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Application of incubation-drying-separation method for viable and dead seeds fractions in *Anogeissus latifolia*

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ABSTRACT

In the present study the IDS (Incubation-Drying- Separation) technique was tested on a seed lot of *Anogeissus latifolia*. Data envisaged that above criteria based on the extent of temperature and drying as found in nature is suitable and provide accurate result for separating viable and dead seed fractions. All drying treatments gave 73.25 to 100% germination in sunken fraction compared to 12.50% for untreated seeds. The aim of this study was to improve seed lots quality by remaining non viable seeds using above technique which is desirable to produce uniform and vigorous seedlings in the nursery. The seed vitality of floated or sink was also determined by 2, 3, 5 triphenyl terazolium chloride test.

Figure : 00 References : 21 Tables : 02

KEY WORDS : *Anogeissus latifolia*, Empty seeds, Filled seeds, Incubation drying separation (IDS).

Introduction

Anogeissus latifolia is an important tropical forest tree species, belongs to the family Combretaceae and popularly known as axlewood, dhau, dhaur, dhawa, It is a slow growing moderate sized deciduous tree occurs throughout the Indian sub continent except Eastern part of Bengal and Assam[19]. It is also found in drier regions of Sri Lanka and Nepal[6]. The leaves are used for tanning as they contain large amount of gallotannins. It is also the source of Indian gum, known as ghatli gum used in calico painting[2]. The germination capacity of *Anogeissus latifolia* seed lots has been reported to be very low hampering the natural regeneration and production of seedlings for plantation.

Incubation- Drying- Separation method is widely used as sorting technique to remove non-viable seeds from seed lots. This technique was developed to upgrade the quality of conifer seed lots by removing empty and/or dead-filled seeds [14,15]. It is based on the principle that water imbibed by live seeds is lost at a slow rate than the water imbibed by dead filled and empty seeds when both are subjected to uniform drying conditions. As a result, seeds are separated in two fractions *i.e.* a non-viable surface fraction and a viable bottom fraction due to their densities. The IDS technique has been successfully tested on seed lots of several tree species[1,3,4,11,13,21].

The Present paper deals with the effect of the incubation and wetting- drying and separation of dead and viable fractions of *Anogussius latifolia* seeds. An attempt has been made to find out the suitability of IDS method to remove the non- viable seed from the seed lots to produce the desired quantity of seedling needed for active restoration program and for ensuring a high percentage of viable seeds for nurseries to reduce the cost of containerized seedlings production.

Materials and Methods

Plant Material:

Seeds of *Anogeissus latifolia* were collected from Orchha, district Tikamgarh (M.P.), India, located at 25.35° N lalitute and 78.64° E longitude during the month of April 2018. Phenotypically sound ten trees were marked and seeds were collected during the later period of maturation presuming that such seeds are usually most viable [8,12]. Immediately after collection, seeds were transported to laboratory in polythene bags. All the seeds collected from different trees were mixed and seed lots were prepared [9].

Incubation Drying Separation (IDS) technique:

Separation of seeds was carried out by IDS method[15] taking 4X100 seeds for each treatments. Seeds were imbibed in distilled water for 16 hours and spread between two moistened blotting paper sheets for incubation at 20 ± 2^{0}C and 90% RH for 56 hours. Incubated seeds were first surface dried and then kept for drying for 3, 6, 9, 12 and 15 hours at 20 ± 2^{0}C. Finally dried seeds

TABLE-1 : Changes in the moisture content and proportion of sunken and floating seeds after different periods of drying.(Mean +SE)

Drying time (Hours)	Moisture Content (%)	Sunken Fraction (%)	Floating Fraction%
3	28.57+1.46	45+0.91	55+1.08
6	24.46+1.45	32+1.29	68+1.47
9	15.31+0.72	27+0.41	73+1.47
12	11.54+0.39	22+0.70	78+1.08
15	10.20+0.80	19+0.71	81+1.22

were placed in vessel containing distilled water for separation. After five min, the buoyant seeds on the surface were removed representing the dead seed fraction. Seeds that sunk and settled in the bottom of the vessel represented the viable seed fraction.

