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Effect of Pre-sowing Kernel Treatments on Viability, Germination and Seedling Performance of Malabar neem (*Melia dubia* Cav.)

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ABSTRACT: Malabar Neem Tree, also known as *Melia dubia* Cav. is a plant with considerable medicinal and industrial economic potential. It is a multi-use tree native to tropical and subtropical areas that is mostly grown for its quick growth and adaptability for agroforestry systems. *Melia dubia* seeds have limited germination owing to their extreme hardness. Its seeds have a tough coat that hampers water penetration and germination accordingly. Seed pre-treatments are one of the factors that can significantly influence germination percentage in seeds of *Melia dubia*. This study is an effort to study seed treatments for enhancing the germination and seedling growth parameters of *Melia dubia* in germinator at laboratory condition. *Melia dubia* kernels were treated with six different pre-sowing treatments. The treatments comprised of [T_1: Normal water soaking for 24 hours, T_2: GA_3 100 ppm for 24 hours, T_3: GA_3 200 ppm for 24 hours, T_4: Thiourea (1%) for 24 hours, T_5: KNO_3 (0.2%) for 24 hours, T_6: Kernel (Control)]. The results revealed that GA_3 @ 100 ppm-soaked kernels for 24 hours significantly enhanced the germination. Maximum germination percentage (29.18) and viability percentage (41.68) was recorded in GA_3 @ 100 ppm for 24 hours (T_2) followed by GA_3 @ 200 ppm for 24 hrs (T_3). Similar trends were observed in shoot length, root length and seedling dry weight. Hard seed coat of *Melia dubia* is the reason for less quality planting material available for this species and less adaptation and awareness among farmers. The current study shows pre-sowing kernel treatments have better effects on viability, germination and seedling performance and hence, can be suggested to farmers production of quality seedling.

Keywords: *Melia dubia*, Germination, Kernels treatment, Viability.

INTRODUCTION

Melia dubia is a fast-growing tree species comes from family meliaceae. *Melia dubia* Cav. is a tree of tropical and sub-tropical regions which is mainly cultivated for its industrial and medicinal importance. It is commonly known as "Malabar Neem, Maha Neem, Gora Neem and Barma dhek". This species is exclusively found at altitudes between 1500 and 1800 m in the Sikkim Himalayas, North Bengal, upper Assam, Khasi hills of Orissa, North Circars, Deccan, and western ghats. The species is native to southern Asia and has been introduced to South Africa, Middle East, America, Bermuda, Brazil and Argentina, Australia, Southeast (SE) Asia-Pacific Islands and southern Europe (Ram *et al.*, 2014). Its existence in several countries across the world demonstrates both its capacity and adaptability to diverse environments. Germination of *Melia dubia* through seeds is very poor (14-34.3%). It has been screened as one of the best alternatives of pulpwood species, except physical barriers natural regeneration or germination has been very good under tree cover. Without any treatment, the seeds hardly germinate. The outermost, pulpy exocarp of *Melia dubia* is followed by a hard, rocky endocarp and, lastly, locules containing black seeds. Therefore, it is presumed that these physical features may prevent seed germination, however in spite of these obstacles, beneath the shelter of trees, spontaneous regeneration or germination has been quite successful. Seed pre-treatments are one of the factors that can significantly influence germination percentage in seeds of *Melia dubia*. It grows up to 6 to 20 m in height with spreading crown and straight clear bole of 10 m with tap root system. It grows in a variety of soils such as, deep fertile sandy loam soils and shallow gravelly soils. *Melia dubia*is becoming popular especially in semi-arid regions because of its hardy nature and good growth rates under diverse soil and low moisture conditions (Goswami *et al.*, 2020; Nanda *et al*. 2021; Kumar *et al*., 2021). It is deciduous tree and remains leafless during December-February. Flowering and fruiting occur in January-March and November-February, respectively. Ripened fruits are collected in Jan-Feb month. The natural germination is less than 25% (Anand *et al.*, 2012). *Melia dubia* (Malabar neem) possesses anti-cancer, anti-diabetic, anti-tumour, anti-inflammatory, antioxidant, antibacterial, anti-viral and fungicidal properties (Nagalakshmi *et al*., 2003; Thangavel *et al.*, 2019). Wood is moderately hard and 450 kg/m^3 in weight with sapwood and heartwood having grey and light red

