Seed and microsite limitations of large-seeded, zoochorous trees in tropical forest restoration plantations in northern Thailand

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A B S T R A C T

On deforested or degraded land, planting mixtures of native forest tree species facilitates establishment of incoming tree seedling species (species recruitment) by rapidly re-establishing canopy cover. However, delayed colonization of plantations by large-seeded, zoochorous (LSZ) tree species can affect the species composition, structure, and function of the developing forest. The objective of this study was to investigate whether microsites limit establishment (germination and early seedling survival) of LSZ tree species in three 13-year-old, seasonally dry, tropical forest restoration plantations in northern Thailand. We conducted a seed sowing experiment that combined five LSZ tree species with four microsite treatments in a split-plot design. All five tree species were previously absent from the understory of the plantations, despite seed sources being present in nearby natural forest. The four treatments simulated potential microsites that naturally-dispersed seeds may encounter. They included seed deposition on the surface of soil and on leaf litter, as well as seed burial beneath soil and beneath leaf litter. We fenced the experimental areas to prevent seed predation and focus on the environmental effects of microsites on the different stages of seedling establishment. Following seed sowing, we measured germination and seedling survival, mean height, and stem diameter over 26 months. The microsites treatments did not significantly affect germination and seedling survival, providing strong evidence that establishment of the tested species was not limited by the applied microsite environments. Furthermore, although there were significant differences in germination and survival among species, relatively high overall germination (37%) and 26-month seedling survival (58.5%), irrespective of microsite treatment, suggested that environmental conditions in the 13-year-old plantations generally met the requirements for germination and early seedling establishment. Microsite treatments also did not correlate with seedling height or stem diameter at 20 months, suggesting that initial germination microsites have no effect on seedling growth and robustness. Taken together, the results of this investigation support the hypothesis that seedlings of LSZ tree species are slow to colonize tropical forest restoration plantations because of seed limitations resulting from inadequate seed dispersal or low seed availability. The successful establishment of LSZ seedlings from sown seeds in this experiment suggests that direct seeding beneath the canopy of restoration plantations may be an effective way to offset seed dispersal limitations in restoration plantations. Furthermore, the absence of microsite treatment effects suggests broadcast sowing of seeds may be a simple way to recolonize closed-canopy restoration plantations with LSZ tree species.

1. Introduction

Tropical forest restoration using mixed-species plantings of indigenous trees can rapidly re-establish tree canopy cover on deforested and degraded lands (de Souza and Batista, 2004, Lamb, 2011, Elliott et al., 2013). These canopies facilitate recolonization of the site by additional, non-planted tree species (species recruitment), by shading out herbaceous weeds, improving soil conditions, ameliorating the understory microclimate, and attracting seed-dispersing wildlife (Lamb,

2011, Goosem and Tucker, 2012). In forest restoration sites that exist close to natural forest, such species recruitment may occur rapidly, increasing tree species richness by accelerating diversification of understory regeneration (Sinhaseni, 2008, Lamb, 2011, Bertacchi et al., 2016). Still most tropical forest restoration plantations are relatively young (< 25 years), thus little is known yet about the long-term recovery of their tree species composition. In some older, naturally regenerating, secondary tropical forests, however, tree species composition remains distinct from that of primary forests even many decades

after canopy closure (Aide et al., 2000, Chazdon, 2003, Brearley et al., 2004, Chua et al., 2013). This suggests that there may be limits to the ability of natural recruitment to recreate the original tree species composition in recovering forest ecosystems. These limits may be particularly strong for large-seeded, zoochorous (animal-dispersed) tree species (LSZ) as they are often among the last to return to secondary forests (Aide et al., 2000, Chazdon, 2003, Brearley et al., 2004, Chua et al., 2013).

Reduced or delayed recruitment of LSZ trees may have consequences, both for regeneration of these species and for the composition, structure, and ecological functioning of the developing forests. LSZ species are often long-lived, shade-tolerant, late-successional species that are able to regenerate in the forest understory (Leishman et al., 2000). Such trees tend to have higher wood density and higher rates of CO_2 fixation than smaller-seeded species and are larger at maturity (Osuri and Sankaran, 2016). Consequently, their absence from forests may alter not only forest composition, but also forest structure and function. In addition, the failure of some LSZ tree species to regenerate may increase their risk of regional extirpation or even extinction (Brodie et al., 2009, Wotton and Kelly, 2011, Caughlin et al., 2014).

The seeds of most late-successional LSZ species are recalcitrant, that is, they are able to germinate rapidly after shedding but lose viability quickly, particularly if they are exposed to desiccation (Pritchard et al., 2004, Daws et al., 2006). Rapid germination may allow LSZ species to avoid seed predation and, because they allocate relatively few resources to physical defense, to be more efficient at seed resource provisioning than orthodox (i.e. desiccation tolerant) seeds (Daws et al., 2005). The low longevity of recalcitrant seeds, however, generally precludes their accumulation in the soil seed bank (Vazquez-Yanes and Orozco-Segovia, 1996). Therefore in order for LSZ species to recolonize the restoration plantings, they must rely on frugivores to disperse their seeds soon after seed maturation. Yet large seeds are less likely to be dispersed than small seeds because the number of frugivorous species that are capable of dispersing a seed generally decreases with increasing seed size (Kitamura et al., 2002, Corlett, 2017). In addition, hunting and habitat loss have reduced populations of large frugivores throughout the tropics and extirpated them from large stretches of their original distributions, further decreasing the likelihood that large seeds will be dispersed (Wright et al., 2007). For these reasons, seed limitation resulting from poor dispersal is a frequently cited reason for the delayed return of LSZ tree species in regenerating forests (Lamb, 2011, Chazdon, 2014, Reid et al., 2015).

