

DESICCATION STUDIES ON MAHOGANY (*SWIETENIA MACROPHYLLA*) SEEDS

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MARZALINA, M. & NORMAH, M. N. 2001. Desiccation studies on mahogany (*Swietenia macrophylla*) seeds. Studies of desiccation effect on mahogany seeds were conducted to determine the best condition to reduce seed moisture content without affecting its viability. Four types of desiccation methods, namely, ambient temperature of 28 °C (in incubator), cold room of 21 °C (in laboratory), circulated air oven at 35 °C and silica gel (in desiccator at 25 °C) were tested on seeds with and without testa. All four desiccation treatments maintained high germination between 70–78% for seeds with testa. Ambient and cold conditioned seeds without testa resulted in 55–70% germination. Overall results showed that a gradual reduction of moisture content to the lowest safe level while maintaining high viability can be achieved using ambient and cold conditioned desiccation techniques. Oven and silica gel desiccation methods increased respiration rate and cellular leachate conductivity, especially for seeds without testa. Light microscopy showed that cells of seeds without testa desiccated using oven and silica gel had damaged cell walls and cytoplasm. These injuries did not occur in cells of seeds with testa. It was concluded that mahogany seeds with testa can be stored for a short-term period if the moisture content is reduced gradually to 2–9% using environmental condition of 21–28 °C.

Key words: Desiccation - embryo axes - seeds - germination - moisture content

MARZALINA, M. & NORMAH, M. N. 2001. Kajian pengeringan terhadap biji benih mahogany (*Swietenia macrophylla*). Kajian kesan pengeringan ke atas biji benih mahogani dijalankan untuk menentukan keadaan yang terbaik bagi mengurangkan kandungan air biji benih tanpa memberi kesan kepada kebolehidupan biji benih. Empat kaedah pengeringan, iaitu pada suhu bilik 28 °C (dalam inkubator), suhu bilik sejuk 21 °C (dalam makmal), oven berkitaran udara 35 °C dan gel silika (dalam desikator 25 °C) telah diuji ke atas biji benih berkulit dan tanpa kulit. Keempat-empat rawatan dapat mengekalkan percambahan yang tinggi antara 70–78% untuk biji berkulit. Suhu bilik dan bilik sejuk mengekalkan 55–70% percambahan biji benih tanpa kulit. Secara keseluruhan, dengan teknik pengeringan pada suhu bilik dan bilik sejuk, keputusan menunjukkan penurunan kandungan air ke tahap selamat tanpa menjejaskan kebolehidupan biji benih. Kaedah penggunaan oven dan gel silika telah meningkatkan kadar respirasi dan konduktiviti luluhlarutan dalam sel

terutamanya ke atas biji benih tanpa kulit. Pemerhatian melalui mikroskop cahaya membuktikan bahawa dinding dan sitoplasma sel biji benih tanpa kulit musnah apabila dikeringkan secara oven dan gel silika. Keadaan ini tidak berlaku ke atas sel-sel biji benih berkulit. Boleh disimpulkan bahawa biji benih mahogany yang masih berkulit dapat disimpan untuk tempoh masa yang singkat jika kandungan air dikurangkan secara perlahan-lahan ke tahap 2–9% pada suhu persekitaran antara 21–28 °C.

Introduction

Mahogany (*Swietenia macrophylla*) is regarded as the world's finest timber for high-class furniture and cabinet work due to its light hardwood quality. Originated from Central America, this exotic species is cultivated in Malaysia for reforestation and as shade trees for urban landscapes. This species grows rapidly, reaching 20 m height and 60 cm diameter in 25 years when planted in deep, fertile and well-drained soils (Appanah & Weinland 1993). Annual volume increment is between 15–20 m³ per hectare which is achieved within 40 to 50 years. To maintain continuous supply of the planting materials for this species, seeds were collected from several experimental plots within forest plantation areas in Peninsular Malaysia. Although the newly collected seeds germinate easily, they are prone to attacks by fungus if their moisture content is not reduced sufficiently.

