Some aspects of soil and seed pre-treatments on germination, growth and biomass production of Tamarindus indica seeds in the nursery

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Abstract
Tamarind is a multipurpose plant. Hence, the study investigated soil and seed pre-treatments on germination, growth and biomass production of Tamarindus indica seeds in a nursery. Completely Randomized Design with four replications was adopted and data were analysed using Analysis of Variance. Mechanically scarified seeds (MS) sowed in top soil (S1), river sand (S2), top soil plus river sand plus manure (S3) (S1MS), (S2MS), (S3MS), seeds treated with 60 % concentrated tetraoxosulphate (VI) acid (H2SO4) for 10 minutes (A10) and sowed in S1, S2, S3 (S1A10), (S2A10) and (S3A10) germinated first, 5 days after sowing (DAS), while the untreated soil (C) germinated last, 9 DAS. A10 gave (100 %) germination, while, A10 in an untreated soil (C) was the least (65 %). There was no significant effect among soil media on plant height (PH), wet weight of root (WWR), dry weight of root (DWR) and net assimilation rate (NAR). Meanwhile, pre-germination treatment of MS had higher significant values (8.30, 31.10) on PH at 2, 12 weeks after sowing (WAS), (0.51, 1.20) on (WWR) at 2, 12 WAS, (0.10, 0.13, 0.24) on (DWR) at 2-6 WAS and (0.08) on (NAR) at 6-4 WAS respectively. Seeds soaked in water for 24 hours (S24h) also showed significant effect (0.53) on DWR at 10 WAS. It is concluded and recommended that, mechanically scarified seeds at the micropyle was the effective treatment for breaking dormancy of T. indica seeds.

Keywords: Seed scarification, Tetraoxosulphate (VI) acid, Dormancy, Sowing, Root

Introduction
Tamarindus indica (commonly known as Tamarind) is a multipurpose tree of the tropics variously used as food, chemical, in pharmaceutical and textile industries, or as fodder, timber and fuel (Pugalenthi et al. 2004). T. indica has a rich plant biodiversity, with many of the plants having medicinal uses. Such a rich biodiversity could be easily lost due to over exploitation (Afolayan and Adebola 2004). However, to the domestication of this important species and its subsequent integration into the agroecosystem requires the mass production of its seedlings. Seed dormancy with its poor, slow and difficult germination hampers agroforestry and afforestation efforts (Zabala 1991).

Therefore, raising seeds of agroforestry tree species has been the fundamental exploitation for both artificial and natural regeneration, which in turn provide high quality trees from where we get high quality seeds for sowing. Propagation of trees is majorly from seeds and it is vital to have a successful plantation raised from suitable and quality seeds (Oyebamiji et al. 2014). The usual process is that seeds must germinate first and then grow seedlings. So, the seeds of T. indica must undergo pre-treatments so as to overcome seed dormancy embedded in it due to its hard seed coat for successful germination of its seeds putting various factors (type of substrate used, and environmental factors like oxygen, temperature and light) of germination into consideration (Hartmann et al. 2001; Oyebamiji et al. 2014, 2019).

A suitable growing medium also provides sufficient anchorage to the plant which serves as reservoir of nutrients and water, permits oxygen diffusion to the roots and gaseous exchange between the roots and atmosphere outside the root substrate (Abad et al. 2002; Bello and Gada 2015). It is also important to note that good and suitable nursery potting media influence the quality of seedlings and biomass production (Agbo and Omaliko 2006). Hence, the quality of seedlings obtained from a nursery has a direct effect on the establishment of any plantation, agroforestry site, a garden or an orchard (Baiyeri 2003; 2006).

Owing to these important highlighted views, it is highly necessary to identify the most appropriate pre-sowing or germination techniques suitable for raising T. indica seedlings under different potting media in the nursery. The objectives of this study were to evaluate the effect of pre-germination techniques on the seed emergence and early growth performance of T. indica, determine the best sowing media suitable for T. indica seeds and assess shoot production of T. indica seedlings in a nursery.

Materials and Methods

Study Location
This study was carried out in the Nursery Section of the Federal University Dutsin-Ma, Katsina State, Nigeria. Dutsin-ma is found within Latitude 12°27′18″ N and Longitude 07°29′29″ E. The area receives an annual rainfall of 700 mm, which is spread from May-September. The mean annual temperatures range from 29-31 C, the high temperature normally occurs in April/May and the lowest in December through February (Tukur et al. 2013).

