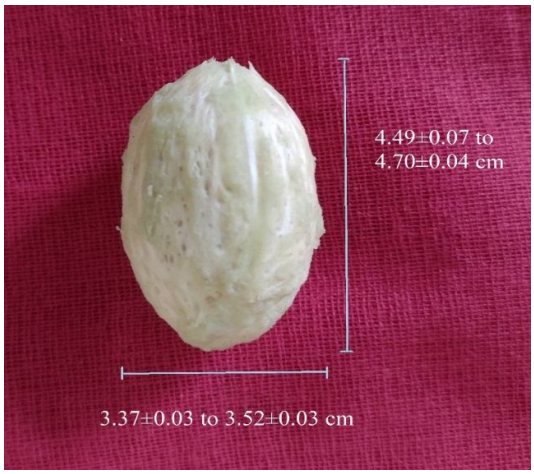

Fig. 1: Seed dimension of *Spondias pinnata.*

3.2 Effect of pre-sowing treatments on seed germination parameters

Seed lots were pre-treated with different pre-treatments to record their effects on germination percentage, germination value and mean germination time. Germination potential of seeds differed significantly due to pre-treatments (Table 1).

3.2.1 Germination percentage

A general trend of low germination response was found in all pre-treatments. The lowest germination (22%) was observed in control. Seeds soaked in cold water for 48 h had average highest germination (54.33%) followed by 50.00% in 1000 ppm of thiourea for 24 h and 48.67% in 200 ppm of KNO_3 for 12 h, respectively. We observed that the germination percentage was increased from 36.33–50.00% with the increasing concentration of thiourea where seeds were soaked for 24 h. In the treatment of KNO_3 for 12 h, the germination percentage was increased initially from 44.33–48.67% in concentration varied from 100–200 ppm and then showed declining trend gradually up to the concentration of 1000 ppm. It is general phenomenon that wide variation of seed lot showed larger variability in germination due to the variation in seed size and reserve food material. The pre-treatment namely, soaking with cold water for 48 h, 1000 ppm of thiourea for 24 h and 200ppm of KNO_3 for 12 h are likely to facilitate water imbibition through softening hard seed coat and gaseous exchange which ultimately lead to higher percentage of germination. The present results of this study are in close agreement with the findings of Dey [31] and Kumar et al. [27] in *Gmelina arborea* but to some extent contradictory with the result of Dey et al. [7] in *S. pinnata*.

3.2.2 Germination value

Irrespective of the pre-treatments, the germination value was varied from 0.63–3.96. The average maximum germination value (3.96) was notice in the seeds treated with cold water for 48 h followed by seeds soaked with 1000 ppm of thiourea for 24 h and 200 ppm of KNO_3 for 12 h recording the value of 3.35 and 3.14, respectively while the minimum (0.63) was observed in control. The germination value was followed the same pattern as germination percentage. Germination value is an expression of total germination at the end of test period with an expression of germination energy or speed of germination. It gives an idea of the vigour of the seed and of the seedling which it produces [28]. The present study strongly supports this hypothesis as seed sources having higher seed germination also had better germination value. This result is well in line with the findings of Dey [31].

Table 1: Effect of pre-treatments on germination percentage, germination value, mean germination time, shoot length, root length, total seedling length and seedling vigour index of *Spondias pinnata* seeds

Treatments	Germination percentage (%)	Germination Value	Mean germination time (Days)	Shoot length (cm)	Root length (cm)	Total seedling length (cm)	Seedling vigour index
T_1–Control	22.00^{h}	0.63^{f}	27.09^{a}	13.47^{f}	10.57^{h}	24.04^{g}	529.01^{g}

	(27.95)						
T_2–Cold Water–24h	33.33^{g}	1.44^{e}	26.70^{abc}	14.24^{f}	12.43^{fg}	26.88^{f}	889.72^{f}
	(35.26)						
T_3–Cold Water–48h	54.33^{a}	3.96^{a}	25.29^{h}	23.43^{b}	14.46^{e}	37.89^{c}	2059.18^{a}
	(47.49)						
T_4– Cold Water–72h	24.33^{h}	0.77^{f}	26.92^{ab}	13.64^{f}	11.72^{g}	25.36^{g}	615.91^{g}
	(29.55)						
T_5–Thiourea–100ppm–24h	36.33^{f}	1.73^{de}	26.55^{abcd}	16.35^{de}	14.88^{de}	31.22^{e}	1133.89^{e}
	(37.07)						
T_6–Thiourea–200ppm–24h	40.00^{e}	2.13^{d}	26.20^{cde}	24.03^{b}	15.38^{cde}	39.41^{b}	1575.13^{c}
	(39.23)						
T_7–Thiourea–400ppm–24h	45.67^{cd}	2.81^{c}	25.94^{efg}	26.99^{a}	16.74^{b}	43.73^{a}	1996.62^{a}
	(42.51)						
T_8–Thiourea–800ppm–24h	47.67^{bc}	3.02^{bc}	25.57^{fgh}	19.60^{c}	18.14^{a}	37.74^{c}	1799.92^{b}
	(43.66)						
T_9–Thiourea–1000ppm–24h	50.00^{b}	3.35^{b}	25.44^{gh}	26.81^{a}	16.02^{bc}	42.83^{a}	2141.87^{a}
	(45.00)						
T_{10}–KNO_3–100ppm–12h	44.33^{d}	2.66^{c}	25.12^{h}	13.91^{f}	12.97^{f}	26.88^{f}	1191.24^{de}
	(41.74)						
T_{11}–KNO_3–200ppm–12h	48.67^{b}	3.14^{bc}	25.74^{ef}	19.46^{c}	15.59^{cd}	35.05^{d}	1705.38^{bc}
	(44.23)						
T_{12}–KNO_3–400ppm–12h	38.67^{ef}	1.98^{d}	26.08^{def}	19.16^{c}	15.08^{de}	34.23^{d}	1324.03^{d}
	(38.44)						
T_{13}–KNO_3–800ppm–12h	38.33^{ef}	1.96^{d}	26.04^{def}	17.28^{d}	14.62^{e}	31.90^{e}	1221.96^{de}
	(38.25)						
T_{14}–KNO_3–1000ppm–12h	36.67^{f}	1.77^{de}	26.51^{bcd}	15.71^{e}	15.57^{cd}	31.28^{e}	1147.43^{e}
	(37.26)						
SEm±	0.90	0.17	0.19	0.36	0.31	0.46	57.69
CD (p= 0.05)	2.61	0.50	0.56	1.05	0.91	1.35	167.71