Only a few studies on mahogany seeds have been reported and our understanding of the desiccation effects on the seeds is limited. Mahogany seeds are unable to tolerate drying to a low moisture content (Lopez 1938). Several suggestions on condition and treatment have been proposed to maintain high viability of mahogany seeds during storage (Perera 1973, Vivekanandan 1978, Martini 1985) but results are inconsistent. Effects of desiccation method and period are very critical to achieve the lowest safe moisture content especially if the mahogany fruits are harvested straight from mother trees and not collected from the ground (Pukittayacamee 1991). The drying period is important if the seeds are immature and have a moisture content between 45–52%. Martini (1985) found that seeds extracted from freshly forced open fruits fail to germinate. However, when the fruits are dried for three days, moisture of the seeds content reduces to 9% and viability increases to 85%. In addition, Martini (1985) observed that germination declines to 54% after the fourth day of drying. Marzalina (1995) and Pukittayacamee *et al.* (1995) found that mahogany seeds behave semi-recalcitrantly (intermediate) during storage. The present experiment was undertaken to study the effects of desiccation methods on mahogany seeds and to determine the best condition to reduce moisture content without affecting the viability of seeds during a short storage period of 30 days. We hoped that the study can further explain the cellular activities of the seeds during the desiccation exposure.

Materials and methods

Freshly harvested fruits of mahogany (*S. macrophylla*) were collected from Lentang Forest Plantation, Pahang, Malaysia. The seeds were extracted from the fruits and were then subjected to four desiccation treatments with their testa intact and removed.

- (1) **Ambient temperature**
Seeds were placed into a wire mesh-based container in an incubator (Hotpack, Model 352601, Philadelphia, USA) at 28 ± 2 °C. The relative humidity (RH) was $65 \pm 2\%$.
- (2) **Cold room**
Seeds were placed into a wire mesh-based container in an air-conditioned room at 21 ± 2 °C. The RH was $55 \pm 2\%$.
- (3) **Circulated air oven**
Seeds were placed into a wire mesh-based container in a circulated air oven (Memmert, Model BM 400, Germany) at 35 ± 2 °C. The RH was $50 \pm 2\%$.
- (4) **Silica gel**
Seeds were placed in a desiccator at 25 ± 2 °C. The RH was $35 \pm 2\%$.

Each treatment had three replications. For treatments 1, 2 and 3, the seeds in each wire mesh-based container were mixed from time to time to distribute the effect of temperature evenly during desiccation. For treatment 4, the silica gel, which was twice the weight of the tested seeds, was replaced every other day to ensure continuous drying effect.

To determine the viability of seeds in each treatment, three replicates of 50 seeds each were sown in sand on every alternate day up to the 30th day of storage. In addition, moisture content on fresh weight basis of three replicates of 10 seeds per replicate was determined at the same interval by oven drying at 103 °C for 16 h. Regression analysis was carried out on these indexes.

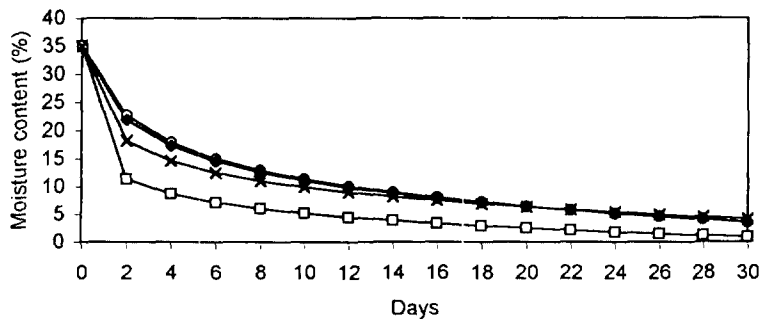
Respiration and leachate conductivity indexes were determined at 0, 5, 10, 15, 20, 25 and 30 days to evaluate cellular activities or damage during drying. Respiration was measured by oxygen uptake of 10 embryonic axes dissected aseptically using a respirometer (Gilson Medical Electronics Inc., Wisconsin, USA). The embryonic axes were placed in conical flasks containing 2.0 ml sterile incubation medium (10 ml streptomycin sulfate (500 ml l⁻¹), 10 ml penicillin (500 ml l⁻¹), 2.51 g Na₂HPO₄ · 12 H₂O, 0.41 g KH₂PO₄ and distilled water to make up the volume to 100 ml). CO₂ was absorbed in 0.2 ml of freshly prepared 20% potassium hydroxide on fluted blotter placed in the middle of the conical flask. The flasks were shaken continuously at a slow speed using an orbital shaker (Hotech, model 720) at 30 °C and the readings were taken two hours after equilibration.