Experimental Materials and Design
The matured and processed seeds of T. indica seeds were purchased from Wednesday market in Dutsin-ma Local Government of Katsina State. Good and healthy seeds were sorted out by simple floatation technique following the procedure of Agbogidi and Eshegbeyi (2006). The river sand used was washed to remove all particles that might affect the proper germination of the seed. The potting mixture was prepared by sieving the top soil, river sand and cow dung with mixture ratio 1:1:1 (top soil plus river sand plus manure) into finest particles using 2 mm sieve. The mixture was then put inside sixty (60) bottom perforated polythene bags of 30 cm x 30.6 cm dimension. Seeds were pre-treated by mechanical scarification using sand paper (emery cloth), seeds soaking in water at room temperature for 24 hours, seeds soaking in 60 % concentrated tetraoxosulphate (VI) (H2SO4) respectively. The total number of six hundred (600) viable seeds were pre-treated, and 10 seeds each were sowed in each polythene bag, making the total of 150 seeds per replicate. The seeds were sown at a depth of 2.0 cm in all the treatments.

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Watering was done regularly to avoid losses of seedlings. The polythene bags were kept weed free throughout the experiment. Seedling emergence was observed to have occurred once the plumule attained a height of at least 1 cm above the surface of the soil following the procedure of Ugese et al. (2007). The seedlings were observed for 12 weeks after germination (WAG) to monitor growth morphology and physiology of plant height, wet and dry weight of root and net assimilation rate respectively.

The experiment was laid in 3 x 5 factorial in Completely Randomized Design (CRD) arrangement with four replicates. The factors were; sowing media and pre-germination treatments which includes: top soil (S1), river sand (S2), top soil plus river sand plus manure (cow dung) (S3); and mechanical scarification of seeds at the micropyple (MS), seeds soaked in water at room temperature for 24 hours (S24h), seeds soaked in 60 % concentrated H2SO4 for five (5) minutes (A5), seeds soaked in H2SO4 for ten (10) minutes (A10) and no treatment (Control) (C) respectively.

Data Collection

Record of data of the experiment were taken at intervals of two (2) weeks to access different parameters which include:

**Days of Emergence:** The number of days for sprouting from each pot was observed and recorded.

**Germination Percentage (%):** the percentages of germination was derived by dividing the total number of seed germinated by the total number of seed sown and then multiply by 100, the formula is estimated below:

\[
\text{Germination percentage} = \frac{\text{Number of seed germinated} \times 100}{\text{Total number of seeds sown}}
\]

**Plant Height (cm):** The height of five seedlings from each pot were measured from the root collar to the tip of the terminal shoot using ruler in (cm).

**Wet and Dry Weight of Root (g):** One seedling from each pot was carefully uprooted washed and matter weight of root were obtained. Sensitive weighing scale was used to measure the initial wet weight of the root. After the wet weight has been taken, root components were placed in the oven and dried at 75° C for several days until constant weight was reached when the biomass was fully dried. The dried weight of biomass (roots) were then measured and recorded.

**Net Assimilation Rate (NAR):** The NAR is a measure of the amount of photosynthetic product going into plant materials i.e. it is the estimate of net photosynthetic carbon assimilated by photosynthesis minus the carbon lost by respiration. The NAR can be determined by measuring plant dry weight and leaf area periodically during growth and is commonly reported as grams of dry weight increase per square centimetre of leaf surface per week. This is also called a unit leaf rate because the assimilatory area includes only the active area in measuring the rate of dry matter production. For calculating NAR, leaf area of individual plant has to be used but not leaf area index.

\[
\text{Net assimilation rate} \ (\text{NAR}) = \frac{(W_t - W_i) \ (\log_{L_t} - \log_{L_i})}{(t - t_i) \ (L_2 - L_1)}
\]

\[L_2 \ \text{and} \ W_1 = \text{are leaf area and dry weight of plant at time} \ t_1, \ \text{and} \ L_2 \ \text{and} \ W_2 = \text{are leaf area and dry weight of plants at time} \ t_2 \]

Data Analysis

Data were analyzed using Analysis of Variance (ANOVA) with the statistical Analysis System (SAS 2003) at 5 % level of significance to determine differences in the treatment effect. The Fisher’s Least Significant Difference (F- LSD; P ≤ 0.05) was used to separate the means of differences among the treatments.

Results

**Days of Emergence**

Seeds mechanically scarified (MS) and seeds treated with tetraoxosulphate (VI) acid (H2SO4) for 5 minutes had the lowest days of emergence at (5) DAS and seeds not treated (C) had the highest days of emergence in top soil + river sand + manure in 1: 1: 1 (S3) (Figure 1).

