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CONSERVATION BIOLOGY FOR SEVEN PALM SPECIES FROM DIVERSE GENERA

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ABSTRACT

Arecaceae are a relatively large family with a considerable number of species of local and global
socioeconomic importance. Many species are also under threat of impending extinction, indicating an
urgent need to improve their conservation prospects. Here we present studies on seven palm species
(*Adonidia merrillii, Caryota urens, Livistona muelleri, Ravenea rivularis, Sabal* [*minor* var. *louisiana*],
Trachycarpus latisectus, and *Wallichia disticha*) from diverse genera in relation to various seed traits,
including germination, desiccation tolerance, and weight. Germination varied from ca. 12–100% and
mean time to germinate ranged from four days to four and one-half weeks at 30°C. Six of the species
were newly screened for seed conservation biology and of these two were found to possess desiccation-
tolerant seeds, indicating opportunities for longer-term storage and improved use.

Key words: Arecaceae, desiccation, germination, palms, seeds, tolerance.

INTRODUCTION

Worldwide biodiversity loss is a widely accepted crisis
that requires strategic action to elicit a global response
(Wood et al. 2000). One example of this is the recently
launched United Nations Global Strategy for Plant Conser-
vation, which encourages government bodies to conserve
90% of threatened plant species by 2010 (Convention on
Biological Diversity, CBD 2001–2004). This can be
achieved by either utilizing in situ (i.e., conservation within
the plants' native habitat) and/or ex situ (i.e., out of the
plants' native habitat) approaches (Miller et al. 1995). Ex
situ methods, including seed banks, are seen as important
components of an integrated conservation approach that
complements in situ techniques (Smith et al. 1998). Indeed,
over the last 25 years seed banking has become well estab-
lished as a method for storing the seeds of both crop and
nondomesticated plants (Plucknett et al. 1987; Chin 1994;
Miller et al. 1995; Linington and Pritchard 2001). However,
the seed biology of tree and herbaceous species is, in gen-
eral, under-researched; seed storage physiology has been de-
scribed for only ca. 9000 species (Tweddle et al. 2003) out
of a predicted number of seed-bearing plants in the region
of ca. 240,000–400,000. For palms, baseline data is partic-
ularly meager, with detailed seed storage and germination
information available for less than 200 species (e.g., Davies
and Pritchard 1998*a, b*; Pritchard and Davies 1998; Tweddle
et al. 2003; Wood and Pritchard 2003; Pritchard et al. 2004)
out of ca. 2700 in the family (Royal Botanic Gardens, Kew
2003). Clearly, a greater knowledge of palm seed biology
will improve the conservation prospects for this highly uti-
lized and threatened group of species (Johnson 1996).

Semina Palmarum, part of the Millennium Seed Bank Pro-

ject, aims to assess experimentally the seed conservation po-
tential of 200 palms species over a five-year period, thereby
approximately doubling baseline knowledge. In the first
phase of the project a screening method has been developed,
using only 100 seeds, which allows the quantification of seed
moisture content, equilibrium relative humidity, morphom-
etry, initial germination, short-term storage effects, desic-
cation tolerance, germination rate (mean time to germinate),
and seedling morphology (Pritchard et al. 2004). To date this
approach has been applied to four species each of *Phoenix*
L. and *Syagrus* Mart. by the authors. Here we present data
for seven more species from diverse genera; six of which
are new to seed conservation studies.

MATERIALS AND METHODS

For five species, commercial lots of 100 cleaned seeds
(fruit removed) were purchased from The Palm Centre
(Ham, Richmond, Surrey, UK). Seeds of the other two spe-
cies investigated (*Caryota urens* and *Ravenea rivularis*)
were provided by Xishuangbanna Tropical Botanic Garden,
China, and Silo National des Graines Forestières, Madagas-
car, respectively. Seeds of *Sabal minor* when purchased were
described as *Sabal minor* var. *louisiana* and is referred to as
such throughout the text. Upon receipt, basic seed charac-
teristics were determined; dimensions were taken from 20
seeds using a digital caliper and the fresh weights of a sim-
ilar number of seeds were also recorded.