For leachate conductivity, 10 seeds per replicate were washed six times in deionised water before being immersed in 50 ml deionised water for one hour at room temperature (22 °C). The leachate conductivity was measured using a conductivity meter (Jenway Ltd., model 4010).

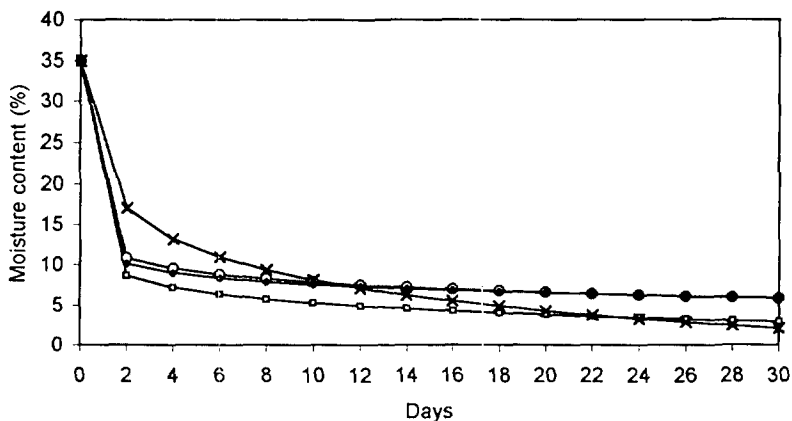
Changes in cells of embryonic axes were observed by light microscopy. Ten embryonic axes were fixed in 50% formalin-aceto-alcohol (FAA) for a week at 24 °C. After fixation, the samples were dehydrated in 70% EtOH followed by increasing concentration of tertiary butyl alcohol (TBA). Once dehydration was completed, the samples were gradually infiltrated in different grades of wax. The block containing embryonic axes was cut at 12 micron thick. The mounted slides were stained in a combination of safranin-fast green as given in Johansen (1940).

Results

Reduction of moisture content of the seeds over 30 days is shown in Figure 1. During the first two days, the reduction of moisture content was rapid for all desiccation treatments for both seeds with and without testa. Desiccation using silica gel showed a more uniform reduction rate down to 2% especially for seeds with testa. Circulated air oven reduced moisture content for seeds with testa much faster from 35 to 9% within the first two days. Desiccation at ambient temperature and in the cold room caused seed moisture content to decline at a slower rate during the 30 days period. These treatments gave a high germination percentage of 55–78% (Figure 2) compared to desiccation methods using silica gel. However, at the end of the 30th day storage all four desiccation treatments attained high germination of 70–78% for seeds with testa, with moisture contents ranging between 2–9%. All germinated seeds showed normal hypogeal growth.