Simply increasing the number of LSZ seeds in the understory, however, may not guarantee their recruitment in restoration forests. Seedling establishment (seed germination followed by survival and growth of the seedling) in all forests may also be limited by the availability of establishment microsites with suitable biotic and abiotic conditions in the understory (Putz, 1983, Molofsky and Augspurger, 1992, Dalling and Hubbell, 2002). Such microsites are defined by features of the forest floor that may be on a scale of no more than a few centimeters, yet they often determine the abundance, survival, and early growth of plants (Harper, 1977, Whittaker and Levin, 1977). On forest restoration plantations, the importance of microsites for seedling establishment may be even greater than in naturally occurring forest, since the initial conditions of land requiring active restoration are often poor (e.g. soil compaction, low levels of soil organic matter and low soil water retention) due to long-term, intensive, agricultural activity, such as repeated weeding and burning (Chazdon, 2003, Lamb, 2011).

The seeds of LSZ species that are naturally dispersed into restoration forests are likely to lodge in microsites that are defined by the seed's location with respect to soil and leaf litter. For example, seeds that fall onto leaf litter or exposed soil may lodge where they have landed or they may work their way below litter or be buried beneath leaves or soil by seed-caching animals. Where seeds end up may determine their exposure to potential threats and affect their ability seeds to access resources such as light, water, and soil. Seeds resting on bare soil may benefit from good seed-soil hydraulic conductivity and favorable aeration (Makana and Thomas, 2005), but exposed seeds on both soil and leaf litter may also be at increased risk of seed predation and desiccation (Doust et al., 2006). Furthermore, seeds lodged on the surface of leaf litter may be blocked from reaching the mineral soil (Molofsky and Augspurger, 1992). By contrast, seeds that have worked their way below leaf litter or have been buried beneath leaves or soil may have access to soil and be shielded from predation and desiccation (Cintra, 1997, Dupuy and Chazdon, 2008), but may also be forced to expend greater amounts of energy to grow above the substrate. This expenditure may reduce the robustness of seedlings that manage to emerge (Molofsky and Augspurger, 1992, Peterson and Facelli, 1992).

Microsites that shield recalcitrant seeds from desiccation may be particularly important in seasonally dry tropical forests (SDTFs). In aseasonal tropical rainforests, microsite variations in light availability rather than moisture are the primary limiting environmental factor affecting seedling survival (Augspurger, 1984, Brown, 1996, Schnitzer and Carson, 2001). SDTFs are distinct from aseasonal tropical forest because they grow where there is at least one prolonged season (≥ 4 months) of severe to absolute drought (Dirzo et al., 2011). Thus in these forests moisture may be as or more limiting than light in SDTF (Vieira and Scariot, 2006, Poorter and Markesteijn, 2008, Ferreira et al., 2015). To maximize the availability of moisture for seedling establishment, many zoochorous SDTF tree species fruit at the beginning of the rainy season (Elliott et al., 1994, Daws et al., 2005), but the high year-to-year variability in timing and quantity of rainfall that is characteristic for this forest type may still leave seeds and seedlings vulnerable to mortality due to desiccation (Blain and Kellman, 1991, McLaren and McDonald, 2003, Vieira and Scariot, 2006).

In order to optimize the return of LSZ tree species to restored tropical forests, we need to understand the role that limiting factors play in their recruitment; but, to the best of our knowledge, the relative importance of seed and microsite limitations in a restoration forest setting has never been explicitly tested. We conducted a seed sowing experiment in13-year-old SDTF restoration plantations in northern Thailand to determine the degree to which recruitment of five LSZ tree species may be limited by seed availability and microsite conditions. Seedlings of the species used in this investigation had not recruited into plantations, despite the presence of mature individuals in the nearby natural forest. The experiment also investigated how initial microsite conditions might differentially affect the stages of seedling establishment, i.e. seed germination and early seedling survival and growth. Here we present the results of the two-year seed sowing experiment, explore the effects of the dry season on seedling survival, and suggest management practices that may assist in overcoming barriers that may prevent LSZ tree species recruitment in tropical forest restoration plantations.

2. Study site

Field work was conducted on three 0.16 ha experimental forest restoration plots along or immediately below the ridge of a watershed (1207–1310 m above mean sea level) in the upper Mae Sa Valley, Northern Thailand, approximately 2 km north of the village of Ban Mae Sa Mai (18°52′N, 98°51′E). Average annual precipitation at the nearest weather station to the plots was 2095 mm (Kog-Ma Watershed Research Station) (Elliott et al., 2003), with nearly all rain falling during the six-month wet season that extends from May to October. During the dry season (from November to April) precipitation averages less than 100 mm per month (Elliott et al., 2003).

The area was originally covered with seasonally dry evergreen forest (EGF) comprised of more than 250 tree species (~75% of which are evergreen) many of which are unique to this forest type (Maxwell and Elliott, 2001). However, much of the original EGF has been cleared for cultivation, tourism developments, and infrastructure. The forest in the study sites was cleared for cabbage cultivation about 20 years prior to restoration plantings. In 1996 local villagers decided to abandoned

cultivation, so that the land could be returned to forest, as part of a national reforestation project to mark the golden jubilee of King Bhumibol Adulyadej (Elliott et al., 2000).

The Forest Restoration Research Unit (FORRU) of Chiang Mai University assisted local villagers and forest department officials to establish experimental forest restoration plots at the site in 1998 (13 years before the start of the study described here) to test the framework species method (FSM) of forest restoration (Elliott et al., 2003). The technique seeks to accelerate forest succession by planting mixtures of 25–30 indigenous forest tree species, with high survival and growth rates, in order to rapidly close the canopy, shade out herbaceous weeds, and ameliorate the understory micro-climate in order to facilitate seedling establishment (Elliott et al., 2003, Lamb et al., 2005, Goosem and Tucker, 2012). After canopy closure, the FSM relies on natural seed-dispersal from nearby remnant trees and forest to increase tree species richness.