Seed moisture contents were gravimetrically determined
before and after drying utilizing a 103°C oven for 17 ± 1
hr (International Seed Testing Association [ISTA] 1999) on
at least five individuals (consisting of the endocarp, endo-
sperm, and embryo; botanically, a pyrene). After oven-dry-
ing, seed weights were used as determinants of seed dry
mass. For each seed lot, duplicate determinations of seed
equilibrium relative humidity (eRH) were also made on sub-
lots of usually no more than 20 seeds over a 1 hr period at

20°C using a Rotronic AWVC DIO sensor attached to a Hygro-Palm 3 unit (Rotronic Ltd., Crawley, UK). In the case of *Sabal minor* var. *louisiana,* because of their smaller size, 50 seeds were utilized for eRH measurements (Table 1).

Seeds were held in the short term (a few weeks) at 15°C at or around their initial moisture content (Table 1) in loosely tied, polythene bags containing a small amount of moistened vermiculite. This phased start to the work removed time constraints from the desiccation experiments, although introducing the dimension of post-harvest maturation. As a consequence, an undried, moist-stored control was sown for germination when the desiccation experiment concluded, which reduced the number of seeds available for post-desiccation testing, but facilitated an assessment of post-harvest maturation.

For germination, replicate batches of 20 seeds were sown, at 30°C on 1% agar in Perspex sandwich boxes (175 × 115 × 60 mm). Light was supplied by cool white fluorescent tubes for 12 hr d^{-1}. Seed germination was recorded when a coleorhiza-like organ had lifted the seed operculum and the emerging tissue protruded by >2 mm in length.

Mean time to germinate (MTG), in days, was calculated according to the following equation:

$$MTG = \sum_{i=1}^{D} \frac{d_i n_i}{N}$$

where n_i is the number of seeds that germinated on day d_i and N was the total number of seeds that germinated over the duration of the germination test, D (typically ca. 90 days).

A method modified from Grout et al. (1983) for drying palm seeds, involved mixing seeds with an equal weight of silica gel in sealed polythene bags and placing them at 25°C (Pritchard et al. 2004). The weight of a sub-sample was monitored regularly after separating the seeds from the silica gel. This allowed the approaching equilibrium weight to be assessed and also facilitated regular ventilation of the bags, which avoided oxygen depletion. The time taken to dry to a target moisture content of 5% varied among seed lots, from ca. 1–2 mo. Seed moisture contents and eRHs were determined after drying as described above.

The species used are detailed in Table 1 and the commitment allocation of seed numbers to each part of the test is shown in Table 2.

RESULTS

Seed Lot Characteristics

Inter- and intra-specific seed characteristics are shown in Table 1. For the seven species detailed mean seed dry mass varied by ca. 1.5 orders of magnitude, from 121 mg (*Ravenea rivularis*) to ca. 3 g (*Adonidia merrillii*). The mean seed dimensions for these two species were 10.5 × 7.4 mm and 30.8 × 17.1 mm, respectively. Variability within seed lots was also evident with the fresh weight of seeds on receipt, by up to 1.5-fold. In the case of *Ravenea rivularis* there was as much as a 2-fold variation in seed fresh weight.

On receipt, equilibrium relative humidities within the test containers were high for each seed lot (69–95%) and whole seed (including endocarp) moisture contents varied from 14–40% (Table 1).

Table 1. Characteristics of the species and the seed lots used in this study.

