Research paper

Germination and Storage Behavior of Seeds of *Scolopia oldhamii* Hance (Flacourtiaceae)

Jeng-Chuann Yang,[1,3] Shing-Rong Kuo,[2] Chiung-Mei Lee[1]

【Summary】

The purpose of this study was to examine the germination characteristics of seeds of *Scolopia oldhamii* Hance. Effects of the seed moisture content (MC) and storage temperature on germination were investigated to determine its seed storage behavior. *Scolopia oldhamii* seeds completely germinated within 4 wk at fluctuating temperatures of 30/20℃ with 8 h of light, and this result showed that *S. oldhamii* seeds were non-dormant. In addition, germination mainly took place during 7~14 d. Stratification at 4℃ was not efficient in improving germination of *S. oldhamii* seeds, but only decreased the mean germination time (MGT) to about 8 d. In fact, such a reduction in the MGT is not practical in the nursery. Moreover, seed viability sharply deteriorated with 4℃ moist storage for more than 6 mo. Thus, low-temperature moist storage is not suitable for temporarily storing *S. oldhamii* seeds. *Scolopia oldhamii* seeds which could be dried to 2.0~7.6% MCs (on a fresh-weight basis) still maintained viability after storage at 4 and -20℃ for 24 mo. This result showed that *S. oldhamii* seeds are orthodox; that is, long-term storage can be applied to this species for future seed demands. We suggest that *S. oldhamii* seeds be dehydrated to 3~7% MCs and then hermetically sealed for storage at -20℃ when a long-term *ex situ* conservation strategy is adopted.

Key words: *Scolopia oldhamii*, germination, seed storage behavior, orthodox, stratification.

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研究報告

魯花樹種子的發芽與儲藏行為

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摘　要

本研究探討魯花樹種子的發芽特性，並以種子含水率與儲藏溫度對發芽率之影響來判定其種子儲藏行為。結果顯示魯花樹種子不具休眠性，以30/20℃變溫經4週後就能完全發芽完畢，發芽主要集中在7~14日。以4℃層積無法有效提升魯花樹種子的發芽率，僅稍能降低平均發芽日數至約8日，但此功效在育苗作業上不具實用價值。而且當4℃低溫濕藏時間超過6 mo後會讓其種子活力迅速下降，故低溫濕藏並非短暫儲藏魯花樹種子之可行方法。魯花樹種子乾燥至含水率2.0~7.6% (鮮重)，經24個月的4及-20℃儲藏後種子均仍能維持活力，故判定其屬正儲型，亦即可將種子進行長期儲藏以供日後利用。建議未來以儲存種子進行本種長期的區外保育時，應先將種子含水率降低到3~7%後密封儲藏在-20℃環境中。

關鍵詞：魯花樹、發芽、種子儲藏行為、正儲型、層積。

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INTRODUCTION

In recent years considerable concern has arisen over the effects of the seed moisture content (MC) and storage temperature in seed longevity and seed storage behavior research. Seed storage behavior was initially divided into orthodox and recalcitrant types. The longevity of orthodox seeds is prolonged with concurrent decreases in the MC and storage temperature; the relationships can be inferred from mathematical approaches (Roberts 1973). However, Hong and Ellis (1996) divided seed storage behavior into 3 categories: orthodox, recalcitrant, and intermediate according to the degree of desiccation tolerance of mature seeds. Orthodox seeds can survive an MC of < 5% (on a fresh-weight basis; all MC values in this study are on a fresh-weight basis). Therefore, orthodox seeds have a high degree of desiccation tolerance.

