

SEED STORAGE STUDIES IN *SYZIGIUM CUMINII*

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Key words: Viability – storability – recalcitrant – desiccation – chilling sensitive – storage container

ANANDALAKSHMI, R., SIVAKUMAR, V., WARRIER, R. R., PARIMALAM, R., VIJAYACHANDRAN, S. N. & SINGH, B. G. 2005. Kajian penyimpanan biji benih *Syzigium cuminii*. Kajian peyimpanan untuk melanjutkan kebolehhidupan biji benih *Syzigium cuminii* dijalankan. Kandungan lembapan awal biji benih berdasarkan berat segar ialah 44.2% dan percambahan awalnya pula ialah 89%. Kami ingin menentukan kandungan lembapan penyimpanan, suhu dan bekas yang paling baik untuk biji benih. Biji benih dikeringkan sehingga mencapai kandungan lembapan sebanyak 35%, 30%, 25%, 20% dan 10% dan disimpan pada suhu bilik (28–30 °C), 20 °C, 15 °C dan 10 °C dalam bekas plastik. Kapasiti percambahan biji benih diuji selepas disimpan selama 30, 75, 120, 165, 180 dan 270 hari. Biji benih didapati tahan walaupun dikeringkan sehingga kandungan lembapan 10%. Suhu penyimpanan yang paling sesuai ber julat antara 15 °C hingga 20 °C. Menyimpan biji benih yang mempunyai kandungan lembapan 10% pada 20 OC dapat melanjutkan kebolehhidupan sehingga 180 hari dengan 92.5% percambahan. Biji benih yang tidak dikeringkan disimpan dalam pelbagai bekas iaitu polibeg (tolok 150, 200 dan 250), polibeg dengan dua liang (tolok 200), beg kain, beg kertas dan bekas plastik dan semuanya disimpan pada 20 °C. Selepas lima bulan, kami dapati polibeg, tidak kira ketebalannya dan sama ada berliang atau tidak, paling baik untuk melanjutkan tempoh kebolehhidupan.

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Introduction

Syzigium cuminii, commonly known as jamun, is a member of the Myrtaceae family. It is one of the most widely distributed tropical fruit trees in India and is found both in wild and cultivated conditions. The fruit is a favourite among the locals and the seed is used as a traditional preparation for treating diabetes (Prince *et al.* 1998).

Syzigium cuminii seeds are recalcitrant. Recalcitrant seeds are relatively high in moisture content and possess a characteristic feature of losing their viability during desiccation (Liang & Sun 2000). Despite the fact that cooling and drying form the basis for preserving all types of seeds (Ruhl 1996), the recalcitrant seeds impose serious storage problems due to their desiccation and chilling sensitivity (Chin & Roberts 1980). These seeds undergo no maturation drying during the final phase of seed development and are thus shed in moist condition. Desiccation below a critical level leads to loss of viability. Recalcitrant seeds are intolerant to drying and low temperatures. Plants that produce recalcitrant seeds are typically restricted to aquatic environments or humid tropical areas, where the environment is suitable for seedling growth throughout the year (Roberts & King 1980).

The main fruiting season of jamun in India extends through late May to July. The tree produces seeds every year. Seeds continue to be viable for three to five months after pre-sprouting. Prolonging the viability of seeds until next fruiting season would facilitate the availability of seeds for various plantation programmes and also for use by local farmers throughout the year. Hence, the present study was aimed at finding suitable storage moisture content, temperature and container for storage of *S. cuminii* seeds.

Materials and methods

Fully ripen fruits of *S. cuminii* were collected from the forest during two successive years and used for the study. The first and second year collections were tagged as seedlot 1 and seedlot 2 respectively. The seeds were extracted after the fruits were soaked in water for an hour. Seeds were then depulped manually and washed. The cleaned seeds were surface dried under shade (28–30 °C) for half an hour.

All the experiments were carried out with four replications of 25 seeds each. The germination trials were conducted in completely randomized design and tested for statistical significance using analysis of variance (ANOVA) at 5% level of significance after arc sine transformation of the values (Panse & Sukhatme 1995) followed by Duncan's multiple range test for multiple comparison.

The processed seeds were tested for moisture content on fresh weight basis by oven dry method (ISTA 1999). Moisture content was studied in four replications of five seeds each. Germination study was conducted in sand medium after the seeds were soaked in water overnight. Due to the polyembryonic nature of *S. cuminii*, a seed giving rise to multiple seedlings was counted as a single entity in the germination test (ISTA 1993). The final count of germination test was taken after 40 days of sowing.