Species[a]	Habitat (Country)	Seed dimensions (L × B mm)[b]	Range of seed fresh weight on receipt [Mean seed dry weight] (mg)	On receipt: Seed moisture content % [eRH]	After drying: Seed moisture content % [eRH]
Adonidia merrillii (Becc.) Becc.	Rainforest (Philippines, New Hebrides, Fiji)	30.8 × 17.1	3451–3978 [2937 ± 49]	33.4 ± 1.3 [95 ± 2]	5.8 ± 0.3 [3 ± 1]
Caryota urens L.	Monsoon to humid (India, Myanmar [Burma], Sri Lanka, Solomon Islands)	16.1 × 19.5	810–1494 [697 ± 63]	36.4 ± 0.2 [93 ± 2]	4.8 ± 0.2 [8 ± 2]
Livistona muelleri F. M. Bailey	Open coastal areas (Australia, Africa, Solomon Islands)	10.1 × 7.4	289–322 [228 ± 29]	24.0 ± 0.2 [94 ± 2]	3.8 ± 0.2 [6 ± 2]
Ravenea rivularis Jum. & H. Perrier	Dry forest to river banks (central Madagascar)	10.5 × 7.4	165–335 [121 ± 10]	40.0 ± 0.5 [95 ± 2]	4.1 ± 0.0 [13 ± 2]
Sabal minor var. *louisiana* (Jacq.) Pers.	Low lying scrub areas (Colombia, NE Mexico, SE United States)	5.4 × 6.9	169–180 [155 ± 20]	13.5 ± 0.1 [75 ± 5]	3.2 ± 0.2 [5 ± 3]
Trachycarpus latisectus Spanner, H. J. Noltie & M. Gibbons	Littoral woodlands (Sikkim Himalayas, NE India)	12.6 × 8.8	581–657 [541 ± 18]	18.0 ± 0.1 [86 ± 4]	4.4 ± 0.2 [15 ± 3]
Wallichia disticha T. Anderson	Moist forest mountain valleys (Sikkim Himalayas, NE India)	10.4 × 7.4	245–311 [203 ± 6]	29.2 ± 0.4 [69 ± 4]	4.7 ± 0.1 [23 ± 10]

[a] Names were accepted as those given by the seed provider and authorities taken from International Plant Name Index (IPNI 2004).
[b] Standard errors are not shown but were a maximum of 0.9 mm, for *Adonidia merrillii*.

Table 2. Seed numbers balance sheet for the 100-seed test (after Pritchard et al. 2004).

Task	Number of seeds in test	Destructive (D) or non-destructive test (ND)	Cumulative number of seeds used
On arrival			
Length and breadth	20	ND	0
Fresh weight on arrival	20	ND	0
eRH	2 × 10	ND	0
Moisture content	10	D	10
Germination	2 × 20	D	50
Dessication study[a]			
eRH	2 × 10	ND	50
Drying curve	ca. 20	ND	50
Moisture content	5	D	55
Germination	2 × 20	D	95
Carpological collection	5	ND	100
		(but retained)	

[a] If initiation of the desiccation trial is delayed, 1 × 20 seeds can be sown to re-check viability and a similar number assessed after desiccation.

Germination and Seed Quality

Initial germination levels were extremely variable across the seven seed lots (Fig. 1–7), from the low level of 35% (*Sabal louisiana* and *Trachycarpus latisectus*, Fig. 5, 6) to 100% (*Adonidia merrillii* and *Wallichia disticha*, Fig. 1, 7). Germination started after about two weeks and concluded by two months (Fig. 1–7). Mean time for germination ranged from as little as 12 days (*Ravenea rivularis*, Fig. 4) to 30 days (*Trachycarpus latisectus*, Fig. 6).

Maximum germination levels of all 74 species tested so far in the *Semina Palmarum* project ranged from 0–100% (Fig. 8). Thirty-nine seed lots had germination values that were <25%, 11 had values ranging from 26–50%, 9 were between 51–75% and 15 species had high germination values between 76–100% (Fig. 8).

Effects of Moist Storage and Desiccation

The response of seeds to moist storage and desiccation is shown in Fig. 1–7. Seed responses fell into four categories: (i) four species failed to germinate after desiccation: *Adonidia merrillii* (Fig. 1), *Caryota urens* (Fig. 2), *Ravenea rivularis* (Fig. 4), and *Trachycarpus latisectus* (Fig. 6); (ii) *Wallichia disticha* (Fig. 7) had slightly reduced germination after desiccation, while the fresh- and moist-stored seeds had similar germination levels; (iii) *Sabal minor* var. *louisiana* (Fig. 5) exhibited improved germination, both after moist storage and drying; (iv) *Livistona muelleri* (Fig. 3) had little germination after drying, although the single surviving seed produced a healthy seedling/plantlet reaching 32 cm in height, with four leaves after ca. 3.5 months of greenhouse growth.

For three of the desiccation-sensitive species (*Adonidia merrillii, Caryota urens,* and *Ravenea rivularis*) germination levels were similar for both the fresh- and moist-stored seeds. In contrast, seed germination in *Trachycarpus latisectus* (Fig. 6) declined after moist storage. For the species

that tolerated desiccation (Fig. 3, 5, and 7), the mean time to germinate appeared not to change after the drying treatment.