Different from orthodox seeds, recalcitrant seeds have a low degree of desiccation tolerance. When recalcitrant seeds are dried to < 12~31% MCs, seed viability decreases with concurrent prolongation of the drying duration (Roberts 1973). Many recalcitrant seeds are extremely sensitive to low temperatures, and at temperatures of < 10~15℃, their viability often deteriorates, especially seeds from tropical areas (Bonner 1990). Hong and Ellis (1996), therefore, differentiated recalcitrant seeds into temperate and tropical subtypes according to the level of temperature tolerance of recalcitrant seeds. Temperate-recalcitrant seeds cannot survive desiccation but can survive a temperature close to 0℃ and MCs of 35~50%. Air circulation plays an important role in the respiration of temperate-recalcitrant seeds during storage; i.e., seed longevity is more likely to reach 1~3 yr after storage at high MCs with appropriate air circulation (Wang et al. 1995). Different from temperate-recalcitrant seeds, tropical-recalcitrant seeds have a weak tolerance of desiccation and low temperatures. They are almost unable to survive a temperature close to 0℃ and are even sensitive to 15~20℃ (Hong and Ellis 1996). As tropical-recalcitrant seeds are very prone to losing viability at low temperatures, the period for which they can be stored is short, even if they are stored at high moisture contents with good air circulation. In other words, viable seeds begin to quickly germinate after storage at a high temperature. To prolong storage duration, we employed a low-temperature approach to inhibit germination. However, the optimal storage conditions are difficult to determine, and seeds often lose their viability. Thus, it is not easy to preserve tropical-recalcitrant seeds for long-term periods. Moreover, relative to the longevity of temperate-recalcitrant seeds, that of tropical-recalcitrant seeds is shorter. Take the Dipterocarpaceae for example. Among 79 species of these tropical-recalcitrant seeds, the maximum seed longevity is 1 yr, and the minimum is only 14 d (Tompsett 1987, 1989).

In addition to orthodox and recalcitrant seeds, Ellis et al. (1990) found that *Coffea arabica* L. seeds survived desiccation to 5~10% MCs but failed to survive at a storage temperature of < 10℃. Such seeds are considered intermediate; that is, intermediate seeds are not like orthodox seeds by showing a strong desiccation tolerance and are vulnerable to low-temperature damage. According to Hong and Ellis (1996), most intermediate seeds can survive desiccation to 10~12% MCs, but their viability declines with a concurrent reduction in the MC.

Scolopia oldhamii Hance, a small evergreen tree species of the Flacourtiaceae, is distributed in southern China, the Philippines, and coastal and lowland areas of Taiwan (Li and Lo 1993). Its globose berries are about 6~8 mm in diameter, and each fruit usually produces 4 or 5 seeds. The fruits are dark purple when mature, and the maturation period is in January and February (Fig. 1).

No previous research on *S. oldhamii* seeds has been published, so 2 seedlots in this study were used to investigate the germination and storage behaviors of *S. oldhamii* seeds to have a careful research design and produce accurate results. The purpose is to offer useful information about nursery operations for afforestation programs of native coastal forest species in the future.

MATERIALS AND METHODS

Seed collection and processing

Detailed collection information on the 2 seedlots of this study is given in Table 1. These 2 seedlots were collected in 2003 and 2006, respectively. At the time of collection, 60% of the fleshy fruits of seedlot 1 were

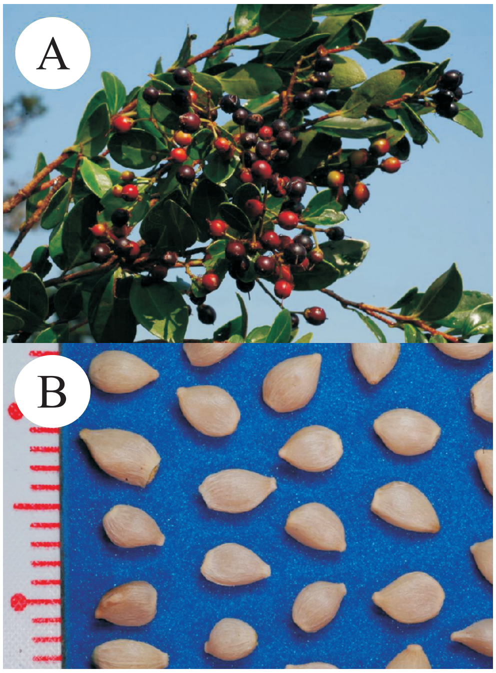

Fig. 1. Mature fruits (A) and seeds (B) of *Scolopia oldhamii*.

Table 1. Information on fruit collection dates and sources of the 2 seedlots of *Scolopia oldhamii*

Seedlot	Collection date	Collection location	Latitude, longitude	Elevation (m)
Seedlot 1	13 Jan. 2003	Chiayi	23°29'N, 120°27'E	35
Seedlot 2	25 Jan. 2006	Hengchuen	21°58'N, 120°48'E	180

light red, and the mature fruits of seedlot 2 were all dark purple. The collected fruits of these 2 seedlots were placed in a greenhouse and kept moist to cause the fruit flesh to ripen and decompose. Once the berries had become softer, they were immediately washed and extracted to obtain cleaned seeds. Then, flotation and manual selection were used to remove empty and damaged seeds from the pure seeds with no debris left. Seed characteristics of fresh mature seeds of the 2 seedlots are given in Table 2. The number of seeds of these 2 seedlots per liter was about 90,000~99,000 (Table 2). The thousand seed weight of these 2 seedlots was about 6.6~7.0 g (Table 2). Tests of germination and storage behavior on selected fresh mature seeds of these 2 seedlots were immediately conducted.

