# STUDIES ON MAINTAINING SEED LONGEVITY AND THE EFFECT OF FRUIT GRADES IN NEEM (AZADIRACHTA INDICA)

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Received November 1990

PONNUSWAMY, A.S., VINAYA RAI, R.S., SURENDRAN, C. & KARIVARA-THARAJU, T.V. 1991. Studies on maintaining seed longevity and the effect of fruit grades in neem (*Azadirachta indica*). Grading of neem (*Azadirachta indica*) depulped fruits by the floatation method gave 18% floaters. These weighed comparatively less than the sinkers, possessed a higher endocarp content and recorded low viability in terms of maximum germinability and also vigour. The sinkers in contrast recorded an increase of 35% in viability and 92% in vigour index. In a separate study, seeds stored at an ambient temperature of 33.8°C recorded a germinability of 8% at the end of three months compared to 90% when freshly collected. This is attributed to desiccation injury as the moisture content dropped from an initial 30.8 to a low 15.5% at the end of three months. In contrast, seeds stored in earthern pots buried in moist sand bed manifested little loss in seed moisture content and recorded a germinability of 62% at the end of three months. Density grading for maximising viability and vigour and moist storage in earthern pots for prolonging longevity is recommended for the species.

Key words: Neem - fruit density grading - viability and vigour - fruit storage

#### Introduction

The terms 'orthodox' and 'recalcitrant' were introduced to describe the storage behaviour of seeds (Roberts 1973). Orthodox seeds are shed from the parent plant at low moisture content, having undergone maturation drying and are capable of tolerating dehydration down to 5 to 6%. When dry, the viability of these seeds can be prolonged by keeping them at the lowest temperatures and moisture contents possible (Roberts 1972, IBPGR 1976). In contrast, recalcitrant seeds do not undergo maturation drying and are shed at relatively high moisture contents. Seeds of most tropical and subtropical trees and shrubs are recalcitrant (King & Roberts 1979, Come 1982, Hanson 1984, Roberts *et al.* 1984). These seeds are not storable under conditions suitable for orthodox seeds and can only be stored in wet medium to obviate dehydration injury and at relatively warm conditions as these are mostly sensitive to chilling injury (Lyons *et al.* 1979, Chin & Roberts 1980, Graham & Paterson 1982). To date there has been no successful method for long term storage of recalcitrant seeds (Farrant *et al.* 1988). Whether neem (*Azadirachta indica*) is a genuine recalcitrant

or short lived orthodox species, however, is still nebulous (Willan 1985). On the basis of low moisture content of seeds (12.5%) from a Haiti plantation it has been argued that neem is not a recalcitrant species (Chaney & Knudson 1988). Again since neem occurs in dry tropical forests while most recalcitrant tropical species are found in moist tropical forests, it was suggested that neem may have short lived orthodox seed (Willan 1985). However, recognising several facts like advocacy by many for sowing the seeds within two or three weeks after collection (FAO 1956, 1957, NAS 1980, Kamweti 1982), occurrence of high moisture content (37.2%) in fresh seeds (Maithani et al. 1989) and manifestation of chilling damage by the seeds (Emah 1986, Maithani et al. 1989), the species is indicated to be a recalcitrant one. Nevertheless, the polemics remain still unresolved. The present study was carried out to assess the effect of moist storage on seed longevity on the assumption that neem seeds are recalcitrant. Furthermore, as information on the effect of seed grades on seed viability and vigour in the species is scant, an experiment was also made to investigate this influence.

#### Materials and methods

#### Effect of grading of drupes on seed germination

Ripe fruits (15 samples) collected from a 20-y-old tree were depulped, washed thoroughly, air dried for 8 h and resolved into sinkers and floaters by the liquid floatation technique (Willan 1985). Each sample comprised a replication. The two components were then air dried for four days and weighed. Each component was resolved into two lots. In one lot the endocarp and kernel fractions were manually separated and weighed. From the other lot, 25 each of sinkers and floaters (one replication) were germinated on sterilized quartz sand filled in enamelled trays (Maithani et al. 1989). Owing to non-availability of adequate seeds the size of replication could not be kept larger. Twenty-one days after sowing (Chaney & Knudson 1986), the number germinated was counted and the percentage germination computed. After count, dry weight of ten random seedlings was obtained by oven drying at  $105^{\circ}C$  for 72 h. Vigour index is the integral of percentage germination and seedling dry weight (Abdul Baki & Anderson 1973). The data were subjected to an analysis of variance and treatment differences tested (t-test) for significance ( $P \le 0.05$ ) after Panse and Sukhatme (1967).

#### Effect of wet storage on seed viability

Sinkers separated by the floatation method and air dried after treatment with Dithane M-45 were transferred to 12 earthern pots (@ 50 seeds per pot) and the pots buried up to neck level in 20 to 25% moist sand bed under a thatched shed. The mouth of the pots was kept open. The sand moisture content was

maintained by replenishing water once every three days. A similar number of seed lots stored in the ambient served as control. The treatments were set up in quadruplicate in a complete randomised block design. At monthly intervals for three months (one, two and three months after storage), four pots (replications) each were drawn from the two storage media and 25 seeds from each pot tested for germination. Ten seeds were assayed for moisture content following Willan (1985). Initial moisture content and germinability were determined prior to storage. While the mean temperature during the storage period was  $33.8^{\circ}C$  in the ambient, it was  $25.6^{\circ}C$  inside the pots buried in moist stand. Data were subjected to statistical scrutiny as described earlier.