The restoration plots used in our study were planted with the same mix of 29 tree species at a density of 3125 ha^{-1} (averaging 1.8 m between trees). Canopy closure on the plots was complete within four years after planting (Anusarnsunthorn and Elliott, 2004, FORRU, 2005) and a dense, two-layered canopy had developed by the start of this study (Wydhayagarn et al., 2009). Each of the three plots was separated from its nearest neighboring replicate plot by at least 0.7 km. The land between the plots included older and younger restoration plantations, small secondary forest patches, and agricultural land. At the time of this investigation, most of the slopes below the watershed ridge were still being cultivated by the villagers of Ban Mae Sa Mai, a Hmong community with a population of about 1700 (Neef et al., 2004). The nearest patch (> 100 ha) of relatively intact mature EGF from the plots was Pah Dong Seng, the community's sacred forest, just behind the village and approximately 1–2 km east of the plots. Additional details regarding the planting, maintenance and monitoring of the plots can be found in Elliott et al. (2003).

We selected the 13-year-old plots because they were the oldest successfully-established restoration plantations in the area. Although FORRU began restoration planting trials a year earlier in 1997, survival of planted trees was mixed in that first year's plantings (Elliott, personal communication). The following year, FORRU adjusted both the composition of planted species and the post-planting fertilization and weeding schedule. This increased seedling survival and within four years the plantings had achieved canopy closure (Anusarnsunthorn and Elliott, 2004, FORRU, 2005). Understory tree seedling communities developed soon after. Seedling surveys conducted across all three restoration plots 8–9 years after establishment identified 369 seedlings belonging to 30 colonizing EGF species within 942 m^2 of sample subplots (Sinhaseni, 2008). The presence of colonizing species indicated that recruitment from seed dispersal into the plantations was occurring. Furthermore, most of the seedlings were zoochorous (82%), though most of the zoochorous colonizers (93%) had smaller-sized seeds with seed lengths ≤ 1 cm (Sangsupan, 2017).

3. Methods

3.1. Species selection

We selected five LSZ tree species characteristic of EGF: *Aglaia lawii, Baccaurea ramiflora, Calophyllum polyanthum, Horsfieldia amygdalina,* and *Mangifera caloneura* (Maxwell and Elliott, 2001). Adults of these species are present in intact, primary EGF in DSNP at low to moderate abundances (Maxwell and Elliott, 2001), but colonizing juveniles of these species had never been identified in the understory of the restoration plots (Sinhaseni, 2008).

Two of the five selected species, *A. lawii* and *H. amygdalina* (formerly known as *Horsfieldia thorelii* Lecomte), were among the species FORRU planted to establish the restoration plots in 1998. Both species, though, grew poorly under initial open field conditions and had only

Table 1
Families and approximate sizes of seed length, width, and mass of planted species.

Species	Family	Length (mm)	Width (mm)	Mass (g)
Aglaia lawii (Wight) Saldanha ex Ramamoorthy[a]	Meliaceae	21.5	13.3	1.89
Baccaurea ramiflora Lour.[b]	Phyllanthaceae	12.6	12.0	0.45
Calophyllum polyanthum Wall ex Planch. & Triana[c]	Guttiferae	22.0	15.0	8.75
Horsfieldia amygdalina (Wall.) Warb.[d]	Myristicaceae	33.4	18.3	4.92
Mangifera caloneura Kurz[d]	Anacardiaceae	45.3	29.7	6.4

[a] Saldanha and Nicholson (1976).
[b] Yu et al. (2008).
[c] Nair et al. (2005).
[d] FORRU (unpublished data).

moderate or poor survival, so they were not recommended for future plantings (Elliott et al., 2003). A few of the initially planted individuals persisted on the plots during the period of this investigation (2011–2013), but no naturally established juveniles of these species were found.

The selected species represent a range of seed sizes, but all five produce seeds with a mass > 0.4 g and a length > 1 cm (Table 1). Thus, based on length, these species possess seeds that are within the top third of seed sizes for all EGF tree species (Sangsupan, 2017).

The seeds used in this investigation originated from mature trees within nearby remaining natural forest. We collected seeds from 2 to 3 trees per species for *A. lawii, B. ramiflora,* and *H. amygdalina* and one tree each for *M. caloneura* and *C. polyanthum. M. caloneura* and *C. polyanthum* fruit irregularly and we were unable to locate additional fruiting trees for this investigation. We collected seeds from four of the five species in early June of 2011, two weeks prior to the initiation of the field experiment. Fruiting of the fifth species, *H. amygdalina,* occurred earlier than the other species, and we acquired seeds of this species three weeks prior to the nursery trial. After collection, we removed fruit flesh from the seeds and rinsed them gently with water. We then packed the seeds in moistened peat moss and stored them at 10–15 °C until sowing, to prevent desiccation and fungal growth.

3.2. Field experiment design

A split-plot design field trial was started on June 22, 2011, about a month into the rainy season, by sowing the seeds of the five selected tree species in combination with four microsite treatments.

On each of the three forest restoration plots, we established two 5 × 4-m subplots, at least 10 m from plot edges and at least 30 m from one another. To prevent seed predation by small mammals, we fenced the perimeter of each subplot with a 1.1 m tall chicken wire fence. A 10-cm chicken wire skirt extended outwards from the base of each fence and we buried the skirts beneath a layer of soil and litter to keep out burrowing rodents.