Effects of temperature

Fresh non-desiccated seeds (moisture content 42.5%) were stored in plastic containers at different temperatures, namely, ambient (28–30 °C), 20 °C, 10 °C and 0 to -5 °C. The stored seeds were subjected to germination test to study the effects of temperature after 20, 40, 60, 75, 100 and 170 days of storage.

Effects of storage temperature and seed moisture content

The interactive effects of storage temperature and seed moisture content with varying storage durations were studied. Freshly collected seeds (moisture content 44.2%) were desiccated to different moisture contents. While taking samples for estimation of initial moisture content, five other samples were taken at random. The fresh weights of these samples were determined. As soon as the moisture content of the seedlot was known, the five samples were each dried to different target moisture contents, i.e. 35, 30, 25, 20 and 10%. The target weights for drying seeds to the required moisture content were determined using the formula given by DFSC (1999). The seeds were dried to the different moisture levels under shade (28–30 °C). After the seeds had attained the targeted moisture contents, their actual moisture contents were determined. These values turned out to be 35.8, 31.1, 24.1, 19.6 and 11.4% respectively. The seeds were then stored in plastic containers at ambient (28–30 °C), 20, 15 and 10 °C. The stored seeds were tested for germination after 30, 75, 120, 165, 180 and 270 days.

Effects of storage containers on storability

Fresh non-desiccated seeds (moisture content 44.2%) were stored in different storage containers, namely, as sealed polybags of varying gauges (150, 200 and 250), 200-gauge polybag with ventilation (polybags were with two 5 mm diameter pores), cloth bag, paper bag and air-tight plastic container. The size of polybags, cloth bags and paper bags was 15 × 20 cm. The plastic containers used for this study were of 1 l capacity. Seeds were filled to 50% capacity of these containers and stored at 20 °C for five months and tested for germination.

Results and discussion

The initial moisture contents of seedlots 1 and 2 were found to be 42.5 and 44.2% with 92 and 89% germination respectively. Seedling emergence was noticed 10 days after sowing. Generally moisture contents of recalcitrant seeds are reported to be 30 to 70% (Chin *et al.* 1984).

Effects of temperature

The viability of seeds stored at ambient was only 20 days (Table 1). *Syzigium cumini* seeds did not withstand chilling below 10 °C. At 0 to -5 °C, seeds completely lost their viability after 20 days of storage. Chilling temperatures are known to adversely affect the viability of seeds of tropical origin (Chin *et al.* 1989, Bedi & Basra 1993). Storage of seeds between 10 to 20 °C was found to prolong the viability of the seeds. After 170 days of storage at 10 and 20 °C, the germination percentage was 65 and 69% respectively (Table 1). Though seeds stored at 10 and 20 °C showed good germination, maximum germination was observed in seeds stored at 20 °C even after 75 days of storage. The seeds in these storage temperatures reached peak germination after a short interval of storage and later declined with increasing storage period. This could be ascribed to the phenomenon of after-ripening. Non-desiccated seeds of *S. cumini* had been successfully stored at 16 °C for four (DFSC 1998) and 19 weeks (DFSC 1999). Mittal *et al.* (1998) reported that *S. cumini* seeds with 43.6% moisture content could be successfully stored up to 20 weeks in untied polybags at 16 and 5 °C without serious impact on their germination.

Effects of storage temperature and seed moisture content

Seeds in this study with actual moisture contents 35.8, 31.1, 24.1, 19.6 and 11.4% showed 47.17, 49.24, 52.00, 51.61 and 40.98% germination respectively (Table 2). This showed that germination percentage was not greatly affected by desiccation. This suggested that seeds could even be desiccated up to 20% moisture content with minimum loss of seed viability.

Table 1 Effects of storage temperature on germination of non-desiccated seeds stored for up to 170 days

Temperature	Germination (%)					
	20 days	40 days	60 days	75 days	100 days	170 days
Ambient (28–30 °C)	24 <i>29.28^d</i>	0 <i>0^e</i>	0 <i>0^e</i>	0 <i>0^e</i>	0 <i>0^e</i>	0 <i>0^e</i>
20 °C	89 <i>70.96^{ab}</i>	87 <i>68.85^b</i>	94 <i>77.94^a</i>	91 <i>72.61^{ab}</i>	82 <i>65.20^b</i>	69 <i>56.54^c</i>
10 °C	88 <i>70.06^{ab}</i>	89 <i>70.96^{ab}</i>	87 <i>69.25^b</i>	82 <i>65.57^b</i>	83 <i>65.60^b</i>	65 <i>54.06^c</i>
0 to -5 °C	16 <i>22.24^d</i>	0 <i>0^e</i>	0 <i>0^e</i>	0 <i>0^e</i>	0 <i>0^e</i>	0 <i>0^e</i>

Values in italics denote angular transformation values. Means with the same letter do not differ significantly as per DMRT at 5% level of significance.

