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Effect of temperature and storage conditions on viability and longevity of recalcitrant (Homoiohydrous) sal (*Shorea robusta* Gaertn. F) seeds

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Abstract

The effect of five storage conditions *viz*., open, closed, partial vacuum, with calcium chloride and silica gel under three temperature regimes of 10°C, 15°C and ambient temperature were observed on freshly collected sal (*Shorea robusta* Gaernt. F.) seeds. The initial seed moisture content and germination was 45.5% and 100% respectively.

Among all the storage conditions, seeds stored at 10°C showed a rapid decline in moisture content upto 10 days. Maximum seed moisture retention was found with storage in closed and partial vacuum conditions at 15°C after 50 days of storage, which also showed maximum viability/germinability (30% and 42% respectively). Statistically significant interaction was observed between storage temperature and storage period. Slow desiccation leads to good storage and longevity of seeds.

Key words: Recalcitrant, Storage physiology, Moisture content, *Shorea robusta*

Introduction

Roberts (1973) classified seeds as orthodox which can be dried of drying to low moisture content with intact viability and for a long period of time at low temperature as orthodox and those which cannot be dried below a relatively high moisture content without subcellular damage and cannot be stored for a long period at low temperature with retention of viability as recalcitrant. Drying of recalcitrant (homoiohydrous) seeds is detrimental to seed viability in nature and storage conditions Berjak *et al*., Berjak and Pammenter, 1997; Varghese *et al*., 2004). Desiccation of recalcitrant seeds to very low moisture content has not been successful, viability of these seeds could be retained by maintaining low moisture content (IBPGR, 1976) under low temperature and humidity (Ellis and Roberts, 1981; Ellis *et al*., 1991). Degree of desiccation varies among recalcitrant species with reported to tolerance and survival of seeds as reviewed by Varghese *et al*. (2004). The seeds of some species tolerate only a slight degree of dehydration as in case of *Shorea robusta* (Chaitanya and Naithani, 1994; 1998).

Shorea robusta Gaertn. F in one of the most important timber tree distributed in humid tropics of Asia. It is a typical dipterocarp species with gregarious fruiting at an interval of 4 years (NG, 1981). Seeds exhibit recalcitrant seed storage behaviour (Nautiyal and Purohit, 1985; Tompsett, 1985; Chaitanya and Naithani, 1994). These seeds lose their viability within an average period of 10 days after full ripening under natural conditions (Troup, 1921; Yadav, 1989; Chaitanya *et al*., 2000). Loss of viability within a short period of time even in storage condition has greatly hampered the species trials and plantation project (Tompsett, 1992).

The present study aims to increase the longevity of *Shorea robusta* seeds under different storage conditions and temperature regimes.

Materials and Methods

Collection of Seeds

Mature seeds of *S. robusta* were collected from tropical moist deciduous forest of Amarkantak, India, situated between 22°-40°N latitude and 81°-46°E longitude. Trees with sound physiognomy were marked and fruits were manually collected. Immediately after collection, fruits were transported in polyethylene bags within 15 hours to the laboratory. Dried wings of the fruits were manually detached and uninfected seeds were separated for the storage.

Seed Moisture Content

Seed moisture content was determined as per the recommended by International Seed Testing Association

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(ISTA, 1985) after drawing two independent samples contained 50 seeds each. Seeds were predried after spreading in a layer at 70°C for 2ï 5 hours in a hot air oven. Dried seeds were ground to small pieces and dried in covered metal containers in oven at constant temperature of 103±2°C for 17 hours and cooled in desiccator. Percent moisture content was calculated on fresh weight basis.

Germination

Surface sterilized seeds (100 ´ 4) were placed on moistened sterilized filter paper sheets in seed germination incubator at 27±2°C. Emergence of radicle was considered as germination and observations were made at an interval of 24 hours.

Seed Storage

Since seeds of *Shorea robusta* are known to possess recalcitrant physiology, seed storage was done within 24 hours of its collection with initial seed moisture content of 45.5%. Seeds were stored in sterilized glass containers under five storage conditions *viz*., open, closed, with silca gel, with calcium chloride and in partial vacuum. For partial vacuum condition, seeds were filled in vacuum desiccators and vacuum was created with rotatory vacuum jump ñGevivekò compressor ¼ Hp at 28" Hg and the desiccators were sealed. All the containers were kept at three temperature regimes *viz*., ambient, 15°C and 10°C. Sample containing 400 seeds were taken out at an interval of 10 days for determination of seed moisture content and germination.

Statistical analysis for interactions among different storage conditions, moisture content and viability was followed after Mather (1966).

Results and Discussion

Results of the present study is based on the freshly collected sal (*Shorea robusta*) seeds with 45.5% moisture content having 100% germination. Seed storage was done by taking two variables into consideration *i.e.* storage temperature and conditions of storage. Seeds lost their viability at ambient temperature after 20 days irrespective of the storage conditions (Table 1). Sal seeds exhibited gradual loss of moisture content under all storage conditions with maximum (20% after 50 days) at room temperature in open storage (Table 2).

Seeds did not survive for long at 10°C temperature however, germination varied under different storage. After 30 days of storage, germination was less than 10%. The seed moisture content did not drop much and remained above 30%. Successful storage of sal seeds was found at 15°C under partial vacuum where viability was 40.25%. Similarly, at 15°C under open and closed storage conditions, seeds showed viability of 11.25% and 30%, respectively. Seed moisture content did not decline much except at ambient temperature and remained above 30% under other storage conditions.