Determination of MC

Seed MCs were determined gravimetrically with 4 replicates. For each replicate, seedlot 1 of 20 seeds and seedlot 2 of 25 seeds at 0.12~0.15 g each were randomly selected and cut into pieces of < 4 mm in length. Then, the picked seeds were dehydrated. The MCs of the seeds were determined using a low-constant-temperature oven method (103±2℃ for 17±1 h) (International Seed Testing Association 1999). All MCs are presented on a percentage fresh-weight basis.

Germination assay

To avoid imbibition damage by rapid rehydration, seeds with different treatments were placed above water in a sealed container for 1 d before the germination test, so that these seeds could take up water at ambient temperatures. The imbibed seeds were then thoroughly mixed with clean sphagnum moss in sealable polyethylene (PE) bags (14×10 cm, and 0.04 mm thick) with adequate air inside. The excess water of the moss was squeezed out until a water content of about 450% by mass (on a dry-weight basis) was reached (Lin and Chen 1993). To estimate the germination capability of the fresh mature seeds and seeds for dry storage, 35 seeds from seedlot 1 and 50 from seedlot 2 were randomly sampled in 4 replicates per treatment for the germination test at fluctuating temperatures of 30/20℃ (day/night) with 8

Table 2. Information on seed characteristics and germination of fresh mature seeds of the 2 seedlots of *Scolopia oldhamii*

	Seedlot 1	Seedlot 2
Moisture content (%, on a fresh-weight basis)	12.2±0.1	10.1±0.1
Number of seeds/L	98762±902	90825±766
Thousand seed weight[1)] (g)	6.92±0.09	6.61±0.06
Germination of fresh mature seeds (%)	55.0±8.2	99.0±1.0
Mean germination time of fresh mature seeds (d)	14.3±0.4	14.1±0.1

[1)] The thousand seed weight was estimated at the moisture content shown.

h of light (50~80 μmol m^{-2} s^{-1}). During the 8-wk test, the number of protruding seeds was counted once per week. Meanwhile, 3 ml of water was added to the moss. Seeds with a radicle reaching 5 mm were counted as having germinated.

Moist storage treatment

Seeds of seedlots 1 and 2 were stored for 9 (0, 1, 2, 3, 6, 9, 12, 18, and 24 mo) and 13 periods (0, 1, 2, 3, 4, 5, 6, 8, 10, 12, 15, 18, and 24 mo) for up to 24 mo, respectively. In addition, 35 seeds from seedlot 1 and 50 seeds from seedlot 2 were randomly sampled in 4 replicates for each moist storage period, and then these seeds were incubated at fluctuating temperatures of 30/20℃ (day/night) with 8 h of light (50~80 μmol m^{-2} s^{-1}) for the 8-wk germination test. Freshly selected seeds of the 2 seedlots were thoroughly mixed with wet sphagnum moss in a PE bag (14×10 cm, and 0.04 mm thick) and then placed at 4℃ for storage, which was the same procedure used for stratification. The extra water of the moss was removed until a water content of about 390% by mass (on a dry-weight basis) was achieved.

Dry storage and determination of the moisture level of dehydrated seeds

Sub-seedlots exhibited different moisture levels, and the range was between approximately 2% and the initial MC of fresh mature seeds. The initial MCs of fresh mature seeds of the 2 seedlots are shown in Table 2. When a sub-seedlot reached the desired moisture level, the seeds were placed in an aluminum foil bag and stored at 4℃ for about 5 d before the average equilibrium MC was determined. As soon as the desired MC of each sub-seedlot was achieved, the seeds were sealed in double-layered foil bags to stabilize the MCs. In seedlot 1, a factorial combination of 7 different storage durations (0, 3, 6, 9, 12, 18, and 24 mo) and 3 different temperatures (-20, 4, and 15℃) was used, while in seedlot 2, that of 8 different storage durations (0, 3, 6, 9, 12, 15, 18, and 24 mo) and 3 different temperatures (-20, 4, and 15℃) was employed.