(a) Testa intact

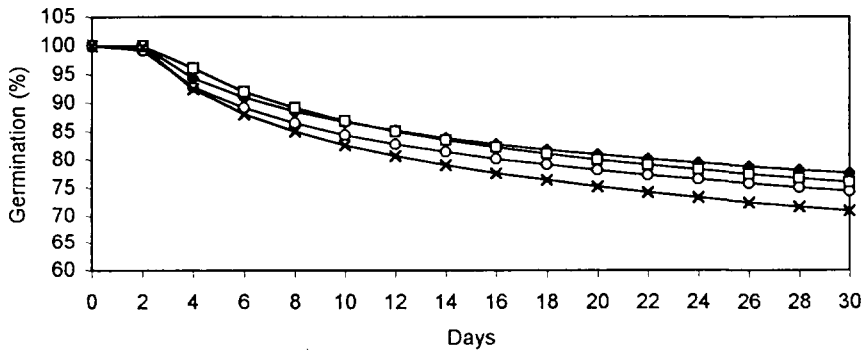


(b) Testa removed.

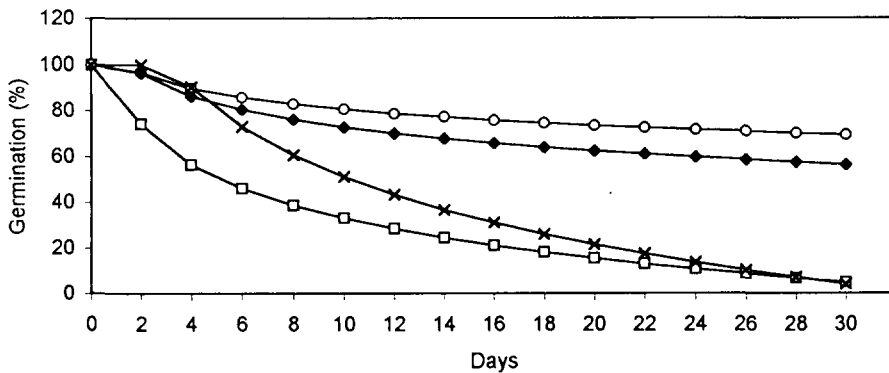
Desiccation conditions on seeds for 30 days
 —○— Ambient —●— Cold room —□— Oven —×— Silica gel

Figure 1 Comparative graph of regression analysis for percentage of moisture content for seeds (a) with testa intact (b) testa removed

Desiccation of seeds without testa using oven drying caused almost total loss of moisture content (Figure 1 (b)) resulting in seeds being largely not viable at the end of this treatment (Figure 2(b)). Germination of seeds without testa placed in silica gel also declined rapidly with moisture reduction reaching 3% at the end of the treatment. However, when kept in the cold room and at ambient temperature, the final moisture content was maintained at 5% and this only had slight effect on the viability of seeds (55-70% germination).



(a) Testa intact

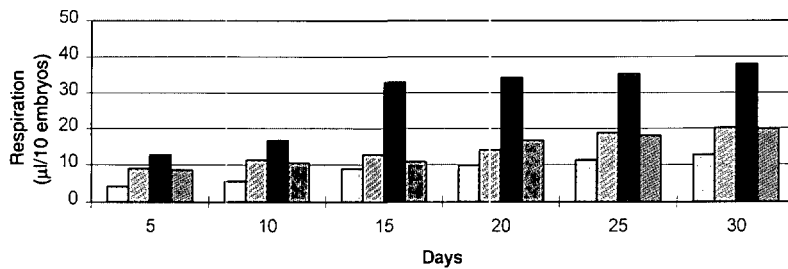


(b) Testa removed

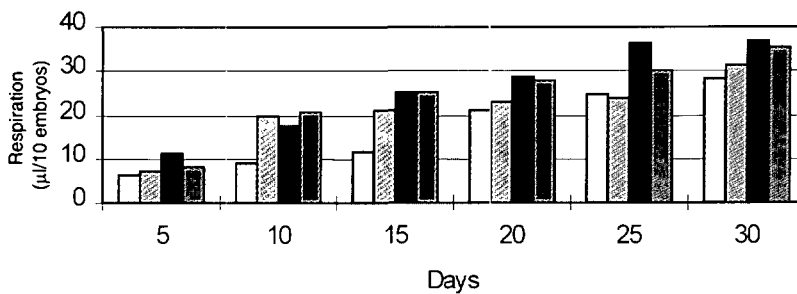
Desiccation conditions on seeds for 30 days
 ○ Ambient ● Cold room □ Oven × Silica gel