Moisture Content:

The moisture content of seed lot was determined on a fresh weight basis by oven drying of two replicates of 100 seeds at 103^0C for a period of 17 hours[9,10]. Seed moisture content was also determined after incubation period as well as after each drying time on wet weight basis.

Assessment of germination and viability :

Untreated seeds were used as control. Sunken and floating seeds (4x 100) from five drying treatments were germinated at 27± 2^0C for 21 days. Germinates were counted everyday when the radicle has reached the size of the seed with a normal appearance. All the non-germinated seeds from both fractions were tested for viability with 2,3,5, triphenyl tetrazotium chloride[9].

Results and Discussion

Results of the present study are based on the fresh seed collected directly from the selected trees during the peak fruiting period. The initial germination and moisture content of *Anugeissus latifolia* was 12.50% and 7.25% respectively. Incubation increased the seed moisture content upto 32.43%. The results indicate that with increasing drying period the moisture content decreased upto 10.20%. The proportion of sunken seeds decreased from 45 to 19 %, while the proportion of floating

TABLE-2 : Percentage germination, TTC viability and dead seeds in sunken and Floating fractions.

Drying Time (Hours)	Sunken fraction			Floating Fraction		
	G	N.G.		G	N-G	
		Viable	Dead		Viable	Dead
3	73.25	6.75	20	03	07	90
6	86.5	05	08.5	01	06	93
9	89.5	02	08.5	01	04	95
12	94.0	02	04	00	02	98
15	100	00	00	00	00	100

* G= Germination: N-G= Non-Germinated

seeds increased from 55 to 81% consistently (Table1).The percentage of viable seeds was maximum after 15 hours of drying than after the other drying periods. Hence, this fraction was considered as most efficient in terms of net gain of the viable seeds after sorting (Table-2). In all drying treatments, viable fraction of seeds showed germination from 73% to 100% against the floating seeds both viable and dead seeds. However, in dead fraction seed germination was found maximum upto 3%.

The study revealed that *Anogeissus latifolia* seeds have poor germination capacity because of emptiness and insect damage. However, seeds particularly consumed by insect larvae usually showed less vigor while severely infested seeds were completely empty of their contents and hence failed to germinate [18-20].

The incubation drying separation technique is largely useful for separation of viable and dead seeds. The physiological property involved may be the deferential ability to absorb and or retain water. The use of differences in the rate of imbibitions of empty, dead and viable seeds for grading of seed quality of several coniferous species[16]. In the present study, the separation of viable and dead seeds showed similar differences.The two fractions, dead and viable seeds are separated owing to their buoyancy which in the former may have been increased due to damage of physical agencies, insect attack[11,14,15,18,21].

Higher drying temperature *i.e.*20^0C in the present study was taken in view of species habitats, is slightly higher[15]. Among different drying periods, fifteen hours drying was found effective in terms of yielding 100% seed germination in sink fraction. Our finding accords with previously reported for *Platanus acerifolia*[7], *Pinus petula*[4], *Schinus molle L*[5] and *Juniperus polycarpus*[3]. The tetrazolium chloride test at the end of germination test revealed 2 to 6.75% non-germinated seeds in the viable fraction and 1 to 2 % in dead fraction in different drying periods were found containing intact and viable embryo, which suggest that they might be dormant with low vigor[5].

The study provides evidence that the low seed germination performance of *Anogeissus latifolia* is due to large quantity of empty, insect damaged and shriveled seeds as observed after cutting test. Such nonviable seeds can be effectively removed by IDS test. Interestingly, this method was found not only suitable for separation of viable seeds from dead seeds, but also rendered a seed lot with higher seed germination as compared to the control from which dead and viable seeds were separated.

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