Mature seeds of the 2 seedlots were placed in a hermetically sealed acrylic box with a small fan installed to circulate the air, and molecular sieves and pure water were used to dry and moisten the seeds, respectively. In seedlot 1, the MC of fresh mature seeds was 12.2%. After seed treatment, the 5 moisture levels were controlled to 2.0±0.2% (48 h of desiccation), 5.3±0.1% (4.5 h of desiccation), 7.6±0.2% (1 h of desiccation), 10.8±0.2% (0.7 h of desiccation), and 14.5 ±0.4% (7.5 h of humidification). In seedlot 2, the MC of the fresh mature seeds was 10.1%. After treatment, the 5 moisture levels were 2.3 ±0.3% (42 h of desiccation), 4.9±0.1% (4 h of desiccation), 7.2±0.2% (0.9 h of desiccation), 11.3±0.1% (3 h of humidification), and 14.3±0.3% (40 h of humidification).

Data analysis

The equation for calculating the mean germination time (MGT) is: $MGT = \Sigma(n \times d)/N$, where n is the number of germinated seeds after each period of incubation in days (d), and N is the total number of emerged seeds recorded by the end of the test (Hartmann et al. 1989).

Analysis of variance (ANOVA) was used to analyze the seed germination percentage and MGT to evaluate the effects of the stratification period on germination. Additionally, germination results of the combination of different storage temperatures, seed MCs, and storage periods were statistically analyzed by variables implemented in Tukey's test of the PROC GLM procedure of the SAS statistical package (SAS, Cary, NC, USA).

RESULTS

Germination of fresh mature seeds

The following observations were made of the germination of fresh mature seeds of the 2 seedlots. In seedlot 1, when fresh seeds were incubated at the fluctuating temperatures of 30/20℃ for 8 wk, their germination percentage and MGT were respectively 55.0% and 14.3 d (Table 2). Seeds mainly emerged during the 2nd wk, and there was no further germination after the 4th wk. In seedlot 2, the germination percentage and MGT of fresh seeds were respectively 99.0% and 14.1 d under the same germination conditions as seedlot 1 (Table 2). The seeds mainly emerged during the 2nd wk, and germination was completed within 3 wk. From the above results, *S. oldhamii* seeds exhibited no dormancy and displayed a high germination speed.

Effects of moist storage at 4℃ on germination

Figure 2 shows the effects of moist storage at 4℃ on germination of the 2 seedlots. In seedlot 1, seed germination percentages were 43.6~50.0% after moist storage at 4℃ for 6 mo. There was no significant difference in germination ($p > 0.05$) compared to fresh seeds (55.0%). However, the germination percentage decreased to 25.7% after 9 mo of moist storage, and the seeds had completely lost viability after 12 mo (Fig. 2). Moreover, after moist storage for 1~6 mo, the MGTs of seeds were 7.9~8.3 d. Clearly, a significant decrease in MGTs ($p < 0.05$) was found compared to fresh seeds (14.3 d). However, the MGT of seeds after 9 mo of moist storage had returned to 13.3 d, and this result showed no significant difference compared to fresh seeds ($p > 0.05$) (Fig. 2). In seedlot 2, after moist storage at 4℃ for 6 mo, seed germination percentages were 80.5~91.5%, which suggested that the seeds maintained good viability. However, a significant decrease in germination ($p < 0.05$) was found compared to fresh seeds (99.0%). In addition, the germination percentage sharply decreased to 66.0% after moist storage for 8 mo. The seeds had almost lost all viability after 4℃ moist storage for 12 mo (4%) and had all died after 15 mo (Fig. 2). Moreover, after moist storage for 1~6 mo, the MGTs of the seeds were 7.5~9.2 d. A significant decrease in MGTs ($p < 0.05$) was found compared to fresh seeds (14.1 d). However, the MGTs of the seeds after moist storage for 8 and 10 mo had returned to 12.5 and 16.5 d, respectively, which showed no significant difference compared to fresh seeds ($p > 0.05$) (Fig. 2).