We further subdivided the subplots into twenty 1 × 1-m split-plots. We then randomly assigned split plots to one of the twenty combinations of species and microsite treatments. In each split-plot 25 seeds of a single species were planted in five rows of five, with each seed at least 15 cm from its nearest neighbor. We applied one of four microsite treatments to each split-plot by placing seeds (i) on top of the pre-existing leaf litter, (ii) on the mineral soil beneath pre-existing leaf litter, (iii) on mineral soil cleared of leaf litter, and (iv) 2-cm beneath mineral soil cleared of leaf litter (i.e. by burying seeds). The depth of pre-existing leaf litter on uncleared split-plots varied from 5 to 10 cm. To bury seeds, we carefully lifted the litter by hand at each planting location, placed the seed on the soil, and replaced the leaf litter. We inserted 15-
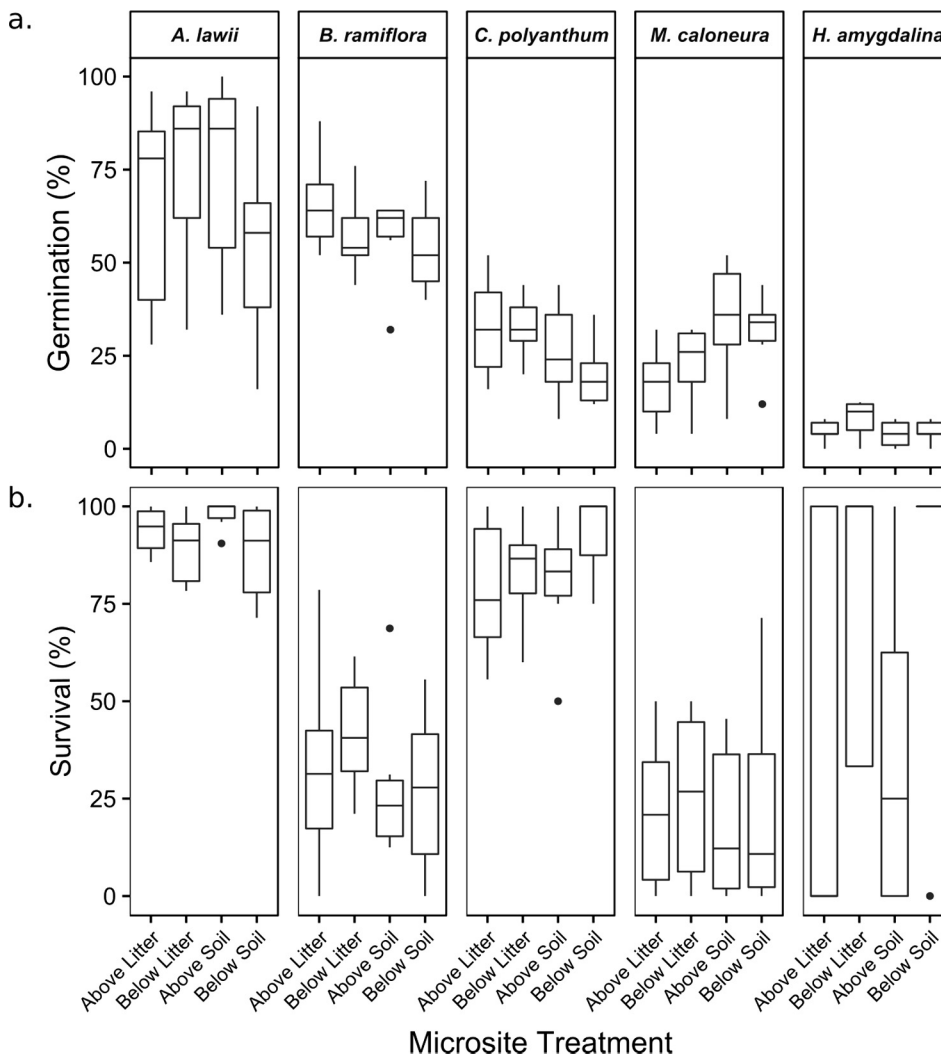
Fig. 1. Box plots displaying (a) the percent germination of planted species by microsite treatment, 4 months after seed sowing and (b) the percent survival of seedlings of planted species by microsite treatment, 26 months after seed sowing. Each box represents the interquartile range (IQR) of the data distribution. The horizontal line across the box represents the distribution median. Boxplot whiskers represent the most extreme data within 1.5 times of the IQR. Outliers outside of 1.5 times the IQR are represented by an individual dot. Note that the boxplots of *H. amygdalina* seedling survival were strongly skewed due to the small number of initial seedlings (28).

cm bamboo skewers into the substrate, beside each seed, to allow us to relocate seeds easily for subsequent monitoring.

We monitored seed germination and seedling survival once a week for the first 10 weeks after planting, then once every three weeks through to week 16. We defined germination as visible emergence of the hypocotyl from the seed or above the substrate. To maintain treatment integrity, at each monitoring event for the first 16 weeks we cleared leaf accumulation from split-plots assigned to the two microsite treatments requiring bare mineral soil, being careful not to disrupt seeds or developing seedlings on the split-plots. By week 16, most new germination had ceased; therefore subsequent monitoring events did not include microsite treatment maintenance. We conducted additional germination and survival monitoring 9, 20, and 26 months after planting.

3.3. Measuring seedling height and diameter

Nine months after seed sowing we selected a subset of surviving seedlings for measurement. We randomly selected eight seedlings for measurement from split-plots with more than eight surviving seedlings and we measured all seedlings from split-plots with eight or fewer seedlings. We tagged each selected seedling by encircling the base of the stem with a labeled, 1-cm-wide aluminum cable tie. After tagging we measured seedling stem diameter 2 cm above the soil and marked the stems with acrylic paint at the measurement location to ensure accurate re-measuring. We also measured stem height from 2 cm above

the soil to the tip of the apical bud. At 20 months after seed sowing (11 months after the initial measurements) we re-measured the height and stem diameter of the tagged seedlings.

3.4. Data analyses

We used three linear mixed models (LMMs) to test the effects of microsite treatment, species and the interaction between microsite treatment and species on seed germination (LMM1), seedling survival (LMM2) and seedling growth (LMM3).

For LMM1, we defined germination as the proportion of planted seeds that had germinated at any time within the first four months of planting, when microsite treatments were still being maintained. For LMM2, we defined survival as the proportion of seedlings alive at the four-month monitoring event that were still alive at the 26-month monitoring event. For LMM3, we defined growth as the height and stem diameter of seedlings 20 months after seed sowing.