colour, respectively. Its wood is termites resistant, used for making furniture (Saravanan *et al*., 2013) musical instruments, packing cases and agricultural implements and house construction (Suprapti *et al*., 2004), as alternative pulp wood species, fuel wood and leaf used as a fodder. Recently, *Melia dubia* is gaining popularity for generating higher income because of its fast growth habit in short duration and no transient allelopathic effect on under storey crops (Kumar *et al*., 2017a; Thakur *et al*., 2017a; Thakur *et al.*, 2017b) *i.e*., it is being widely adopted as tree component in different types of agroforestry systems (Parmar *et al*., 2019; Prajapati *et al*., 2020). *Melia dubia* tree can be a viable option which can lead to productive and profitable utilization of degraded ravine lands. In this paper, improvement in kernel germination and seedling quality parameters through pre-sowing treatments which can reduce cost of the seedling production is discussed.

MATERIAL AND METHODS

The experiment was laid out in Completely Randomized Design. The treatments comprised of [T_1: Normal water soaking for 24 hours, T_2: GA_3 100 ppm for 24 hours, T_3: GA_3 200 ppm for 24 hours, T_4: Thiourea (1%) for 24 hours, T_5: KNO_3 (0.2%) for 24 hours, T_6: Kernel (Control)] each replicated four times. The experiment was conducted in the laboratory of Department of Seed Science and Technology, CCS Haryana Agricultural University, Hisar (Haryana). The fruits were collected from plus trees of *Melia dubia* at Coimbatore (Tamil Nadu). Kernels were removed by breaking the hard seed coat of fruits by use of bench vice device. In T_1, Kernels were soaked in normal water for 24 hours. In T_2, kernels were soaked in 100 ppm solution of gibberellic acid. In T_3, kernels were soaked in 200 ppm gibberellic acidsolution for 24 hoursThe solution was prepared dissolving gibberellic acid in ethanol and dissolving100 mg of GA_3 in one litre of water and 200 ppm by 200 mg in one litre of water, respectively. In T_4, kernels were soaked in thiourea (1%) solution for 24 hours. In T_5, kernels were soaked in KNO_3 (0.2%) for 24 hours. Chemical treatment was given to seeds selected phenotypically to remove seed dormancy and enhance the germination of seedling emergence. The thiourea solution (1%) was prepared dissolving one gram of thiourea in 100 ml of water while, KNO_3 solution was prepared by dissolving 0.2 gram of potassium nitrate in 100 ml of water, respectively. In T_6, kernels which were removed from the fruits by breaking the hard seed coat were placed in sand and later into germinator at 30°C.

Viability. Kernel viability refers to the capability of a seed to germinate and produce a normal seedling. Viability of the kernels was determined through TZ test (tetrazolium test). Kernels were soaked in 0.1% solution of 2,3,5-triphenyl tetrazolium chloride in dark room at 30-35º C temperature for 24-48 hours at a pH of 6-8 and then washed with plenty of water. Viability of kernels was visually judged on the basis of colour change in the embryo. The embryo colour of the viable kernels changed to bright red while dead or non-respiring kernels remained colourless. The reason for change in colour of living kernels was due to the reduction of colourless 2, 3, 5-tripheny tetrazolium chloride to red coloured formazan by the dehydrogenase enzyme activity of the living tissue (ISTA, 2011). The kernels viability percentage was calculated using the following formula:

$$\text{Viability (\%)} = \frac{\text{Number of stained kernels}}{\text{Total number of kernels used}} \times 100$$

Germination. The number of normal seedlings was counted on the 30th day of germination in laboratory condition and pre-treated seeds were subjected for germination in a seed germinator which was maintained at 30°C and 90% humidity. Germination percentage was calculated using the following formula:

Germination (%) =

$$\frac{\text{Number of normal kernels germinated}}{\text{Total number of kernels sown}} \times 100$$

Vigour Indices. The sum total of those properties of the seed which determine the level of activity and performance of the seed lot during germination and seedling emergence.