Seed Characteristics and Quality

Many of the collections used in this study were obtained from a specialist commercial source, the exceptions being *Caryota urens* (Xishuangbanna Tropical Botanic Garden, China) and *Ravenea rivularis* (Silo National des Graines Forestières, Madagascar) that were provided through collaboration. Although the commercial supplier used is well established, herbarium vouchers could not be provided with the material. Therefore, we have taken long-term measures to address concerns over species identification: (i) for each species, five plants are "grown-on" from germination tests so that identification may be possible in the future; (ii) digital photographs of both the seed surface and cross sections have been taken; (iii) seed mass is recorded and a small number of seeds are maintained as a carpological collection (see Table 1), again for reasons of comparison and identification.

A chi^2 test has been carried out on all the maximum germination data (all treatments) for the 74 species investigated so far in the *Semina Palmarum* project (Fig. 8) to determine whether the distribution of the data was random or skewed. The results were 28.378 ($P = 0.05$, 3 degrees of freedom) with a critical value of 7.815, revealing that seeds received from the commercial source were of a lower quality than, (i) would be expected if the seeds were of randomly spread viability (Fig. 8 is skewed towards lower germination %), and (ii) those of the botanic garden donations (*Caryota urens* and *Ravenea rivularis*). This suggests a problem with the commercially sourced material that could have arisen at harvest or during processing and/or transportation. However, it has been noted that less than 20% of palm seeds germinate under natural conditions (Robinson 2002). This, however, may be a function of seed maturity; as for a variety of species, highest germination levels are achieved only when seeds are harvested fully ripened (Hay et al. 1997; Pritchard and Davies 1998). Palm seeds do not mature simultaneously, developing sequentially on the infructescence, which may account for our mixed or low germination results (Fig. 8). Obtaining more donations from botanic gardens, particularly those with conservation stands, may help in securing higher quality material, as well as providing a more adequate means of initial species identification.