At the 9- and 26-month monitoring events, we observed a small number of new *A. lawii* and *C. polyanthum* seedlings, but we excluded these from all LMM analyses because they had emerged after microsite treatments had ceased.

We excluded one species, *H. amygdalina*, from both LMM2 and LMM3 due to insufficient survival data. This species had low germination (approximately 5% of planted seeds germinated) and consequently, 20% of split-plots planted with *H. amygdalina* had no germinating seeds from which to obtain survival proportions or growth

Table 2
Percent germination (Germ) for planted seeds (*n*) in field trials, irrespective of microsite treatments, and the results of FORRU nursery germination trials conducted between 1995 and 1997 (FORRU, unpublished data).

Species	Field		FORRU Nursery	
	n	Germ (%)	*n*	Germ (%)
A. lawii	598	68.6	72	55.6
B. ramiflora	600	58.5	72	51.4
C. polyanthum	600	30.3	72	31.9
M. caloneura	600	26.5	72	70.8
H. amygdalina	599	5.3	72	94.4

measurements. In addition, in LMM2, the number of surviving seedlings varied widely among the four included species. LMM2 therefore weighted each split-plot's survival proportions by the number of seedlings on the split-plot that were alive at the 4-month monitoring event, to compensate for bias from survival proportions derived from small numbers of seedlings.

To account for the nested structure of the experimental design, we included both plot and subplot as random effects in all LMMs. In addition, we logit transformed germination and survival proportions and log transformed height measurements, to stabilize variance and normalize the data. For each model, we calculated the summary statistic R^2_{GLMM}, as described in Nakagawa and Schielzeth (2013), to quantify the amount of variance explained by the model, as well as to provide an absolute value describing the model's goodness-of-fit. We conducted all analyses in the R 3.0.2 software environment (R Development Core Team, 2013).

4. Results

4.1. Seed germination

In the field, the mean germination percentage for all split-plots (*n* = 120) was 37.0% (sd = 27.4%). Mean germination percentages across split-plots by species (*n* = 24 for each species) ranged from a minimum of 5.4% (sd = 3.9%) for *H. amygdalina* to a maximum of 68.6% (sd = 27.2%) for *A. lawii* (Fig. 1a).

Microsite treatments did not significantly affect overall germination ($F_{3,92} = 1.11$, *p* = 0.35), nor was there a difference among species in response to treatment ($F_{12,92} = 1.31$, *p* = 0.23). Species germination

rates (irrespective of treatments), however, were significantly different from one another ($F_{4,92} = 70.7$, *p* < 0.0001; Fig. 1a). The statistical model explained 70% of the variance in germination ($R^2_{GLMM} = 0.70$).

Since treatment had no significant effect on rates of seed germination, we compared seed germination rates by species to germination rates obtained in nursery germination tests previously conducted by FORRU as part of their efforts to identify species suitable for restoration planting (Table 2). For three of the five tested species, field germination rates in this investigation were similar to or greater than those reported in FORRU germination tests. *H. amygdalina* and *M. caloneura*, however, had considerably lower germination rates in the field experiment than in the FORRU germination tests.

4.2. Seedling survival

The mean 26-month survival of seedlings across all split-plots (*n* = 120) was 58.5% (sd = 36.8%). Mean survival across split-plots by species (*n* = 24) ranged from a minimum of 23.0% (sd = 46.6%) for *M. caloneura* to a maximum of 92.4% (sd = 9.1%) for *A. lawii* (Fig. 1b).

Microsite treatments did not significantly affect overall seedling survival ($F_{3,75} = 0.74$, *p* = 0.53), nor did treatments affect seedling survival differently among species ($F_{9,75} = 0.85$, *p* = 0.58). Seedling survival irrespective of treatment, however, was significantly different among species ($F_{3,75} = 15.97$, *p* < 0.0001; Fig. 1b). The statistical model explained 18% of the variance in survival ($R^2_{GLMM} = 0.18$).

4.3. Seedling height and diameter

Height and diameter growth differed among species, between 9 and 20 months after seed sowing (Fig. 2). *C. polyanthum* seedlings grew the most rapidly in height, attaining a final height nearly twice that of the next tallest species, *A. lawii*. *A. lawii, H. amygdalina,* and *M. caloneura* had similar growth rates and heights, with growth occurring most rapidly within the first 9 months after seed sowing. *B. ramiflora* seedlings grew more slowly than seedlings of the other four species and appeared to develop more slowly as well. At the conclusion of the investigation at 26 months, many *B. ramiflora* seedlings still possessed their cotyledonous leaves and a few had only just developed their first true leaves.

We calculated relative growth rate of height and diameter for tested species, but this provided no additional insight into growth patterns beyond that which could be determined from the visual comparison provided by Fig. 2.
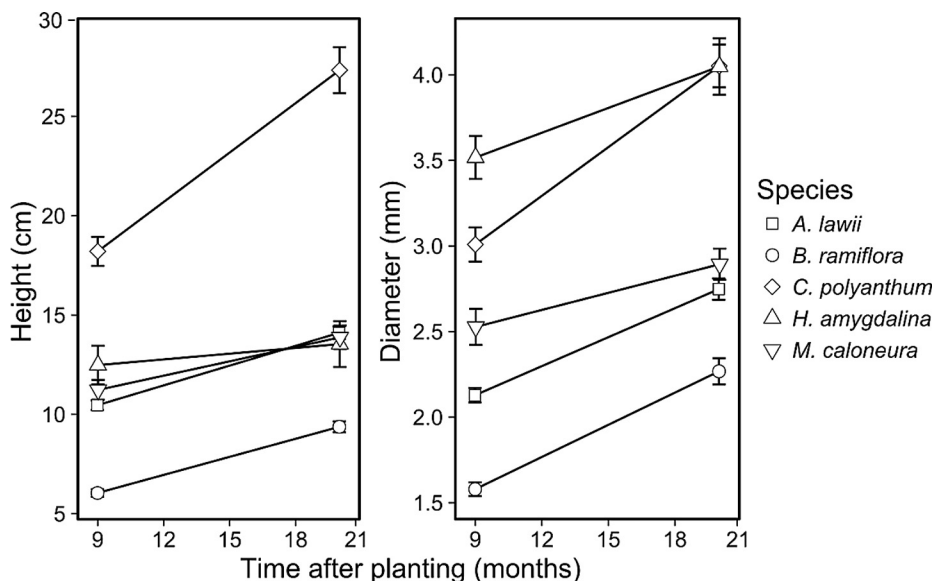