Figure 2 Comparative graph of regression analysis for percentage of germination for seeds with (a) testa intact (b) testa removed

All treatments caused respiration rate (oxygen uptake) to increase with time (Figure 3). Circulated air oven drying of both seeds with and without testa had the highest rate of oxygen uptake among all treatments, followed by silica gel, cold and ambient temperature methods respectively. Respiration rate was higher in seeds without testa and correlated to lower viability.



(a) Testa intact



(b) Testa removed

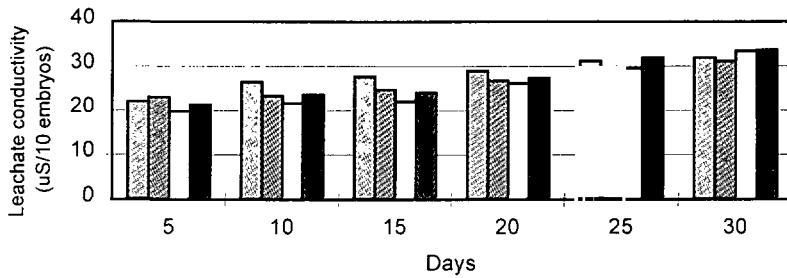
Desiccation conditions on seeds on 30 days
 LSD ($p = 0.05$) between two points = 0.18

Ambient
 Cold room
 Oven
 Silica gel

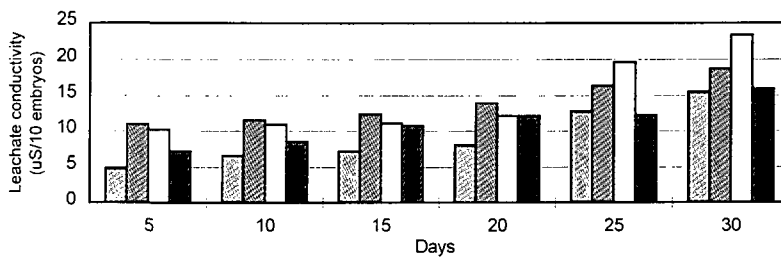
Figure 3 Respiration ($\mu\text{l}/10$ embryos) for 2 hours for mahogany seeds with (a) testa intact and (b) testa removed under various desiccation treatments for 30 days

Leachate conductivity of seeds with testa was higher compared to seeds without testa (Figure 4). Circulated oven drying method produced highest leachate conductivity in seeds without testa.

Figure 5 shows the normal cell condition without treatment, served as control. Figures 6–13 show cells under various desiccation treatments. Ambient conditions caused cytoplasm of the seeds with testa to shrink slightly during the 30 days of storage (Figure 6) compared to seeds without testa (Figure 7). Not much difference was observed in the cells for seeds with testa exposed to cold room treatment (Figure 8) compared to the control cells (Figure 5). However, slight reduction of cell walls and shrinkage of cytoplasm were observed in seeds without testa (Figure 9). The same results were also observed in seeds with testa under circulated air oven (Figure 10) and silica gel treatments (Figure 12). However, cells from seeds without testa under such conditions were found damaged (Figures 11 & 13).