Effects of MC and storage temperature on seed longevity

Figure 3 shows the effects of different MC levels and storage temperatures on the germination percentage of *S. oldhamii* seeds of seedlot 1 after dry storage for 0~24 mo. Germination percentages of these seeds at MCs of 14.5, 10.8, 7.6, 5.3, 2.0% were 56.4, 57.1, 56.4, 60.7, and 54.3%, respectively. There was no significant difference ($p > 0.05$) among the germination percentages of these 5 moisture levels of dried seeds and the fresh sample (55.0%). This result reveals that seeds of *S. oldhamii* can tolerate dessication. Of the above seeds with 5 moisture levels, the seeds at 10.8 and 14.5% MCs had totally lost viability after 9 and 6 mo of storage at -20℃, respectively (Fig. 3). The germination percentages of the other 3 lower-MC seeds at MCs of 7.6, 5.3, and 2.0% were 50.0, 55.0, and 60.7% after 24-mo storage, respectively (Fig. 3). There was no significant difference in germination compared to fresh seeds ($p > 0.05$). After storage at 4℃ for 24 mo, no significant decrease in germination was revealed among seeds at the 4 lower moisture levels compared to fresh seeds ($p > 0.05$). However, a significant difference in germination ($p < 0.0001$) was found in seeds at a 14.5% MC (7.1%) after 18 mo of storage, and the seeds had totally lost their viability after 24 mo. After 24 mo of storage at 15℃, only 2 sub-seedlots of seeds at 5.3 and 2.0% MCs remained viable. Germination of seeds at a 7.6% MC decreased to 4.3% after 24-mo storage, while seeds at 10.8 and 14.5% MCs had quickly lost viability and had all died after storage for 18 and 9 mo, respectively.

Figure 4 shows the effects of different MC levels and storage temperatures on the germination of *S. oldhamii* seeds of seedlot 2 after dry storage for 0~24 mo. When the drying process was achieved, freshly dried seeds with 5 moisture levels were incubated at the fluctuating temperatures of 30/20℃ for 8 wk. Of these seeds, germination percentages at MCs of 14.3, 11.3, 7.2, 4.9, and

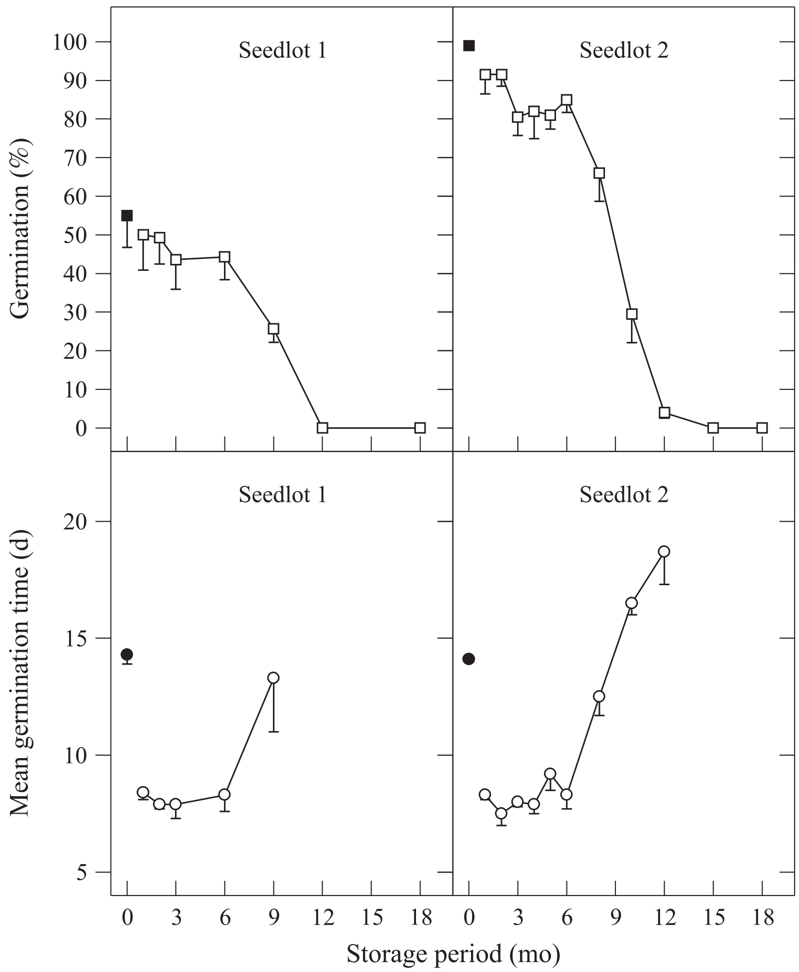

Fig. 2. Effects of 4℃ storage with moist sphagnum for 1~18 mo on the mean germination time and germination percentage of *Scolopia oldhamii* seeds. Filled circles (●) and squares (■) represent the mean germination time and germination percentage of fresh mature seeds. Vertical bars represent the standard error of the mean.

