Some Aspect of Dormancy Studies and Vitamin D Content of Four Tree Seed Species

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Abstract:
Selected leguminous tree seeds of Tamarindus indica, Parkia biglobossa, Prosopis africana and Albizia lebbeck play prominent roles in nutrient recycling of soils in both savanna and tropics of Nigeria. These trees are recklessly fell without a corresponding afforestation programme. Therefore, the danger of being extinct is imminent. However the seeds of these trees were found to be dormant. The viability test used was floating methods. Efforts to alleviate their dormancy led to the usage of some dormancy-breaking methods which involve chemical scarification of concentrated sulphuric acid at 5min, 10min and 15mins. The viability test of the seeds showed 90-100%. Initial germination trials gave 0-20% in all the seeds. About 80-100% germination was observed when treated with concentrated sulphuric acid under 5mins duration within 10days. At 10mins duration the treatment was 100% in A. lebbeck while maximum percentage germination were obtained in A. lebbeck (100%) and P. biglobossa (60%) at 15min duration. The highest vitamin D content was found in P. africana. This work will assist in the propagation of these valuable leguminous seeds; and revealed the potentials of these tree seeds for commercial and domestic purposes.

Key words: Dormancy, Vitamin. D, Germination, Prosopis africana, Tamarindus indica, Parkia biglobossa, Albizia lebbeck

INTRODUCTION

Dormancy in seed is regarded as a state whereby an intact and viable seed fails to germinate or to complete germination under favourable conditions. The seeds of some leguminous plants are prevented from completing germination because the embryos are sometime constrained by its surrounding structures. This phenomenon could be termed coat-enhanced dormancy state. Embryos isolated from these plants are not mostly dormant. A second category of dormancy is found in which the embryo itself is dormant, in such that it does not support the completion of germination. This state could be termed embryo-enhanced - dormancy. Studing germination may be difficult because population of seeds do not complete the process. Synchronous release from dormancy may be more erratic because of the threshold stimulus require to promote germination varies widely among individual seeds. A “biotime” concept has been introduced which incorporates mathematical model to characterize and predict seed germination behaviour with respect to dormancy and the factors that influence it. Is dormancy the result of a deficiency in some vital cellular event of germination? Is there some dormancy imposed event that must be negated before germination can be completed? A broader issue is weather release from dormancy could be triggered by a variety of environmental and chemical stimulus is mediated through a common signal transduction chain that coordinates diverse cellular responses but may differ between the seeds the seeds of different species and dormancy types. It has been suggested that there are related or common receptors for dormancy breaking agents within the plasma of the responsive embryonic cells. When triggered, these receptors then initiate a signal transduction cascade, perhaps involving
synthesis of or sensitization to germination-promoting gibberellins (GAs) that leads to the completion of germination (Hilhorst, 1993; Vleeshouwers, et al., 1995). Changes in the phosphorylating activity of membrane associated Ca^{2+}-dependent protein kinases that lead to dormancy or germination has been proposed as well. Vitamins are organic compounds required in trace amount in the diet for healthy growth and reproduction. They are natural materials that can be isolated from biological organisms (Bieri, 1976). The vitamin D are classified as one of the fat-soluble vitamins, because of their ability to dissolve in organic solvents. It can be transported and stored for long period of time in a manner generally similar to that of fats. Unlike the water-soluble vitamins, the fat-soluble vitamins generally do not function as co-enzymes and rarely utilized by microorganisms (Carpenter, 1974). Vitamin helps to increase the calcium absorption and assist in bone growth and teeth development.

This work focus on the dormancy studies and vitamin contents of valuable leguminous tree seed species in Nigeria.

MATERIALS AND METHODS

Seed Collection and Processing:
Matured seeds were collected and extracted from their parent plant after fruit fall. Pods of Tamarindus and Parkia seeds were soaked for 24hours while that of Prospopis and Albizia were hit with gentle strokes of moderate size stone. Dried pods of the latter were collected from the campus of the University of Ilorin, Ilorin, Kwara state of Nigeria. The seeds of Tamarindus were bought in Murbi market of Adamawa state, Nigeria. The dried pulps of Parkia were collected from its tree found at the back of female hostel of the University of Agriculture, Abeokuta, Ogun state, Nigeria.

Seed Viability Study:
The viability of the seeds was carried using simple floating test. Seeds were immersed in distilled water contained in 500ml beaker. Those seeds found floated were considered to be damaged or bad for germination. Such seeds were discarded. Whereas, those that settled at the bottom of the water were termed to be viable for germination. Such seeds were put in container and placed in the refrigerator set at 15-20 oC. However, the percentage germination were observed at each case.

Initial Germination Studies:
The seeds could not germinate when slated for it under natural conditions. This established the fact that the seeds exhibit physical dormancy, having considered that the seeds were viable and could readily germinate if not for dormancy problems.

Chemical Scarification:
Seeds were prepared at different lots and immersed in concentrated Hydrogen Tetraoxosulphate VI acid for the period ranging from 5mins, 10mins and 15mins. The set-up were raised in several changes of distilled water and air dried at room temperature. Thereafter, they were put in 9cm diameter petridishes containing two sterile filter papers soaked in distilled water. The set up were observed for some days while percentage germination of each was observed.