(a) Testa intact



(b) Testa removed

Desiccation conditions on seeds for 30 days
 LSD ($p = 0.05$) between two points = 0.28
 Ambient Cold room Oven Silica gel

Figure 4 Leachate conductivity ($\mu\text{S}/10$ embryos) for mahogany seeds (a) with testa intact and (b) testa removed under various desiccation treatments for 30 days

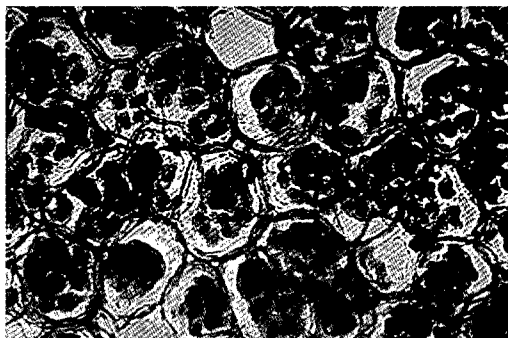


Figure 5 Cells structure of mahogany seed with testa serve as control

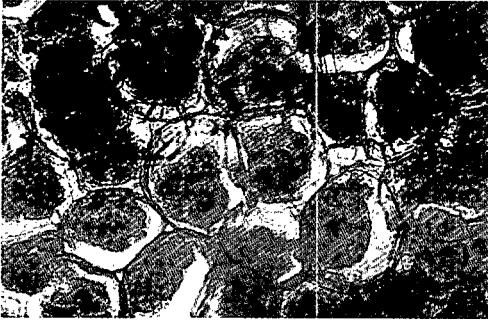


Figure 6 Cells structure of seeds with testa after 30 days ambient treatment (28 °C)

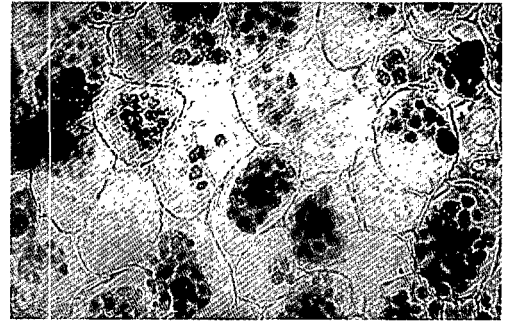


Figure 7 Cells structure of seeds without testa after 30 days ambient treatment (28 °C)

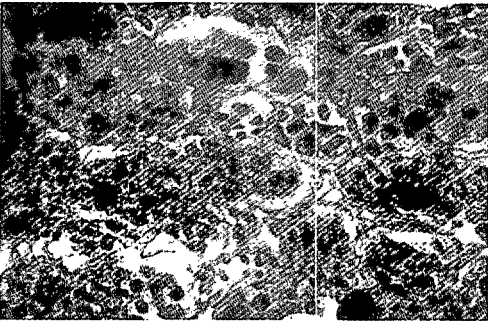


Figure 8 Cells structure of seeds with testa after 30 days cold room treatment (21 °C)

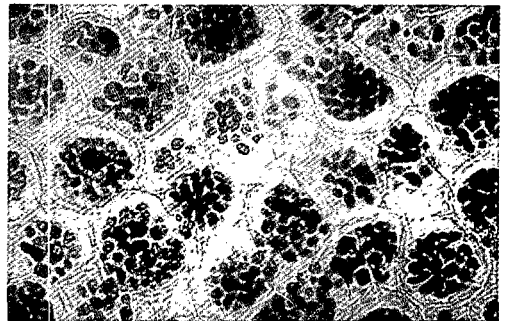


Figure 9 Cells structure of seeds without testa after 30 days cold room treatment (21 °C)

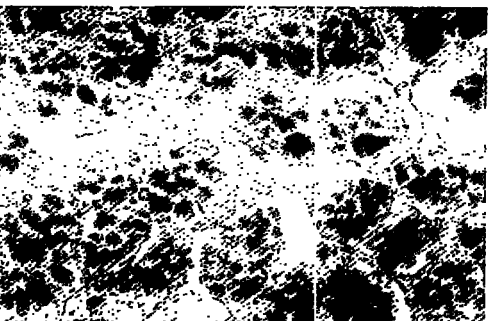


Figure 10 Cells structure of seeds with testa after 30 days oven treatment (35 °C)