Germination

Germination of all the seven species detailed was relatively rapid, starting from as early as ca. four days and as late as ca. three weeks (Fig. 1–7). Germination times have previously been reported for all but two (*Adonidia merrillii* and *Trachycarpus latisectus*) of the seven species presently studied (Ellison and Ellison 2001). However, germination in this study was quicker than the 2–10 months reported by Ellison and Ellison (2001); only *Sabal minor* var. *louisiana* previously had germination times near those (4–8 weeks) suggested by Ellison and Ellison (2001). It has been noted that pre-soaking of palms seeds of some species can either
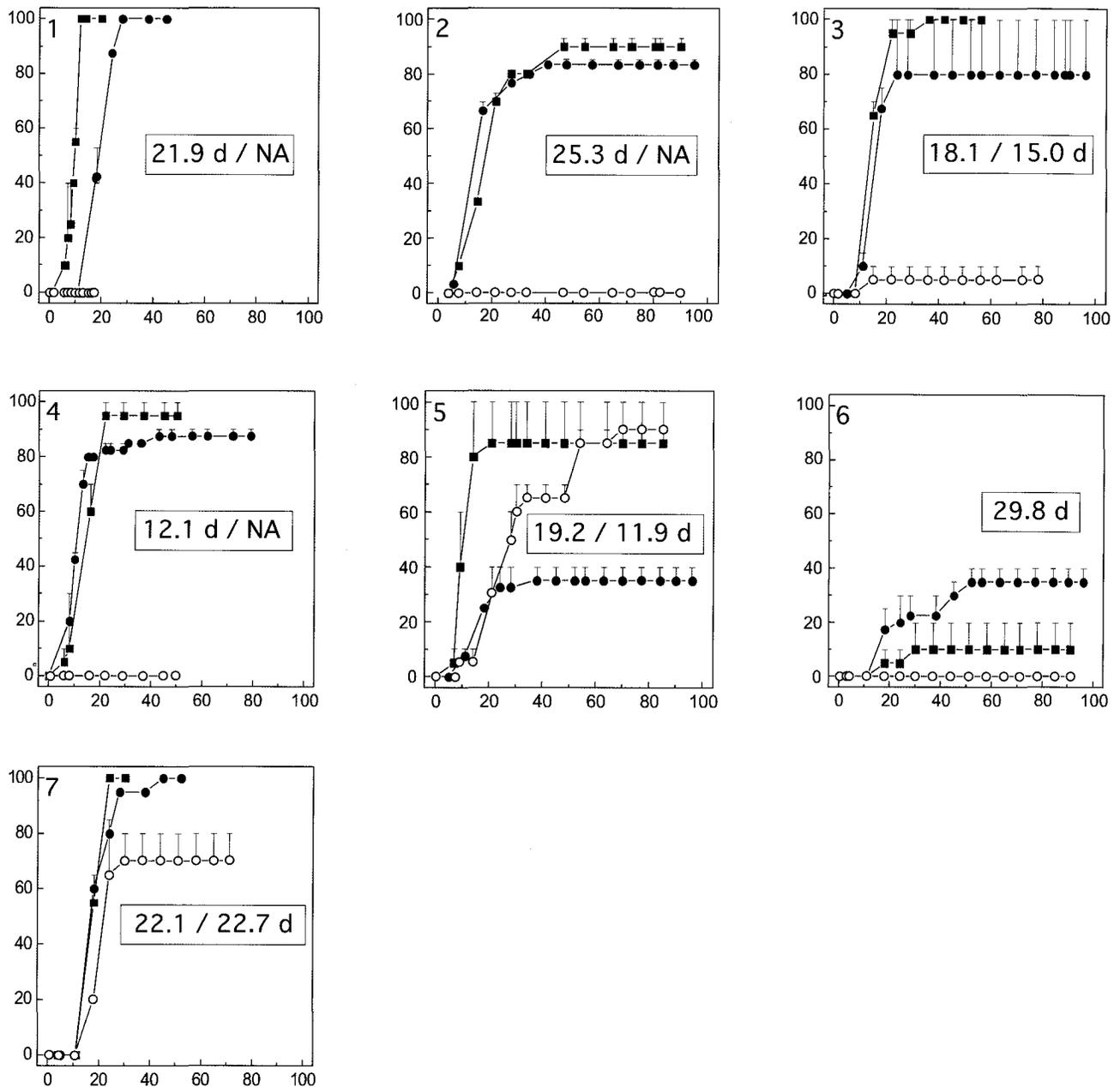
Fig. 1–7.—Effects of moist storage and desiccation on seed germination (N = 20).—1 *Adonidia merrillii.*—2. *Caryota urens.*—3. *Livistona muelleri.*—4. *Ravenea rivularis.*—5. *Sabal minor* [var. *louisiana*].—6. *Trachycarpus latisectus.*—7. *Wallichia disticha.* Seeds were sown at 30°C in the light (12 h d^{-1}) when either fresh (closed circles), stored moist for a few weeks (closed squares), or after drying with silica gel (open circles) to low relative humidity (5–23%). Mean times to germination (MTG) in days are shown for the fresh and dried seeds. NA = not applicable.

improve germination per se (Carpenter 1987; Davies and Pritchard 1988a; Ellison and Ellison 2001) or reduce the mean time for germination, presumably by removing the need for prolonged imbibition periods when water is presumably taken up slowly from the germination medium (Wood and Pritchard 2003). However, this treatment did not appear to be necessary for satisfactory germination at 30°C in this study.

An incubation temperature of 30°C was selected to simulate those in the natural environments of the majority of the species. This temperature has previously been demon-

strated to favor high levels of germination in other palm species, e.g., *Phoenix dactylifera* L., *Phoenix humilis* Cav., and *Hyophorbe lagenicaulis* (L. H. Bailey) H. E. Moore (Sento 1976; Wood and Pritchard 2003).