Result of the analysis of variance for temperature, duration and storage conditions and their interaction reveal significant effects of temperature and storage days (Table 3). The interaction of temperature and days was

Table 1: Germination (%) of *Shorea robusta* seeds at different temperatures and storage conditions.

Storage condition	Temperature (°C)	Storage period (days)					
		0	10	20	30	40	50
Open	RT	100	86.25	00.00	00.00	00.00	00.00
container	15	100	97.25	72.25	70.25	68.00	11.25
	10	100	84.25	61.75	32.00	07.00	00.00
Closed	RT	100	30.5	20.25	00.00	00.00	00.00
container	15	100	96.25	67.75	60.00	40.00	30.00
	10	100	92.25	72.25	10.00	08.00	00.00
Partial	RT	100	12.25	00.00	00.00	00.00	00.00
vacuum	15	100	99.25	85.25	65.25	48.25	40.25
	10	100	76.00	48.25	24.00	00.00	00.00
With	RT	100	14.00	00.00	00.00	00.00	00.00
Calcium	15	100	97.25	98.00	31.50	12.00	00.00
chloride	10	100	54.75	26.25	14.00	2.25	00.00
With	RT	100	62.00	20.25	00.00	00.00	00.00
silica gel	15	100	98.50	95.50	50.25	14.00	00.00
	10	100	72.25	60.25	40.25	24.50	00.00

RT = Room Temperature

Table 2 : Variations in moisture content (%) of *Shorea robusta* seeds at different temperatures and storage conditions

Storage condition	Temperature (°C)	Storage period (days)					
		0	10	20	30	40	50
Open container	RT	45.50	35.84	34.37	27.97	26.59	20.00
	15	45.50	38.82	37.69	34.17	30.27	28.12
	10	45.50	34.20	33.77	32.84	32.29	31.27
Closed container	RT	45.50	37.7	35.10	28.92	28.38	26.00
	15	45.50	38.30	37.28	36.00	35.01	34.75
	10	45.50	39.60	37.45	36.66	36.38	36.18
Partial vacuum	RT	45.50	40.12	39.30	38.51	32.10	25.61
	15	45.50	38.71	38.22	36.61	36.06	30.09
	10	45.50	41.61	38.98	37.15	37.27	34.04
With Calcium chloride	RT	45.50	40.21	37.37	34.59	35.55	30.24
	15	45.50	36.25	34.74	32.64	32.34	30.04
	10	45.50	40.79	38.08	37.75	38.00	36.48
With silica gel	RT	45.50	42.64	39.38	38.08	35.06	32.96
	15	45.50	38.33	38.22	36.78	36.57	36.24
	10	45.50	40.00	39.12	37.07	35.00	33.75

RT = Room Temperature

Table 3: Analysis of variance for seed germination in recalcitrant seed (*S. robusta*) stored at different temperature, duration and storage conditions.

	Item	Sum of squares	Degree of freedom	Mean square	Variance ratio
Main effects	Storage days (D)	42821.57	4	10705.39	49.01***
	Temperature (T)	28894.56	2	14447.28	66.14***
	Storage condition (C)	2178.91	44	544.72	2.49^{NS}
First order	D ´ T	5712.18	8	714.02	3.26**
Interaction	D ´ C	2248.22	16	140.51	0.64^{NS}
	T ´ C	1790.64	8	223.83	1.024^{NS}
Second order	D ´ T ´ C	6989.89	32	218.43	é .
	Total	90635.97	74		

(Significance: ** - 0.01, *** - 0.001, NS ï not significant)

also found significant ($p<0.05$). However, there was no significant effect of these parameters with storage conditions.

Mature *S. robusta* seeds with 100% germination showed rapid loss of viability during storage at ambient temperature. Complete loss of viability was exhibited at room temperature after 10 days of storage. Seeds retained viability upto 50 days of storage under partial vacuum conditions at 15°C temperature. More than 50% loss in seed viability is exhibited in seeds stored at 10°C beyond 20 days of storage period. Surprisingly, seed moisture content did not decline much except in open storage.

Varghese *et al*. (2004) also reported the viability loss of *S. robusta* seeds (dried and undried) at freezing and above freezing temperatures. They also found maximum seed viability at 15°C storage temperature. Seeds of tropical recalcitrant species have been found to lose viability completely above freezing temperature *e.g. Dipterocarpus* at < 14°C, *Cacao* at < 10°C and *Mangifera indica* at < 3 ï 6°C (King and Roberts, 1979).

Storage of undried seeds of *S. robusta* showed an increase in longevity of seeds at higher moisture content as already reported by Varghese *et al.* (2004). However, chilling damage at lower temperature (below 15°C) was found in seeds with higher moisture content by Khare *et al.* (1987) and Yadav *et al.* (1987). Seed storage trials in the present study showed higher viability and longevity for those seeds which were subjected to rapid drying and low initial vigour. Slow dried seeds have sufficient time to undergo relatively substantial germination changes before the hydration level become limiting factor (Berjak *et al.*, 1990). Khare *et al.* (1987) found that the critical moisture content of *S. robusta* seeds as 28.2%, below which seeds do not survive.

The storage behaviour of *S. robusta* seeds in the present experiment is characteristic of typical tropical recalcitrant seeds. Similar behaviour in loss of viability on desiccation and storage has been reported in other tropical tree species namely, *Aesculus indica* (Uniyal and Nautiyal, 1996), *Mangifera indica* (Corninean and Come, 1989), *Madhuca indica* (Varghese *et al.*, 2002).

It is evident from the results that seeds with unaltered moisture content and slow desiccation can be stored for comparatively longer period of time than those stored after rapid desiccation.

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