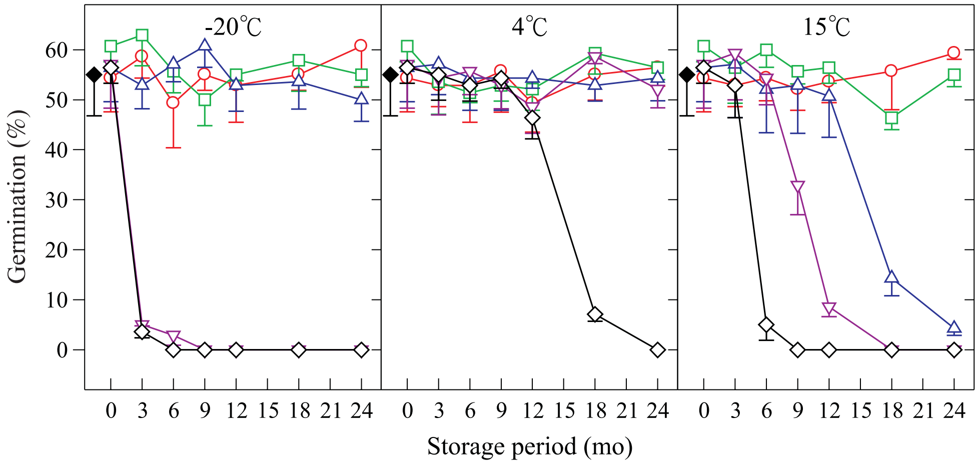

Fig. 3. Effects of storage temperatures (-20, 4, and 15℃) and moisture contents (2.0~14.5%, on a fresh-weight basis) on the germination percentage of seedlot 1 seeds of *Scolopia oldhamii* stored for up to 24 mo. Survival of seeds at the 5 moisture contents significantly differed ($p < 0.0001$), but no significant difference was found if seeds of sub-seedlots at moisture contents of 7.6, 10.8, and 14.5% were excluded from the analysis for all temperatures. The estimated initial germination percentage of fresh mature seeds was 55.0 ±8.2% (◆). Moisture contents of seeds: ○, 2.0±0.2%; □, 5.3±0.1%; △, 7.6±0.2%; ▽, 10.8±0.2%; ◇, 14.5±0.4%. Vertical bars represent the standard error of the mean.

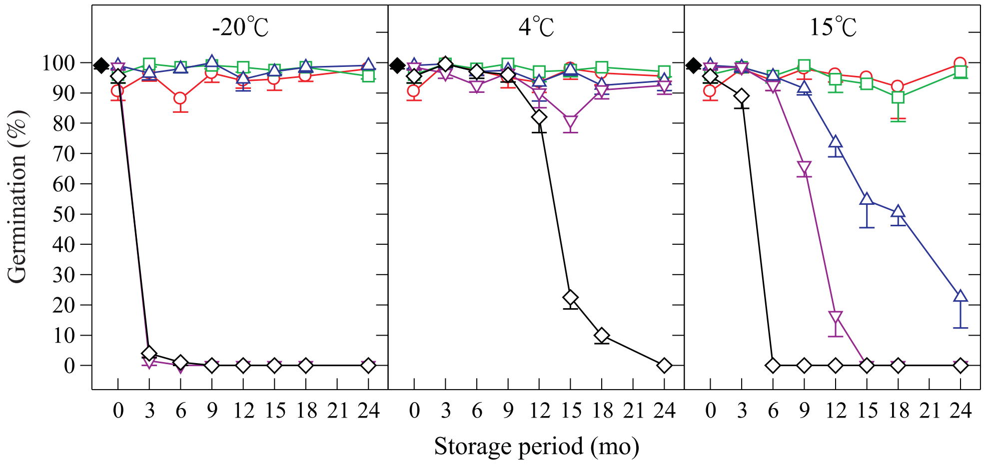

Fig. 4. Effects of storage temperatures (-20, 4, and 15℃) and moisture contents (2.3~14.3%, on a fresh-weight basis) on the germination percentage of seedlot 2 seeds of *Scolopia oldhamii* stored for up to 24 mo. Survival of seeds at the 5 moisture contents significantly differed ($p < 0.0001$), but no significant difference was found if seeds of sub-seedlots at moisture contents of 7.2, 11.3 and 14.3% were excluded from the analysis for all temperatures. The estimated initial germination percentage of fresh mature seeds was 99.0 ±1.0% (◆). Moisture contents of seeds: ○, 2.3±0.3%; □, 4.9±0.1%; △, 7.2±0.2%; ▽, 11.3±0.1%; ◇, 14.3±0.3%. Vertical bars represent the standard error of the mean.