While the effects were subtle in some seed lots, for six of the species presently studied germination improved after short-term moist storage, usually in terms of both quantity and rate. This response was especially pronounced in *Sabal minor* var. *louisiana* (Fig. 5). This, again, may be related to seed maturity. During moist storage the embryo continues to develop, which is then reflected in the apparent increase in
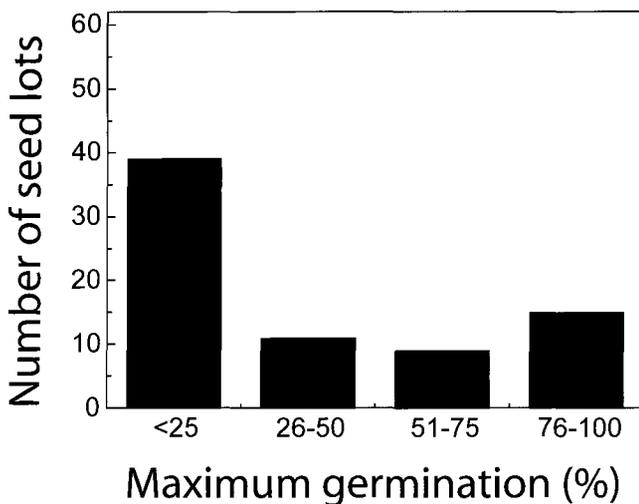
Fig. 8.—Histogram showing the maximum germination upon receipt of all 74 seed lots currently investigated during the *Semina Palmarum* project. Seeds were sown at 30°C in the light (12 hr d^{-1}). The vast majority of seed lots represented were purchased commercially, with two exceptions: (i) *Caryota urens* (Xishuangbanna Tropical Botanic Gardens, China) and (ii) *Ravenea rivularis* (Silo National des Graines Forestières, Madagascar). Both of these seed lots had germination levels of >80%.

seed quality with respect to germination (Hay and Smith 2003). A similar response was also seen for seeds of *Sabal minor* var. *louisiana* when dried. Maturation during drying is also known to improve seed quality traits in some desiccation-tolerant species (Hong and Ellis 1990; Hay and Probert 1995), although this is often a feature of slower desiccation rather than the rapid enforced silica drying used here (Hay and Smith 2003).

Desiccation Tolerance and Seed Ecology

Seed storage characteristics have previously been described for only one of the seven genera examined (*Sabal* Adans.; Tweddle et al. 2003). In the present study *Sabal minor* var. *louisiana* and *Wallichia disticha* exhibited desiccation tolerance. The genus *Sabal* comprises about 14 species occurring from Colombia to northeast Mexico and the southeast United States. Their habitats are diverse; for example, *S. minor* is found in swampy areas, other species occur in sandy coastal and/or dry, open areas (Uhl and Dransfield 1987). Leon (1961) classified the genus as having long-lived seeds, possibly implying that the seeds could survive when equilibrated to ambient relative humidity (RH) conditions. The storage behavior of *S. minor* has previously been described as "probably orthodox" (i.e., desiccation tolerant: Roberts 1973; Hong et al. 1998) on the basis that 32 months of storage at 3–5°C after air drying had no effect on seed viability (Sento 1976). Thus, the findings from this study corroborate the hypothesis that *S. minor* var. *louisiana* is desiccation tolerant. For the remaining six species examined, there are apparently no seed storage type indicators (Hong et al. 1998; Tweddle et al. 2003). At the genus level only three are mentioned by these authors: *Caryota* L., *Livistona* R. Br., and *Trachycarpus* H. Wendl. It is suggested that the storage classification may be uncertain for *Caryota*

and desiccation tolerant for the remaining two groups (Hong et al. 1998; Tweddle et al. 2003).

The ability of the seeds to tolerate desiccation appears to relate closely to the native habitat in which the plants grow. *Adonidia merrillii,* for example, is classified within a genus of about 18 species from the New Hebrides to Fiji and the Philippine Islands, commonly growing in coastal areas or in cloud forests (Uhl and Dransfield 1987). This may explain the seed sensitivity to dehydration (Fig. 1) as dry conditions are unlikely to prevail. *Caryota urens* is also desiccation sensitive and grows in wet (monsoon) climates from Sri Lanka through Southeast Asia to the Solomon Islands (Uhl and Dransfield 1987). Similar correlations may be made for species of *Livistona.* Seeds of *Livistona muelleri* appeared to be desiccation sensitive in this study. *Livistona muelleri* belongs to a genus of 28 species inhabiting areas ranging from the Horn of Africa to the Solomon Islands and Australia. A great diversity is found within this genus, particularly in those species found in the Solomon Islands and Australia (Uhl and Dransfield 1987). Some are adapted to freshwater and peat-swamp forest (*L. saribus* Merrill ex A. Chev.) and others to dry savannah woodland (*L. humilis* R. Br. and *L. carinensis* (Chiov.) J. Dransfield and N. W. Uhl). However, *L. muelleri* is commonly found along coastal areas and may therefore remain hydrated for extended time periods. However, although germination was severely reduced after drying, one seed of this species tolerated desiccation (Fig. 3), producing a healthy plant. Thus, the possible desiccation tolerance of *L. muelleri* warrants further investigation. In contrast, *Ravenea rivularis* is clearly desiccation sensitive (Fig. 4) and grows on, or near, watercourses (Ellison and Ellison 2001), again suggesting a broad correlation with habitat type. The anomalous species in this ecological trend is *Trachycarpus latisectus.* Species from this genus are found on limestone hills (Uhl and Dransfield 1987), where surface water availability may be in short supply. However, *Trachycarpus* palms also grow at altitudes of up to 2400 m in the Himalayas, where water may be available either in dew/mist formations or from melt-water run offs. However, the initial quality of the *T. latisectus* seeds presently used was low (ca. 40%) and had fallen by the time of the desiccation experiment (ca. 10%), which may have made the seeds more intolerant of drying (see Black and Pritchard 2002). Clearly, a more detailed investigation of the relationship between moisture content and seed germination/viability in this species is recommended, as well as for *Livistona muelleri* where survival of drying was low.