#### **Results and discussion**

### Effect of grading of drupes on seed viability

In the total depulped seedlot, floaters accounted for 18% (Table 1). These were characterised not only by lesser weight but also by higher proportion of endocarp. This may be attributed to ill-filling of kernels. In addition, their germinability (55%) and vigour index (3850) were also low. The sinkers in contrast, recorded higher values for not only viability measured as maximum germinability but also vigour measured in terms of seedling dry weight and vigour index. The magnitude of increase over the floaters was 35.0, 17.1 and 91.6% respectively. A close parallel between seed/fruit size/weight and seed viability and vigour has been documented in many hardwoods like teak (*Tectona grandis*) (Eidmann 1934), *Leucaena leucocephala* (Pathak *et al.* 1974, Gupta *et al.* 1983, Natarajan & Vinaya Rai 1984) and kapok tree (*Ceiba pentandra*) (Gawande 1985), which emphasises the need for fruit or seed grading. But grading by weight or size alone was reported to be ineffective in eliminating

Fruit density grade	Percentage to total depulped	100 fruit weight depulped	Proportion depulped f %		Germina- tion (%)	Dry weight (mg seedling <sup>-1</sup> )	Vigour index
0	fruit lot		Kernel	Endocarp			
Sinker	82.0	24.8	64.0	36.0	90.0	82	7380
	(64.9)		(53.1)	(36.8)	(71.5)		
Floater	18.0	21.5	57.0	42.0	55.0	70	3850
	(25.1)		(49.4)	(47.8)			
SEd	0.35	0.10	0.46	0.33	0.68	0.7	1.7
CD (5%)	0.75	0.21	1.00	0.72	1.47	1.5	3.7

Table 1. Effect of fruit density on seed viability and vigour in neem

(Figures in parentheses are arc-sin transformations)

inferior seeds from a seedlot and density-grading based on fullness of seed was emphasised (Ferguson & Turner 1971). It is a good indicator of seed maturity (Bartee & Krieg 1974) as well as seed quality (Krieg & Bartee 1975).

Density grading in preference to size or weight-grading has also been advocated by Tupper *et al.* (1971). Enhanced germination obtained with density graded fruits in the present study is consistent with similar associations reported earlier in *Casuarina equisetifolia* (Kaja Maideen *et al.* 1989). It is therefore advocated that depulped drupes in neem be density graded and only the sinkers used for seedling production.

#### Effect of wet storage on seed viability and moisture

The initial moisture content of the air-dried seeds was 30.8% and the initial germinability was 90.0% (Table 2). In the seeds stored in the ambient, there was a progressive decline in moisture content over time. The reduction during the first, second and third months of storage was 8.2, 5.2 and 1.9% respectively, the cumulative loss being 15.3% or half as much as the initial moisture content. Concomitant with this fall in moisture, viability of seeds measured as maximum germinability also progressively diminished to a low of 8% at the end of three months, the magnitude of reduction compared to the initial being as high as 82.0%. In contrast, the seeds stored in earthern pots recorded a germinability of 62% at the end of three months, the loss compared to the original being only 28.0%. The moisture content of these seeds also showed no appreciable loss over time and was maintained at an almost constant 30%.

	Seed moisture content (%)				Seed germination (%)			
Months after storage	0	1	2	3	0	1	2	3
Earthern pot	30.8	30.5	30.4	30.1	90.0	86.0	78.0	62.0
•	(33.7)	(33.5)	(33.4)	(33.2)	(71.5)	(68.3)	(61.7)	(51.1)
Ambient	30.8	22.6	17.4	15.5	90.0	74.0	26.0	8.0
	(33.7)	(28.3)	(24.6)	(23.1)	(71.5)	(58.6)	(30.4)	(16.3)
	SEd		CD (P	$\leq 0.05)$	SEd	_	CD (P	$\le 0.05)$
Medium	0.05		0.10		0.52		1.11	
Period	0.07		0.15		0.73		1.58	
M * P	0.10		0.21		1.04		2.23	

Table 2. Effect of wet storage on seed viability and vigour in neem

(Figures in parentheses are arc-sin transformations)

The poor storability of seeds in the ambient may be attributed to their rapid dehydration. Seeds of four species rich in moisture (*Mangifera indica, Shorea roxburghii, Hopea odorata* and *Symphonia globulifera*) lost moisture during dry storage ( $20^{\circ}C$ , 55% RH) and the seeds lost their viability as they dried, though sensitivity to dehydration varied with the species (Corbineau & Come 1988). All seeds were dead when their moisture content fell to about 17% for *S. roxburghii* and *H. odorata*, 30% for *M. indica* and 37% for *S. globulifera*. Recalcitrant seeds have been suggested to lack the mechanism allowing drying to low moisture contents (Berjak *et al.* 1984). Also, neem seeds contain a high

percentage of both saturated and unsaturated fatty acids and free fatty acids within seeds could be responsible for loss of viability (Chaney & Knudson 1988). Long chain unsaturated fatty acids cause swelling of isolated mitochondria and impairment of their normal function (Bewley & Black 1982). Absence of appreciable seed senescence in seeds in earthern pots may be ascribed to maintenance of moisture content and possibly also to non-formation of free fatty acids. This inexpensive method which stores well the seeds for three months is recommended for the species.

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