2.3% were 95.5, 98.5, 99.0, 96.0, and 90.5%, respectively. No significant difference ($p > 0.05$) was observed among the germination percentages of these 5 moisture levels of dried seeds and fresh sample (99.0%). Of the above seeds with 5 moisture levels, seeds at 11.3 and 14.3% MCs had almost completely lost their viability after 3 mo of storage at -20℃ (Fig. 4). The other 3 lower-MC seeds had similar germination results after storage for 24 mo, and their germination percentages at MCs of 7.2, 4.9, and 2.3% were 99.0, 95.5, and 98.0%, respectively (Fig. 4). Thus, there was no significant difference in germination compared to fresh seeds ($p > 0.1$). After storage at 4℃ for 24 mo, no significant decrease in germination was found among seeds at the 4 lower moisture levels compared to fresh seeds ($p > 0.05$). However, a significant difference was found in seeds at a 14.5% MC (82.0%) after storage for 12 mo ($p < 0.05$). After 15 mo of storage, the germination percentage had sharply decreased to 22.5%, and the seeds had totally lost viability after 24 mo. Moreover, the germination percentages of seeds at MCs of 11.3, 7.2, 4.9, and 2.3% were 92.5, 94.0, 97.0, and 95.5% after 24-mo storage, respectively. There was no significant difference in germination compared to fresh seeds ($p > 0.05$). After storage at 15℃ for 24 mo, only 2 sub-seedlots of seeds at 4.9 and 2.3% MCs still remained viable. The germination percentage of seeds at a 7.2% MC decreased to 22.5%, while seeds at 11.3 and 14.3% MCs lost viability quickly and had all died after storage for 15 and 6 mo, respectively. The results above reveal that when *S. oldhamii* seeds were dehydrated to 2.0~7.6% MCs, they still maintained good viability after storage at 4 and -20℃ for 24 mo. Due to their tolerance of desiccation and low temperatures, *S. oldhamii* seeds are qualified as orthodox as defined by of Hong and Ellis (1996).

DISCUSSION

Fresh seeds of seedlot 1 only showed a 55% germination percentage, even if these seeds were carefully selected. A possible explanation for this may lie in the degree of their maturation. Collected in the middle of January, the fruits of seedlot 1 were almost all light red, while some were still green. The after-ripened fruits turned red after being moistened in a greenhouse. Then, the seeds were washed and extracted. The initial germination was relatively low because the seeds might have been not fully mature. However, seedlot 2 showed different results from seedlot 1. Fruits of seedlot 2 were mature and dark purple at the time of collection. Thus, these seeds had a higher initial germination of up to 99%. To reach maximum germination, we recommend that *S. oldhamii* fruits be collected when they are fully mature and appear dark red or dark purple. Moreover, seed collection is in approximately late January to the middle of February, but the collection time should depend on the color of mature fruits at the time of fruit harvest.

Hong and Ellis (1996) used 2 parameters, thousandseed weight and MC, to categorize seed storage behaviors of the species examined. Their samples were collected from mature seeds of 94 species of 5 families, including the Aceraceae, Araucariaceae, Dipterocarpaceae, Fagaceae, and Myrtaceae. The findings of their study clarified seed storage behaviors as follows. The MC of mature recalcitrant seeds was 36~90%. Mature seeds with a MC of < 35% were not seen as recalcitrant seeds and those with an MC of < 23% are orthodox seeds. In addition, mature seeds at MCs of 23~55% are intermediate seeds. Furthermore, a thousand seed weight of < 25 g is classified as orthodox while a weight above 13,000 g is seen as recalcitrant.

If the weight is between 30 and 13,000 g, the seeds could be orthodox, recalcitrant, or intermediate. In this study, seeds of *S. oldhamii* were determined to be orthodox seeds. However, the storage behavior of *Zelkova serrata* (Thunb.) Makino seeds is intermediate, although a thousand seed weight of fresh mature seeds of *Z. serrata* is about 12.5 g, and the seed MC is about 13~21% (Yang et al. 2007). The MC of mature seeds of *S. oldhamii* was 10~12%, and thousand seed weight was only about 7 g (Table 2). For the species examined, the relation between the MC and thousand seed weight to seed storage behavior was consistent with Hong and Ellis's criteria (1996).