Fig. 2. Height and diameter growth between 9 and 20 months after seed sowing with standard deviation bars.
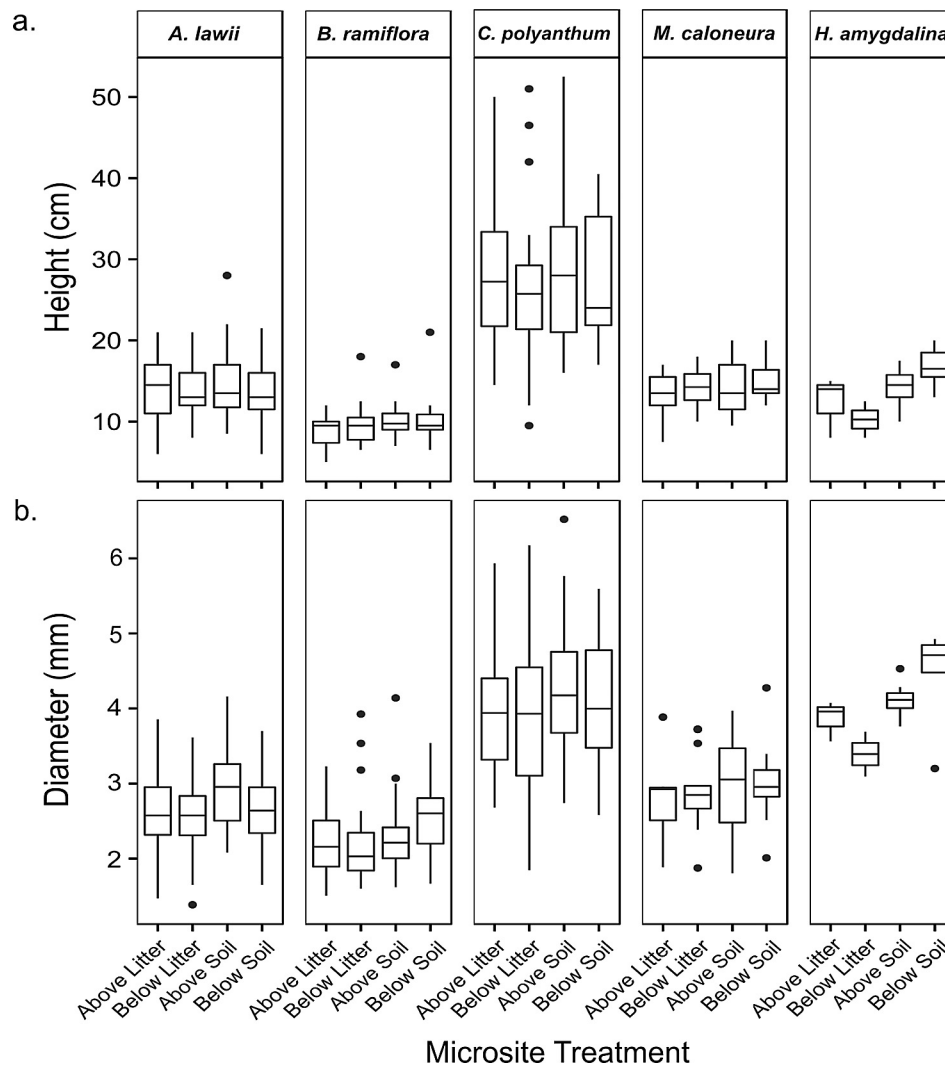
Fig. 3. Box plots displaying (a) height (cm) of surviving seedlings of planted species by microsite treatment, 20 months after seed sowing and (b) the diameter (mm) of surviving seedlings of planted species by microsite treatment, 20 months after seed sowing. See Fig. 1 for a description of the box plot display of distributions.

Twenty months after seed sowing, the mean height averaged across all measured seedlings ($n = 400$) was 17.2 cm (sd = 8.8) and the mean stem diameter was 3.1 mm (sd = 1.0). Mean height by species ranged from a minimum of 9.7 cm (sd = 2.5) for *B. ramiflora* to a maximum of 27.6 cm (sd = 8.7) for *C. polyanthum*. Mean stem diameter by species ranged from a minimum of 1.6 mm (sd = 0.3) for *B. ramiflora* to a maximum of 3.0 mm (sd = 0.6) for *C. polyanthum*.

Twenty months after seed sowing, microsite treatments did not significantly affect seedling height ($F_{3,362} = 0.23$, $p = 0.88$), nor did the treatments affect the height of species differently ($F_{9,362} = 0.51$, $p = 0.87$). However, the mean seedling heights of each species (irrespective of treatments) did differ significantly ($F_{3,362} = 81.39$, $p < 0.0001$). By 20 months, *C. polyanthum* seedlings had grown nearly three times taller than *B. ramiflora* seedlings and were about twice as tall as *A. lawii, M. caloneura,* and *H. amygdalina* seedlings (Fig. 2). The statistical model explained 97% of the variance in height measurements ($R^2_{GLMM} = 0.971$).

Microsite treatments also did not significantly affect overall seedling stem diameter ($F_{3,362} = 2.22$, $p = 0.085$) and there was no evidence that the treatments affected the stem diameter of species differently ($F_{9,362} = 0.26$, $p = 0.98$). However, differences in mean stem diameter among species (irrespective of treatments) were significant ($F_{3,362} = 40.86$, $p < 0.0001$) (Fig. 3b). The statistical model explained 90% of the variance in diameter measurements ($R^2_{GLMM} = 0.902$).