![Figure 1: Showing days of emergence of Tamarindus indica seeds after sowing](image)

**Germination Percentage (%)**

Seeds treated with tetraoxosulphate (VI) acid (H2SO4) for 10 minutes (A10) sowed in river sand (S2) had the highest germination percentage (100 %) (S2A10) follow by seeds mechanically scarified (MS) in top soil (S1) had (97.5 %) (S1MS) and the lowest is the seeds not treated (C) with germination percentage of (65 %) in top soil (S1) (S1C) (Figure 2).

![Figure 2: Showing germination percentage (%) of Tamarindus indica seeds after sowing](image)

**Plant Height (cm)**

There was no significant effect on plant height across all the period of the experiment by the various soil media. However, seeds mechanically scarified at the micropyple had significantly higher values (8.30, 31.10) on plant height at 2 and 12 WAS respectively among other treatments (Table 1).

![Table 1: Data recorded for different parameters of Tamarindus indica seedlings](image)
Table 1: Response of various soil media and pre-germination treatments on plant height at interval of 2 WAS

| Soil media | Treatment | 2 WAS | 4 WAS | 6 WAS | 8 WAS | 10 WAS | 12 WAS |
|------------|-----------|-------|-------|-------|-------|--------|--------|
| S1         | 0.20      | 0.30  | 0.44  | 0.82  | 1.00  | 1.20   |
| S2         | 1.53      | 0.30  | 0.50  | 0.83  | 0.93  | 1.20   |
| S3         | 0.20      | 0.30  | 0.50  | 0.83  | 1.00  | 1.20   |
| SE±        | 0.455     | 0.012 | 0.019 | 0.027 | 0.037 | 0.021 |

Table 2: Response of various soil and pre-germination treatments on wet weight root at interval of 2 WAS

| Soil media | Treatment | 2 WAS | 4 WAS | 6 WAS | 8 WAS | 10 WAS | 12 WAS |
|------------|-----------|-------|-------|-------|-------|--------|--------|
| S1         | 0.20      | 0.30  | 0.44  | 0.82  | 1.00  | 1.20   |
| S2         | 1.53      | 0.30  | 0.50  | 0.83  | 0.93  | 1.20   |
| S3         | 0.20      | 0.30  | 0.50  | 0.83  | 1.00  | 1.20   |
| SE±        | 0.455     | 0.012 | 0.019 | 0.027 | 0.037 | 0.021 |

**Dry Weight of Root (g)**

Seed sowed in soil different media had no significant effect on dry weight of root across all the period of the experiment. Nevertheless, seeds mechanically scarified (MS) and seeds soaked in water for 24 hours (S24h) had significantly higher values (0.10, 0.13, 0.24) and (0.53) on dry weight of root at 2-6 WAS and 10 WAS respectively (Table 3).

Table 3: Response of various soil and pre-germination treatments on dry weight root at interval of 2 WAS

| Soil media | Treatment | 2 WAS | 4 WAS | 6 WAS | 8 WAS | 10 WAS | 12 WAS |
|------------|-----------|-------|-------|-------|-------|--------|--------|
| S1         | 0.08      | 0.11  | 0.21  | 0.47  | 0.49  | 0.62   |
| S2         | 0.08      | 0.11  | 0.22  | 0.43  | 0.48  | 0.62   |
| S3         | 0.09      | 0.12  | 0.22  | 0.42  | 0.52  | 0.63   |
| SE±        | 0.006     | 0.005 | 0.011 | 0.016 | 0.021 | 0.011  |

**Net Assimilation Rate (NAR)**

There was no significant effect of soil media on net assimilation rate across all the period of the experiment. However, seeds mechanically scarified had significantly higher value (0.08) on net assimilation rate at 6-4 WAS (Table 4).