Vigour index value was computed using the following formula suggested by Abdul-Baki and Anderson (1973) and expressed as whole number.

(i) *Vigour Index-I.* It was calculated by using following formula:

Standard germination (%) × Average seedling length (cm)

(ii) *Vigour index-II.* It was calculated by using following formula:

Standard germination (%) × Average seedling dry weight (g)

Shoot length. Ten normal seedlings from four replications of each treatment were randomly selected. Shoot length was recorded with the use of scale and average shoot length was calculated which is expressed in centimetre.

Root length. Ten normal seedlings from four replications of each treatment were randomly selected at the time of final count of standard germination. Root length was measured with scale and average root length was calculated which is expressed in centimetres.

Seedling dry weight. Seedling dry weight was measured after the final count in the standard germination test (30 days). Seedlings were dried in a hot air oven. The dried seedlings of each replication were weighed and average seedling dry weight was calculated.

Statistical Analysis. The data was analysed statistically by using the model suggested by Panse and Sukhatme (1967).

RESULTS AND DISCUSSION

Viability and germination. The perusal of data from Table 1 reveals that kernels viability and germination was significantly affected by all the treatments. The highest viability and germination (41.68% and 29.18%) was recorded in GA_3 100 ppm for 24 hours (T_2) which was significantly higher than the rest of treatments, followed by GA_3 200 ppm (T_3) for 24 hours and (38.88% and 25.88%) for 24 hours, (T_5) KNO_3 (0.2%)

for 24 hours (27.33% and 16.92%), (T_4) Thiourea (1%) for 24 hours (26.73% and 16.22%) and (T_1) normal water soaking for 24 hours (24.66% and 15.63%). Whereas, minimum was recorded in control (T_6) (20.87% and 14.78%). Maximum viability and germination percentage in gibberellic acid @ 100 ppm for 24 hours, might be because GA_3 activates the cytological enzymes which stimulate α-amylase enzyme that converts insoluble sugar into soluble sugar (Babu *et al.,* 2008), increases cell plasticity, better absorption of solvents and enhance cell division which makes early emergence of radicle. GA_3 is found to improve viability and germination in many tree species (Cavusoglu and Sulusoglu 2015). The results are in conformity with Anand *et al.* (2012) that suggested application of GA_3 enhance seed germination of *Melia dubia*. Suresh and Devakumar (2017) reported that GA_3 enhances the seed viability, germination per cent and seedling growth of *Melia dubia.* Similarly, (Babu *et al.,* 2008) discovered that soaking the seeds in GA_3 significantly increased the percentage of papaya cultivars that germinated. From 0 to 100 ppm, the GA_3 concentration considerably increased seed germination (66.17 percent) compared to the control (42.40 percent), and above this concentration, seed germination declined.

Shoot and root length. The data presented in Table 1 reveals that maximum shoot and root length (15.33 cm and 6.33 cm) was observed in GA_3@ 100 ppm for 24 hours (T_2) which was statistically at par with GA_3@ 200 ppm (T_3) for 24 hours (15.16 cm and 5.99 cm) for 24 hours, followed by (T_5) KNO_3 (0.2%) for 24 hours (12.11 cm and 4.82 cm), (T_4) Thiourea (1%) for 24 hours (11.83 cm and 4.34 cm) and (T_1) normal water soaking for 24 hours (9.77 cm and 3.66 cm). Whereas, minimum was recorded in control (T_6) (8.22cm and 2.99 cm). Babu *et al.* (2008) found GA_3 treatment had a significant effect in enhancing the seedling length and it was maximum with GA3 100 ppm (17.38 cm) as against control recording only 10.98 cm. The increase in shoot length of seedling might be due to the fact that GA_3 stimulates the cell wall to transport the calcium ions into the cytoplasm that provide more water absorption by increasing the size and eventually increase the amylase which digests the carbohydrate into sugars (Vishwakarma and Sharma 2020) and untimely roots elongation occurs for more nutrient uptake by the seedlings for photosynthesis. The results are in agreement with Anand *et al*. (2012); Krishna *et al.* (2013) who reported that soaking of fruits of *Melia dubia* in GA_3 enhanced germination and seedling growth parameters.