Table 2 Effects of temperature, moisture content (MC) and storage period on the germination percentage of *Syzigium cumini* seeds

Storage temperature	Seed MC (%)	Germination (%)						Temperature × MC
		30 days	75 days	120 days	165 days	180 days	270 days	
Ambient (28–30 °C) (5.7%)	35.8	0 ³	0 ³	0 ³	0 ³	0 ³	0 ³	0
	31.1	0 ³	0 ³	0 ³	0 ³	0 ³	0 ³	0
	24.1	90 ^{a-i}	0 ³	0 ³	0 ³	0 ³	0 ³	15
	19.6	79.75 ^{d-p}	0 ³	0 ³	0 ³	0 ³	0 ³	13.3
	11.4	0 ³	0 ³	0 ³	0 ³	0 ³	0 ³	0
20 (73.3%)	35.8	85.25 ^{c-m}	97.5 ^a	88.25 ^{b-k}	86.75 ^{b-l}	67.5 ^{b-w}	45 ^{v-1}	78.4
	31.1	71.75 ^{g-t}	87.5 ^{a-h}	88.25 ^{e-r}	76.75 ^{b-k}	57.5 ^{h-z}	45 ^{v-1}	71.1
	24.1	77.5 ^{b-l}	65 ^{d-r}	83.5 ^{e-y}	83.25 ^{b-l}	60 ^{m-x}	47 ^{u-1}	69.4
	19.6	71.75 ^{g-t}	72.5 ^{f-s}	60 ^{e-y}	96.5 ^{ab}	85 ^{b-k}	44 ^{w-1}	71.6
	11.4	81.75 ^{c-o}	77.5 ^{d-r}	81.75 ^{c-o}	74.75 ^{e-s}	92.5 ^{a-c}	48 ^{t-1}	76
15 (72.6%)	35.8	89.75 ^{a-e}	75 ^{d-p}	86.75 ^{a-j}	83.25 ^{c-n}	75 ^{e-s}	33 ^l	73.8
	31.1	88.25 ^{b-k}	97.5 ^a	90 ^{a-i}	88.5 ^{a-g}	87.5 ^{a-g}	35 ^{v-1}	81.1
	24.1	90.25 ^{a-f}	75 ^{e-s}	78.5 ^{d-r}	86.75 ^{b-l}	65 ^{h-u}	39 ^{y-1}	72.4
	19.6	86.5 ^{b-l}	72.5 ^{g-s}	78.5 ^{d-r}	55 ^{r-1}	72.5 ^{g-s}	38 ^{v-1}	67.2
	11.4	61.75 ^{n-y}	85 ^{b-k}	80 ^{d-q}	76.5 ^{d-r}	67.5 ^{h-w}	39 ^{y-1}	68.3
10 (41.3%)	35.8	56.5 ^{q-1}	85 ^{c-m}	70 ^{i-u}	0 ³	7.5 ²³	0 ³	36.5
	31.1	68.5 ^{j-v}	90 ^{a-d}	80 ^{d-q}	15 ²³	15 ²	0 ³	44.8
	24.1	96.5 ^{ab}	90 ^{a-i}	78.25 ^{d-r}	35 ^{v-1}	7.5 ²³	0 ³	51.2
	19.6	68.5 ^{j-v}	80 ^{e-p}	80.25 ^{e-p}	55 ^{r-1}	42.5 ^{x-1}	0 ³	54.4
	11.4	56.5 ^{q-1}	52.5 ^{s-1}	8.5 ²	0 ³	0 ³	0 ³	19.6

Means with the same superscript do not differ significantly as per DMRT at 5 % level of significance. Values in parentheses are overall treatment means for germination (%).