Vitamin Extraction:
Approximately 0.125gram of sample was measured into a set of clean test tubes and marked to 500ug volume with distilled water. About 10g/L of ascorbic acid was added as an antioxidant and shake for 15mins followed by 5mins of sonication. Triton of 0.5g/L was added as detergent and 400uL acetonitrile was added as welland mixed thoroughly, then 400uL n-hexane was measured and added and shook vigorously for 4 units. The mixture was centrifuged for 2mins at 800RPM. The supernatant for vitamins determination was collected over HPLC. Separation of vitamin was carried out using BDS hypersil CN 150min, 5um column in combination with a Javely NH_{2} guard column at wavelength 325 (Cook, 1998).

Results:
Table 1 showed percentage viability of various leguminous tree seed species after “floating testing”. The T.indica, A.lebbeck and P. africana had 100% germination. P.biglobossa gave 90% germination.
Table 1: Percentage Viability Test.

<table>
<thead>
<tr>
<th>Tree Seed Species</th>
<th>Percentage Viability</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tamarindus indica</td>
<td>100%</td>
</tr>
<tr>
<td>Parkia biglobossa</td>
<td>90%</td>
</tr>
<tr>
<td>Albizia lebbeck</td>
<td>100%</td>
</tr>
<tr>
<td>Prosopis Africana</td>
<td>100%</td>
</tr>
</tbody>
</table>

The poor germinability of the leguminous seeds were exhibited in the initial studies shown in Table 2. Seeds prepared for germination without prior germination treatments that is, breaking the barrier of germination had reduced the rate of germination to 0-20%, 0-10%, 0-10%, and 0-10% for T.indica, P.biglobossa, A.lebbeck and P.biglobossa respectively (Table 2).

Table 2: Initial Germination Studies.

<table>
<thead>
<tr>
<th>Tree Seed Species</th>
<th>Percentage germination</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tamarindus indica</td>
<td>0-20%</td>
</tr>
<tr>
<td>Parkia biglobossa</td>
<td>0-10%</td>
</tr>
<tr>
<td>Albizia lebbeck</td>
<td>0-10%</td>
</tr>
<tr>
<td>Prosopis Africana</td>
<td>0-10%</td>
</tr>
</tbody>
</table>

Treatment of the seeds with concentrated sulphuric acid gave rise to the percentage germination of the seeds. This is evident in the in Albizia, Parkia and Tamarindus having 80-100% germination under 5minute duration treatments. The control showed 0% germination within 10days (Fig.1).

Fig. 2, showed at 10mins, the treatment of concentrated sulphuric acid and its treatment on percentage germination of the leguminous seeds. A.lebbeck seeds showed percentage germination of 100%. However, 20% germination was showed for P.africana and P.biglobossa at each case while T.indica and control were 0% germination within 10days.

At 5mins scarification duration, maximum percentage germination of the seeds were observed to be 100% and 60% in A.lebbeck and P.biglobossa respectively. However, P.africana, T.indica and control showed 0% germination within 10days (Fig.3).

The highest vitamin D content of the seeds was found in P.africana having 0.8μg/ml while T.indica, P.biglobossa and A.lebbeck gave 0.7μg/ml, 0.75μg/ml and 0.75μg/ml respectively (Fig.4).

Discussion:

Viability remained high in T.indica, P.biglobossa, P.africana and A.lebbeck seeds. This implied that the embryos are in good state and that the physiological being of these seeds were not affected. Therefore, the seeds were in readiness for germination to take place, provided all the conditions necessary for germination were present.

The initial germination test revealed that the seeds exhibited physical dormancy due to hardness of the seed coats, when raised for germination without pre-germination treatments. This had led to high reduction of their germination potentials. Seeds may exhibit one form of dormancy or the other which has to do with the inability of water and other important materials such as gases and other metabolites to pass into the
embryo through the seed coats. The seed coats of dormant seeds prevented the influx of these materials to mobilize the food reserves in the embryo via imbibition of water in it that may lead radicle protrusion and later plumule. These are evidence of germination in viable and dormant-free seed. Some seeds lose their dormancy while in dry state when the rate of metabolism is very low. However, imbibed dormant seeds are metabolically active and this may receive an external signal like light, chilling alternating temperature and chemical or hormonal treatments that can stimulate germination (Bewley, 1997).

Fig 2: Effect of Sulphuric acid scarification on germination of leguminous tree seeds at 10mins duration.

Fig 3: Effect of sulphuric acid scarification on germination of leguminous tree seeds at 15mins duration.

Fig 4: Vitamin D contents of Four Leguminous Tree Seed species.
Termination of dormancy in seeds of *T.indica, P.biglobossa, A.lebbeck* and *P.africana* occurred by pre-treating them with concentrated sulphuric acid as a pre-sowing agent to rupture the surrounding walls of the hard seed coats after 5min, 10min and 15 min of duration. The primary events in the release from dormancy are the reception of stimulus by the reception by the embryo and the immediate signal transduction chain that led to the secondary events which could involve metabolic and hormonal changes (Bewley, 1997). The result is emergence of the embryo axis from the seed, which is the completion of germination. (Bewley, 1997). *P. Africana* showed the highest content of vitamin D (0.8ug/ml) content. This implied that the seeds are relatively rich in its content. Therefore it could be used as alternative for vitamin D in daily diets. Vitamin D is good in improving healthy growth and development. The seeds could therefore be used as a substitute of vitamin D in daily diets.

REFERENCES