Inga Mill. species were first used to successfully determine that using small numbers of seeds could indicate desiccation tolerance or sensitivity, by drying below critical target moisture contents for germination (Pritchard et al. 1995). In this study, only one species not previously investigated demonstrated a convincing tolerance to drying: *Wallichia disticha.* Again, the seed response of this species to desiccation correlates well with the natural environment of the plant. The genus *Wallichia* comprises seven species that grow from the Indian Himalayas through Burma and China, and southwards to the Thai peninsula (Uhl and Dransfield 1987). *Wallichia disticha* is described as growing on steep sandstone declivities in eastern Sikkim (Anderson 1869; Uhl and Dransfield 1987). Although this area is typified by hu-

mid tropical forests, sandstone is an extremely porous material and therefore water availability may be limited, thus favoring desiccation-tolerant seeds.

It is also noticeable that two of the three smallest seeded species (*Sabal minor* var. *louisiana, Wallichia disticha,* but not *Ravenea rivularis*) have desiccation-tolerant seeds. The relationship between seed size and sensitivity to drying is a topic of recent debate (see Dickie and Pritchard 2002), where it has been noted that intolerance to drying is generally a feature of larger seeds (Table 1). The ecological reasons for this may be related to either the negation of a rehydration period for desiccation-sensitive seeds falling during a limited "wet-season" and/or their being able to germinate rapidly upon shedding.

The initial quality of some of the seed lots screened for desiccation tolerance was relatively low (< ca. 25%; Fig. 8) and this can affect the way in which seeds respond to desiccation. Desiccation decreased the mean time to germinate, albeit slightly, for three of the four species that exhibited desiccation tolerance, compared with the fresh- or moist-stored seeds (Fig. 1–7). In earlier studies on *Hyophorbe lagenicaulis,* mean time to germinate increased after desiccation as a result of the time requirement for the seeds to reach full hydration before germination could proceed (Wood and Pritchard 2003). Only one species detailed in this study followed this trend (*Wallichia disticha*). However, it is unlikely that a similar process is responsible for the increase in mean time to germinate for this species as the change in germination rate was insignificant (22.1 compared to 22.7 days; Fig. 7).

Seed Numbers

There are concerns when using small seed numbers to indicate desiccation tolerance. For instance, there may be variations in individual seed moisture contents during drying, such that some may be above target moisture contents after drying. Additionally, the use of target moisture contents during drying (Hong and Ellis 1996) to determine desiccation tolerance is debatable; e.g., differences in seed composition (physiological and biochemical) can result in seeds of the same moisture content having markedly different eRHs, and therefore water activities (both issues are discussed further in Pritchard et al. 2004). However, the seeds that survived drying (<5% moisture content; Table 1) in this study are clearly desiccation tolerant (Pammenter and Berjak 1999) and the phase of drying reached by the seeds in this study (data not shown) is consistent with them being below a population mean moisture content (Finch-Savage 1992).

Summary

In conclusion, we have provided new information on seed desiccation tolerance for six palm species. These data enable the conservation status of these species as ex situ collections to be judged. The data also suggest a relationship between tolerance to desiccation and the environment in which the plant naturally occurs. As the seed desiccation test is tailored toward the rapid production of such information, it could be used for any family or genus of interest, such as those targeted for conservation or sustainable use in restoration projects.

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