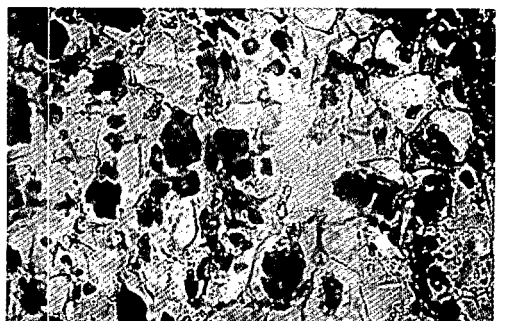


Figure 11 Cells structure of seeds without testa after 30 days oven treatment (35 °C)

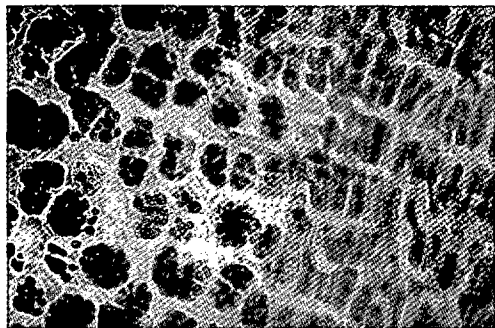


Figure 12 Cells structure of seeds with testa after 30 days silica gel treatment (25 °C)

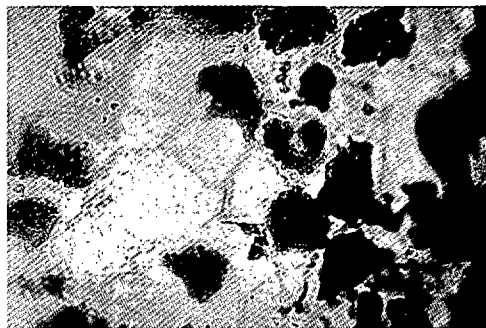


Figure 13 Cells structure of seeds without testa after 30 days silica gel treatment (25 °C)

Discussion and conclusion

From the experiment conducted, the lowest safe moisture content was concluded to be between 2–9%, which was lower than 12–14% proposed by Schmidt (2000) but within the range of 7–12% reported by Hong and Ellis (1996). Desiccation using a temperature of 35 °C (oven) could not maintain the viability of seeds. However, treatment within the range of 21–28 °C maintained high viability of both seeds with and without testa and this concurred with the study carried out by Corbineau & Come (1988) on recalcitrant species like *Shorea roxburghii*, *Hopea odorata*, *Mangifera indica* and *Symphonia globulifera* as well as the desiccation study by Maury-Lechon *et al.* (1989) on *Shorea parvifolia* and *Dipterocarpus humeratus*.

The circulated air oven and silica gel desiccation methods increased the respiration rate and leachate conductivity of seeds without testa. Light microscopy study showed that cells in seeds without testa suffered shrunk or ruptured cell walls due to the oven and silica gel desiccation treatments. Germination studies showed that seeds with testa were able to revive after 30 days ambient and cold room desiccation treatments, indicating that the shrinking of cytoplasm was temporary and seeds recovered after regaining water. Seeds that reduce moisture content rapidly can tolerate loss of water (Farrant *et al.* 1988). Recalcitrant seeds cannot withstand such condition due to its morphology and to the lack of intolerable desiccation component within its structured membrane (Bochicchio *et al.* 1994). The high leachate conductivity observed in seeds with testa suggested that there were chemical components that fused out from the damaged cells but these were not determined chemically. It was reported, however, that mahogany seeds contain terpenoids, swietenin and swietenolide chemical compounds (Chan *et al.* 1976).

From this study, we concluded that respiration rate and leachate conductivity could assist in determining the viability of seed without testa. High respiration rate and leachate conductivity indicated low viability. It could also be concluded that mahogany seeds with testa could be stored for short-term period if the moisture content was reduced gradually to 2–9% at a temperature of 21–28 °C. This definitely

favour the practical approach of storing large quantities of mahogany seeds with testa intact in a cold room.

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