The germination percentage of the seeds stored with different moisture contents varied with days of storage (Table 2). The seeds tested after frequent intervals showed peak in germination after a certain period of time. The time taken to reach the peak germination increased with decrease in moisture content. This phenomenon is clear in seeds stored at 20 °C. At this temperature, seeds stored with 35.8, 31.1, 24.1, 19.6 and 11.4% moisture contents showed peaks in germination after 75, 120, 120, 165 and 180 days of storage with 97.5, 88.25, 83.5, 96.5 and 92.5% germination respectively. Hence, we recommend that seeds of *S. cumini* be stored with 10% moisture since peak germination was delayed at this parameter. Storage of seeds with 20% moisture or below has the advantage of protection from

fungi. Attacks by pathogens has been reported as a serious cause for loss of viability in seeds with high moisture contents like *S. cuminii* (Mittal *et al.* 1998). In this study, seeds stored at moisture contents above 10% showed higher pre-sprouting during storage.

Seeds stored at 15 to 20 °C showed improved viability. Seeds stored at different temperatures, namely, ambient, 20 °C, 15 °C and 10 °C showed 5.66, 73.30, 72.56 and 41.28 % germination (Table 2). The study also proved that storage temperature played a major role in maintaining the viability of the seeds compared with seed moisture content. This is because different levels of desiccation did not affect the germination percentage greatly compared with chilling. Thus, to store seeds up to 165 days with germination as high as 96.5%, we recommend the combination of 20% seed moisture and 20 °C. For still prolonged storage up to 180 days it is best to store seeds with 10% moisture content at 20 °C.

Effects of containers

Seed storage in suitable containers would prevent the direct contact of seeds with storage environment and protect seeds from pests and diseases. Hence, for effective storage, appropriate storage container is essential (Purohit & Doijode 1998). Our results suggested that polybags, irrespective of thickness and presence or absence of pores, are the best for storing *S. cuminii* seeds (Table 3). Doijode (1995) reported that fresh seeds of recalcitrant species should be packed in semimoisture-resistant containers and stored at medium temperature (10–15 °C). Polythene bags with a wall thickness from 0.1 to 0.250 mm is thick enough to prevent excessive moisture loss and thin enough to allow some gas exchange for short-term storage of recalcitrant seeds (Bonner 1996).

Seeds stored in cloth and paper bags did not germinate. These containers did not prevent seeds from desiccation. The moisture contents of the seeds stored in cloth and paper bags were 15.8 and 14.9% respectively while moisture contents of seeds stored in polybags were all above 45%. The pores present in the walls of

Table 3 Effects of containers on storability of seeds at 20 °C

Type of container	MC% of stored seeds before germination	Germination % (after 5 months of storage)
Polybag 200 gauge (2 pores)	46.45	83.3 (66.47) ^a
Polybag 200 gauge	48	88.3 (70.50) ^a
Polybag 150 gauge	47.165	83.3 (66.47) ^a
Polybag 250 gauge	50.475	88.3 (70.50) ^a
Cloth bag	15.825	0.00 (0.00) ^c
Paper bag	14.925	0.00 (0.00) ^c
Plastic container	49.525	68.3 (56.28) ^b

Values in italics denote angular transformation values. Means with the same letters do not differ significantly as per DMRT at 5% level of significance.

cloth and paper bags allowed free movement of air and moisture leading to poor hygienic condition inside the container. This could be the reason for poor germination of seeds stored in these containers.

Conclusions

Recalcitrant seeds do not complete maturation drying. The seeds are shed at relatively high moisture contents and are sensitive to desiccation. In this study, although the initial moisture content of *S. cumini* seeds was 44.2% on fresh weight basis with an initial germination of 89%, desiccating the seeds to 20% moisture did not have a drastic effect on germination. Further, these seeds attained peak germination after a longer period relative to other desiccation conditions. This proved that reduction in moisture contents facilitates longer storability and better germination.

Temperature was the major factor which played a significant role in controlling seed deterioration. As in the case of moisture content, peak germination was extended for more than six months in seeds stored at 20 °C. Ventilation of recalcitrant seeds is essential to maintain viability. Hence, storage containers also play a major role in controlling seed deterioration. The need to replenish oxygen and to remove toxic gases produced during metabolism must be taken into account when deciding storage conditions. However, we need to ensure that seeds do not go beyond the lowest safe moisture content.

To conclude, the fresh and mature seeds of *S. cumini* could store well in polybags when desiccated to 10% moisture content and kept at temperatures ranging between 10 and 20 °C. These conditions would help the seeds retain their viability and vigour for over nine months, making the seeds available for the next planting season.