Seedling dry weight. The data in Table 2 showed that seedling dry weight of *Melia dubia* was significantly affected by different pre-sowing treatments. Maximum seedling dry weight was recorded (0.59g g/seedling) in GA_3@ 100 ppm for 24 hours (T_2) which was significantly higher than other treatments, followed by (T_3) GA_3@ 200 ppm for 24 hours (0.54 g/seedling) and (T_5)KNO_3 (0.2%) for 24 hours (0.39 g/seedling). The minimum seedling dry weight (0.22 g/seedling) was recorded in (T_6) control. The results are in agreement with Anand *et al.* (2012); Krishna *et al*. (2013) who reported GA_3 enhanced germination and seedling growth of *Melia dubia*. Similarly, Cavusoglu and Sulusoglu (2015) reported GA_3 treated seeds gives better seedling growth.

Vigour Index-I and Vigour Index-II. Table 2 reflects that Vigour Index-I of *Melia dubia* seedlings was significantly affected by different pre-sowing treatments. Maximum Vigour index-I was recorded (632.04) in GA_3 @ 100 ppm for 24 hours (T_2) which was significantly higher than other treatments, followed by (T_3) GA_3 @ 200 ppm for 24 hours (547.36), (T_3) and KNO_3 (0.2%) for 24 hours (286.45). The minimum Vigour index-I (165.68) was observed in control (T_6). A similar trend was also observed for Vigour Index-II of *Melia dubia* by different pre-sowing treatments.

Table 1: Effect of pre-sowing treatments on viability, germination, shoot and root length of *Melia dubia* kernels.

Treatment	Viability (%)	Germination (%)	Shoot length (cm)	Root length (cm)
T_1: Normal water soaking for 24 hours	24.66	15.63	9.77	3.66
T_2: GA_3 100 ppm for 24 hours	41.68	29.18	15.33	6.33
T_3: GA_3 200 ppm for 24 hours	38.88	25.88	15.16	5.99
T_4: Thiourea (1%) for 24 hours	26.73	16.22	11.83	4.34
T_5: KNO_3 (0.2%) for 24 hours	27.33	16.92	12.11	4.82
T_6: Kernel control	20.87	14.78	8.22	2.99
C.D. at 5%	2.54	2.15	0.38	0.68

Table 2: Effect of pre-sowing treatments on seedling dry weight and vigour index-I and vigour index-II of *Melia dubia* kernels.

Treatment	Seedling dry weight (g)	Vigour index-I	Vigour index-II
T_1: Normal water soaking for 24 hours	0.29	209.91	4.53
T_2: GA_3 100 ppm for 24 hours	0.59	632.04	17.22
T_3: GA_3 200 ppm for 24 hours	0.54	547.36	13.98
T_4: Thiourea (1%) for 24 hours	0.36	262.28	5.84
T_5: KNO_3 (0.2%) for 24 hours	0.39	286.45	6.59
T_6: Kernel control	0.22	165.68	3.25
C.D. at 5%	0.015	38.76	1.16

CONCLUSION

Native plants like *M. dubia* should be mass-propagated in a standardised manner for future commercial usage. This study suggests that one of the factors that can greatly affect the germination percentage of *M. dubia* seeds is seed pre-treatment. Based on present study, it is concluded that among all the pre-sowing treatments, GA_3 @ 100 ppm for 24 hours proved better germination and viability per cent in laboratory conditions. Similarly, seedling growth parameters such as shoot length, root length, vigour indices and seedling dry weight were observed higher in GA_3 @ 100 ppm for 24 hours. Hence, this pre-treatment could be suggested to farmers for better germination and to raise improved seedling growth of *Melia dubia*.

FUTURE SCOPE

Melia dubia seeds are hardy and resists normal germination therefore, it is necessary to find out alternatives so that planting material demand for commercially important tree species such as *Melia dubia* is fulfilled. Hence, pre sowing treatments could be an effective strategy in enhancing germination and production of quality seedlings of *Melia dubia.* It can solve many issues related to shortage of quality planting material that is being faced by the farmers. Present study could help in the direction of timely availability of quality planting material of *Melia dubia*.

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Conflict of Interest. None.

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