*Values in the parenthesis are angular transformed values.

3.2.3 Mean germination time

Significant effects of various pre-treatments for mean germination time (days) among different treatments were observed. Highest average (27.09 days) was observed in control whereas the lowest (25.29 days) was exhibited when seeds soaked with cold water for 48 h followed by 25.44 and 25.74 days was noticed in 1000 ppm of thiourea for 24 h and 200 ppm of KNO_3 for 12

h, respectively. The present findings support the hypothesis that the better germination will take lesser time (MGT) to germinate. This observation could be explained that soaking seeds with cold water for 48 h plays a great role in breaking hard seed coat very early and activity in seed embryo go faster resulting in the utilization of accumulated food material and early germination. Similar results are obtained by Roy et al. [29] in *Pinus roxburghii,* Sherpa [32] in *Michelia champaca* and Dey et al. [30] in *Baccaurea sapida*.

3.3 Growth behaviour of seedlings

Results presented in Table 1 also depict the significant difference among the pre-treatments on shoot length, root length, total seedling length and seedling vigour index.

3.3.1 Shoot length

Shoot length was showed increasing trend over the control. Among the pre-treatments, seeds soaked in thiourea 400 ppm for 24 h exhibited maximum (26.99 cm) shoot length followed by thiourea 1000 ppm for 24 h (26.81 cm) whereas seeds soaked in cold water for 48 h and KNO_3 of 200 ppm for 12 h showed 23.43 cm and 19.46 cm shoot length, respectively. The minimum (13.47 cm) shoot length was observed in control. It might be due to the cell multiplication and cell elongation in meristematic tissue of the cambium in the internodal region. Similar finding was obtained by Dey et al. [7] in *S. pinnata*.

3.3.2 Root length

Root length also showed increasing trend, but did not follow any trend among the treatments. seeds soaked in thiourea 800 ppm for 24 h exhibited highest (18.14 cm) root length whereas seeds soaked in KNO_3 200 ppm for 12 h and cold water for 48 h showed 15.59 cm and 14.46 cm root length, respectively. The lowest (10.57 cm) root length was found in control. It might be due to thiourea and KNO_3 inducing higher production of photosynthates which enhanced the requirement of abundant moisture and nutrients and increased the root length simultaneously. The finding is similar with the findings of Sherpa [32].

3.3.3 Total seedling length

All treatments showed a positive impact on the total seedling length. Overall, total seedling length was improved in treated seed lots on comparison to control; however, seeds treated with thiourea 400 ppm for 24 h were being the maximum seedling length (43.73 cm) followed by thiourea 1000 ppm for 24 h (42.83 cm) whereas total seedling length (37.89 cm and 35.05 cm)

was obtained in cold water for 48 h and KNO_3 of 200 ppm for 12 h, respectively while minimum was (24.04 cm) in control. The increased total seedling length, a summation of root length and shoot length in all treatments can be attributed to its peculiarity to increase root and shoot growth by cell enlargement and similar with the findings of Dey et al. [7] in *S. pinnata.*

3.3.4 Seedling vigour index

Seedling vigour index represents the potential level of activity and performance of seed during germination. Seeds treated with thiourea 1000 ppm for 24 h were showed highest (2141.87) seedling vigour index followed by cold water for 48 h (2059.18) which was closely followed by thiourea 400 ppm for 24 h (1996.62) whereas seeds KNO_3 of 200 ppm for 12 h exhibited 1705.38 seedling vigour index. The lowest (529.01) was observed in control. The response attributed for enhanced germination and seedling growth in thiourea 1000 ppm for 24 h are many such as diffusion of endogenous hormonal substance. These findings are also akin to the findings of Kumar et al. [27] and Dey [31].

4. Conclusion

Individual seed length and width range from 4.49±0.06 to 4.70±0.04 cm and 3.37±0.03 to 3.52±0.03cm, respectively. Seeds soaked in cold water for 48 hours had the highest germination followed by soaking seeds in thiourea 1000 ppm for 24 hours and KNO_3 200 ppm for 12 hours, in comparison to other pre-treatments. The seedling vigour index was highest in thiourea 1000 ppm for 24 hours. Hence, the above seed pre-treatment can be recommended for overcoming the physiological dormancy.

7. References

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