References

- BEDI, S. & BASRA, A. S. 1993. Chilling injury in germinating seeds: basic mechanisms and agricultural implications. *Seed Science Research* 3: 219–229.
- BONNER, F. T. 1996. Commercial seed supply of recalcitrant and intermediate seed: present solutions to the storage problem. Pp. 27–33 in Ouedraogo, A. S., Poulsen, K. & Stubsgaard, F. (Eds.) *Intermediate/Recalcitrant Tropical Forest Tree Seeds: Proceedings of a Workshop on Improved Methods for Handling and Storage of Intermediate and Recalcitrant Tropical Forest Tree Seeds*. 8–10 June 1995. Humlebaek. IPGRI, Rome and DANIDA Forest seed Centre, Humleback.
- CHIN, H. F. & ROBERTS, E. H. 1980. *Recalcitrant Crop Seeds*. Tropical Press Sdn. Bhd., Kuala Lumpur, Malaysia.
- CHIN, H. F., HOR, Y. L. & MOHAMMED LASSIM, M. B. 1984. Identification of recalcitrant seeds. *Seed Science and Technology* 12: 429–436.
- CHIN, H. F., KRISHNAPILLAY, B. & STANWOOD, P. C. 1989. Seed moisture: recalcitrant vs. orthodox seeds. Pp. 15–22 in Stanwood P. C. & Mc Donald, M. B. (Eds.) *Seed Moisture*. CSSA Special Publication No. 14. Crop Science Society of America, Madison.
- DFSC. 1998. *Syzigium cumini*. The project on handling and storage of recalcitrant and intermediate tropical forest tree seeds. *DANIDA Forest Seed Centre Newsletter* No. 4(April): 6–7.
- DFSC. 1999. Desiccation and storage protocol. The project on handling and storage of recalcitrant and intermediate tropical forest tree seeds. *DANIDA Forest Seed Centre Newsletter* No. 5(April): 12–13.

- DOIJODE, S. D. 1995. Short-term conservation of mango seeds. *Plant Genetic Resources Newsletter* 104: 24–25.
- ISTA. 1993. International rules for seed testing (Supplement Rules). *Seed Science and Technology* 21: 157.
- ISTA. 1999. International rules for seed testing. *Seed Science and Technology* 27: 30–35.
- LIANG, Y. & SUN, W. Q. 2000. Desiccation tolerance of recalcitrant *Theobroma cacao* embryonic axes: the optimal drying rate and its physiological basis. *Journal of Experimental Botany* 51: 1911–1919.
- MITTAL, R. K., HANSEN, H. J. & THOMSEN, K. 1998. Effect of seed treatments and storage temperature on storability of *Syzygium cuminii* seeds. *IUFRO Seed Symposium Recalcitrant Seeds*. Forest Research Institute Malaysia, Kepong.
- PANSE, V. G. & SUKHATME, P. V. 1995. *Statistical Methods for Agricultural Workers*. Indian Council of Agricultural Research, New Delhi.
- PRINCE, P. S. M., MENON, V. P. & PARI, L. 1998. Hypoglycaemic activity of *Syzygium cuminii* seeds: effect on lipid peroxidation in alloxan diabetic rats. *Journal of Ethnopharmacology* 61: 1–7.
- PUROHIT, A. G. & DOIJODE, S. D. 1998. PGR activities at IIHR: germplasm introduction, exchange and conservation with emphasis on fruit crops. Pp. 159 in Arora, R. K. & Ramanatha Rao, V. (Eds.) *Tropical Fruits in Asia: Diversity, Maintenance, Conservation and Use: Proceedings of the IPGRI-ICAR-UTFANET Regional Training Course on the Conservation and Use of Germplasm of Tropical Fruits in Asia*. IIHR. 18–31 May 1997, Bangalore.
- ROBERTS, E. H. & KING, M. W. 1980. Storage of recalcitrant seeds. Pp. 39–48 in Withers, L. A. & Williams, J. T. (Eds.) *Crop Genetic Resources: The Conservation of Difficult Materials*. International Union of Biological Sciences Series B42.
- RUHL, G. 1996. Microscopic methods for subcellular examination of desiccation and chilling sensitivity. Pp. 44–73 in Ouedraogo, A. S., Poulsen, K. & Stubsgaard, F. (Eds.) *Intermediate/Recalcitrant Tropical Forest Tree Seed: Proceedings of a Workshop on Improved Methods for Handling and Storage of Intermediate and Recalcitrant Tropical Forest Tree Seeds*. 8–10 June 1995, Humlebaek.