4.4. Patterns in germination and survival

Germination of all species peaked at or before the fourth month after planting (Fig. 4). After the peak, two distinct patterns in seedling survival emerged. Survival of *A. lawii* and *C. polyanthum* seedlings
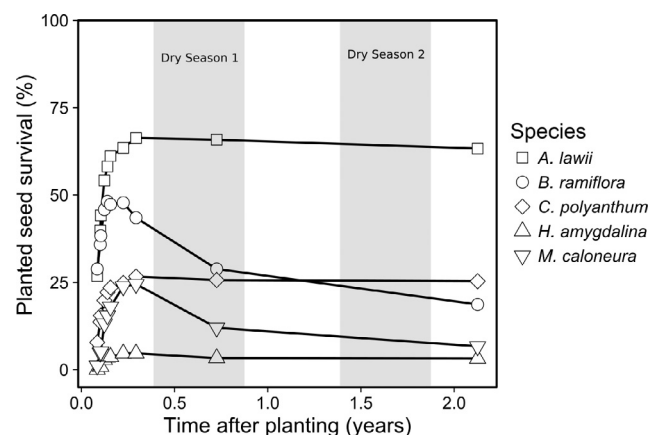

Fig. 4. Percentage survival of sown seeds (i.e. the percentage of sown seeds that were present as seedlings) in the forest restoration plantations.

remained stable throughout the study, but *B. ramiflora* and *M. caloneura* survival declined rapidly between months 4 and 9, a range of time that spanned most of the first dry season after seed sowing. *M. caloneura's* decline appears to correspond with the onset of the first dry season, but *B. ramiflora* survival began to decline shortly before the beginning of the season. Between months 9 and 26, *M. caloneura* and *B. ramiflora* survival continued to decline, though more slowly. This period of time included the second dry season, but there was no survival data immediately prior to the second dry season to assist us in determining whether *M. caloneura* and *B. ramiflora* seedling deaths in the second were a result of continuous mortality or mortality primarily due to the dry season. The survival of *H. amygdalina* seedlings appeared to be relatively stable between months 4 and 26, but trends in this species' survival may have been obscured by the small number of *H. amygdalina* seedlings (28 overall).

5. Discussion

5.1. General overview

The absence of statistically significant treatment effects on germination and seedling survival in the field experiment indicated that recruitment of the five LSZ tree species was not limited by the microsite environments tested. Moreover, relatively high overall germination (37%) and 26-month seedling survival (58.5%), irrespective of microsite treatment, suggests that the basic microsite requirements for germination and early survival for these species were generally met in the 13-year-old restoration plantations. Over the first 20 months of the investigation, surviving seedlings of all five species grew well in both height and diameter, indicating that conditions were sufficient for seedling growth as well as persistence. Furthermore, initial microsite conditions were not correlated with seedling height or diameter at 20 months. This suggests that the investigated germination microsites have no effect on the robustness of seedlings.

Taken together, the results of this investigation support the hypothesis that seedlings of the tested LSZ tree species are slow to colonize tropical forest restoration plantations because of seed limitations resulting from inadequate seed dispersal or low seed availability rather than understory microsite conditions. This also suggests that seed limitations may similarly delay or prevent the recruitment of other LSZ species in the restoration plantations. Our results are consistent with those of other seed sowing studies that have found that seed availability is a key limitation to tree recruitment in both primary and secondary tropical forest (Makana and Thomas, 2004, Svenning and Wright, 2005, Vargas and Stevenson, 2013).

The germination and survival percentages in this investigation were comparable to those reported for LSZ trees sown in similarly-aged, early successional seasonal and moist tropical forest (Bonilla-Moheno and Holl, 2009, Cole et al., 2010). This provides evidence that after a little more than a decade, the understory conditions for seedling regeneration in the forest restoration plantations have become similar to those in naturally regenerating secondary forests. This investigation therefore adds to the growing body of evidence that restoration plantings rapidly ameliorate environmental barriers that inhibit natural forest regeneration (Cole et al., 2010, Bertacchi et al., 2016).

5.2. Seed viability

Since seed viability may interact with seed availability to limit seedling recruitment (Clark et al., 1999), we attempted to isolate viability as a potential factor in recruitment in the field experiment by using a nursery germination experiment to estimate seed germination rates for a subset of the seeds we harvested. However, we were unable to use the nursery trial to estimate seed viability because rates of germination in the nursery were less than half of field germination rates for all but one species, most likely due to overwatering and subsequent

fungal infection (Table A1). Instead, we made some observations based on comparisons between field germination in this investigation and nursery germination tests that FORRU had previously conducted as part of their efforts to identify species suitable for restoration planting (Table 2). Although comparisons of seeds from different harvests is not ideal, similar germination rates for *A. lawii, B. ramiflora,* and *C. polyanthum* in both our field experiment and FORRU's nursery germination tests suggest that field conditions had little effect on germination of these species. However, *C. polyanthum,* had relatively low germination in both our field experiment and FORRU's nursery tests (< 35%). This suggests that lower seed viability may be a characteristic of this species and may act as an additional limitation to natural seed recruitment. Once germinated, though, *C. polyanthum* seedlings had both high survival and growth (Figs. 1b and 2). Germination rates of two other species, *H. amygdalina* and *M. caloneura*, were considerably lower in our field experiment than in FORRU's germination tests. This suggests that either field conditions reduced germination rates or the seeds used in our field experiment were damaged prior to planting. Damage may have occurred during storage or, in the case of *H. amygdalina*, due to late seed harvesting. Future experiments should determine the degree to which seed viability limits recruitment in forest restoration plantations by modifying collection, storage, and watering procedures to minimize impacts to seed survival.