The results of this study show that *S. oldhamii* seeds were non-dormant and had a high germination speed. In addition, 1~6 mo of 4℃ stratification decreased the MGT from 13 to 8 d. However, this method failed to improve germinability and caused the seeds to lose their viability beyond 6-mo storage (Fig. 2). After stratification for 9 mo, a significant decrease in seed viability was found, and the seeds had totally lost all viability after 12 mo. This species, which is intolerant of wet and low-temperature conditions, is similar to *Cordia dichotoma* G. Forst. seeds (Yang et al. 2006a). In short, the method of 4℃ stratification in the nursery of *S. oldhamii* seeds is not only unnecessary but also inadequate for short-time storage.

Generally speaking, when seeds at a MC of > 15% are stored at sub-zero temperatures, their free water turns into ice crystals which cause freeze damage to the seeds (Tompsett 1985). Before sub-zero storage, MCs should be decreased to < 14% in case the available water in cells freezes and causes membranes to break (Copeland and McDonald 1995). Thus, this method keeps seeds from deteriorating. However, when the MC of *S. oldhamii* seeds, which qualify as orthodox seeds, exceeded a specific MC level, marked deterioration in seed viability for frozen storage was more serious than those stored at 4 and 15℃ (Figs. 3, 4). We inferred that *S. oldhamii* seeds suffered freeze damage when the MC was > 11%. Apparently, their ability to protect against freeze damage is weaker than that of other orthodox seeds. In this study, when the MC of *S. oldhamii* seeds was > 11%, freeze damage occurred and seed viability sharply decreased after storage at -20℃ for 3 mo. As the MC increased, the harmful effect of freeze damage was simultaneously strengthened within a short time (Figs. 3, 4). Many species show similar characteristics, including *Chionanthus retusus* Lindl. and Paxt. (Yang and Lin 2004), *Sapium discolor* Muell.-Arg. (Yang et al. 2006b), and *Celtis sinensis* Pers. (Yang et al. 2006a). When these 3 species were respectively dried to 8, 9, and 9% MCs for storage at -20℃, they suffered freeze damage and rapidly died. However, *S. oldhamii* seeds at MCs of 2~8% still maintained good viability after storage at -20℃ for 2 yr. One reason might be that the period of our test was not long enough so that the effects of freeze damage on those seeds at a slightly higher MC of 7.6% were not clear within 2 yr. In conclusion, when the method of long-term sub-zero storage is applied for *S. oldhamii* seeds, we recommend following the rules of FAO/IPGRI (1994). That is, the MC of orthodox seeds should be 3~7% to avoid excess moisture causing seed deterioration. If the strategy for short-term storage within 2 yr is used for *S. oldhamii* seeds, decreasing their MC to < 11% for storage at 4℃ is efficient, and their germination capability does not decline (Figs. 3, 4).

CONCLUSIONS

Seeds of *S. oldhamii* exhibited no dormancy and fully germinated at the fluctuating temperatures of 30/20℃ within 4 wk. In addition, seed germination mainly took place in the 2nd wk.

Stratification at 4℃ was not efficient in improving the germinability of *S. oldhamii* seeds, but only decreased the MGTs. Therefore, this strategy was not effective in the nursery. After moist storage at 4℃ for more than 6 mo, seed viability sharply decreased. In other words, low-temperature moist storage is not effective for temporarily storing *S. oldhamii* seeds.

Seeds of *S. oldhamii* can tolerate dessication. Seed viability did not decrease when dehydrated to an MC of 2%. Moreover, *S. oldhamii* seeds can survive sub-zero temperatures. After storage at -20 and 4℃ for 2 yr, a significant decrease in germination was not found in seeds with 2~7% MCs. Thus, *S. oldhamii* seeds show orthodox storage behavior, and a long-term storage strategy can be adopted for future demands. We recommend that *S. oldhamii* seeds be dehydrated to 3~7% MCs and then hermetically sealed when using the strategy of long-term sub-zero storage. When short-term storage within 2 yr is applied, the MC of the seeds can be decreased to < 11% and then the seeds stored at about 4℃ after being hermetically sealed.

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