Table 4: Response of various soil and pre-germination treatments on net assimilation rate

| Soil media | Treatment | 4 – 2 WAS | 6 - 4 WAS | 8 – 6 WAS | 10 – 8 WAS | 12 – 10 WAS |
|------------|-----------|-----------|-----------|-----------|------------|------------|
| S1         | 0.03      | 0.07     | 0.14     | 0.05      | 0.05       |
| S2         | 0.08      | 0.08     | 0.15     | 0.04      | 0.06       |
| S3         | 0.04      | 0.07     | 0.14     | 0.05      | 0.05       |
| SE±        | 0.006     | 0.005    | 0.074    | 0.008     | 0.005      |

**Pre-germination treatments**

| Soil media | Treatment | 4 – 2 WAS | 6 - 4 WAS | 8 – 6 WAS | 10 – 8 WAS | 12 – 10 WAS |
|------------|-----------|-----------|-----------|-----------|------------|------------|
| S1         | 0.03      | 0.08     | 0.14     | 0.05      | 0.06       |
| S2         | 0.02      | 0.08     | 0.13     | 0.06      | 0.04       |
| S3         | 0.04      | 0.07     | 0.15     | 0.06      | 0.05       |
| SE±        | 0.008     | 0.006    | 0.009    | 0.010     | 0.019      |
Discussion

Of the results, there was no significant difference (P ≤ 0.05) among the soil media on plant height, wet and dry matter weight of root and net assimilation rate. This is an indication that soil media used do not interfere with germination of Tamarindus indica seeds as long as correct and adequate pre-sowing treatments were carried out on them. Seeds that mechanically scarified were significant on plant height at 2 and 12 WAS, wet weight of root at 6 and 12 WAS and dry weight of root at 2-6 WAS and net assimilation rate at 6-4 WAS respectively. Seed germination of T. indica seeds started 5 days after sowing (DAS). Mechanically scarified seeds sowed in top soil (S1;MS), seeds treated with 60 % concentrated tetraoxosulphate (VI) acid (H2SO4) for 10 minutes and sowed in top soil (S2;AI0), mechanically scarified seeds sowed in river sand (S3;MS), seeds treated with H2SO4 for 10 minutes and sowed in river sand (S4;AI0), mechanically scarified seeds sowed in top soil plus river sand plus manure (S1;AI0) and seeds treated with H2SO4 for 10 minutes and sowed in top soil plus river sand plus manure (S1;AI0) germinated first, 5 DAS, while the untreated soil (C) germinated last, 9 DAS. A10 gave (100 %) germination, while, A10 in an untreated soil (C) was the least (65 %). This implies that the difference in the days of emergence and germination rate of the seeds in each treatment was a result of pre-germination treatments carried out on the seeds. Dormancy in seeds was overcome, speeding up their germination as also stated by (Roshetko 1995; Agboola 2002).

Seeds of T. indica treated with 60 % tetraoxosulphate (VI) acid (H2SO4) for 10 minutes (A10) in soil media of top soil plus river sand plus manure (S1) had the highest germination percentage (S2;AI0) (100 %) in all treatments and soil media, followed by the seeds mechanically scarified (MS) in soil media of top soil plus river sand plus manure (S1) (97.5 %) and seeds not treated at all (control) (C) gave the least percentage in top soil (S1) (65 %) of germination respectively. These results indicated that seeds soaked in concentrated acid for a short time tend to show better germination performance and it also improves seed germination rate (Olatunji et al. 2012). It means that seeds that stayed longer in acid have the risk of damaging their cotyledon and this will eventually hamper germination as reported by Ariana et al. (2011). It was generally observed that mechanically scarified seeds had higher effect on plant height, wet and dry weight of root and net assimilation rate. This is possible as a result of abrasion effect that has taken off the hard seed coat of the Tamarindus seeds and thereby provided access for easy penetration of water and oxygen and thereby improved rapid germination of the seeds (Oyebamiji et al. 2019). Hence, the emergence of the seeds was faster in the seeds mechanically scarified and consequently affected its complete germination leading to highest germination percentage (100 %) and is in agreement with Oyebamiji et al. (2014) report who said that seeds scarified at the micropyle region experience faster germination of seeds and higher percentage rate.

Conclusions

Since the seed is fundamental in Silviculture for both artificial and natural regeneration, dormancy of the viable seeds has to be overcome for massive and improved tree establishment. It is obvious from this study that the mechanical scarification of T. indica seeds with sand paper at the micropyle was an effective treatment for breaking dormancy of T. indica seeds. It is therefore concluded that seeds mechanically scarified in all the soil media had the shortest days of emergence with five 5 DAS across all the soil media, while seeds not pre-treated gave the longest days of emergence with 10 DAS. Mechanically scarified seeds at the micropyle showed the highest values (97.5 %) while seeds not treated (control) gave the least germination percentage (65 %). It is then recommended that mechanically scarified seeds at the micropyle should be used by farmers, foresters and tree planter as the effective treatment for breaking dormancy of T. indica seeds for forest establishment.

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