5.3. Microsite effects

5.3.1. Leaf litter

In our experiment, leaf litter had no detectable effect on seed germination and 26-month seedling survival and growth. This contrasts with the results of several authors, who showed that leaf litter reduces establishment of some tropical trees, due to light interception and because it acts as a barrier to both seedling radicles and seedling emergence (Putz, 1983, Vazquez-Yanes and Orozco-Segovia, 1992, Dalling and Hubbell, 2002). Many of these published results though were for small-seeded pioneers. In our experiment, nearly all seeds placed on the leaf litter surface eventually worked their way down through the litter to the soil surface, so leaf litter did not present a mechanical barrier. Large seeds appear to be well-adapted to emergence from burial beneath leaf litter and soil (Kitajima, 2000), since they retain large energy reserves which hasten their growth above competing vegetation and facilitate seedling persistence in deep shade (Leishman et al., 2000). Thus, large seeds typically have higher rates of germination and seedling survival than smaller seeds in tropical forest understories (Molofsky and Augspurger, 1992, Dalling and Hubbell, 2002). Investigations that included late-successional species (with seed sizes comparable to those used in this experiment) also reported a small positive effect or no effect of litter on germination and seedling survival in forest understory (Molofsky and Augspurger, 1992, Makana and Thomas, 2005, Dupuy and Chazdon, 2008). Furthermore, Makana and Thomas (2005) reported that, similar to the seedlings in our study, the presence or absence of litter has little effect on the growth of large-seeded species in the understory. The results of this investigation therefore contribute to the growing body of research indicating that, while leaf litter may strongly influence the establishment of small-seeded species, it is neither an impediment nor a requirement for the establishment of LSZ trees in the understory.

5.3.2. Soil

Although we found that burial of seeds in soil had no discernable effect on germination and survival, several studies have found that burial beneath soil strongly increases tropical tree recruitment, particularly for large-seeded species (Hardwick et al., 1997, Woods and Elliott, 2004, Doust et al., 2006). However, these studies have been conducted in abandoned agricultural fields, where climatic conditions are likely to be harsher and seed predators more common. Under these conditions, burial may enhance recruitment by maintaining the

temperature and humidity at levels required for germination and preventing loss of recalcitrant seeds due to desiccation. In the restoration plantations the canopy cover may be sufficient to maintain adequate temperature and humidity for germination without burial. In addition, 2011, the year this investigation was initiated, was an exceptionally wet year throughout Thailand, with the annual rainfall reaching 24% above normal (Thai Meteorological Department, 2012). High inter-annual variability in timing and quantity of rainfall is characteristic for the seasonal tropics (Murphy and Lugo, 1986) and seedling establishment may be more sensitive to microsite environments in drier years. The importance of microsites may also increase during infrequent dry spells during the wet season. Dry spells as brief as four days have been shown to increase seed and seedling mortality of small-seeded pioneer tree species in Panama (Engelbrecht et al., 2006).

Increased recruitment of buried seeds may also be attributed to protection from seed predators (Cintra, 1997, Brewer and Molly, 2001). In this investigation, however, we attempted to limit the test to the effects of abiotic microsite factors alone on recruitment by erecting fences to exclude small mammalian seed predators. Although we did not take steps to prevent bird and insect seed predation, we also did not observe evidence of either form of predation. There is some evidence that forest restoration reduces the habitat suitability for mammalian seed predators, at least initially. A small mammal survey of the plantation plots in this study as well as other nearby plots found substantially fewer rodent seed predators two and four years after planting than on non-planted control plots (Thaiying, 2003). We were unable to determine if mammalian seed predator populations remained depressed during this investigation since more recent mammal surveys are not available. While it was not possible for us to quantify potential seed predation outside of fenced exclosures, the high density and species richness of naturally-recruited seedlings (1.1 seedlings m^{-1}, 58 species) observed during the understory seedling survey of the forest restoration plots suggests that recruitment limitation due to seed predation in the forest is low (Sinhaseni, 2008). Among the seedlings surveyed were the offspring of a few large-seeded species that had been initially planted to establish the forest restoration plots. One Fagaceous species in particular, *Castanopsis calathiformis*, had begun to form dense seedling stands in the areas immediately surrounding the parent trees. During both years of our investigation this species produced sizable crops of large acorns and we observed that germination of acorns contributed to the expansion of the seedling stands. Since Fagaceous acorns are frequently predated by small mammals in tropical Asia (Wada, 1993, Chang et al., 2009), the presence and expansion of the *C. calathiformis* seedling stands provides additional evidence to suggest that seed predation in the restoration plantations was low.

5.3.3. Seasonal effects

Several studies have suggested that, in seasonal forests, drought acts as a filter to tree seedling survival during the first dry season after germination (Lieberman and Li, 1992, Gerhardt, 1996, McLaren and McDonald, 2003). In our investigation, three of the tested species (*B. ramiflora*, *M. caloneura*, and *H. amygdalina*) experienced substantial declines in survival (≥28.5%) over the course of the first dry season. *H. amygdalina* had very few seedlings due to low germination; therefore, patterns in survival may be obscured by the small sample size. High mortality of *M. caloneura* during the first dry season, though, corresponds with Marod et al. (2002)'s investigation of seedling dynamics in a Thai mixed deciduous forest. In that study, all of the naturally-recruited first-year *M. caloneura* seedlings died as a consequence of dry-season drought. Survival of *B. ramiflora* seedlings contrasted with *M. caloneura* in that *B. ramiflora* seedling survival began to decline a month prior to the start of the first dry season (Fig. 4). This suggests that factors in addition to drought may have contributed to reducing early survival of *B. ramiflora* seedlings. One potential factor may have been insect predation, which we frequently observed on young *B. ramiflora* leaves. Both *B. ramiflora* and *M. caloneura* survival continued to decline

after the first dry season, but there were too few monitoring events to determine whether the declines were continuous or corresponded with the second dry season. If the declines were continuous, this would suggest that other factors such as low light availability may be contributing to the filtering of seedlings from the understory. In contrast to *B. ramiflora* and *M. caloneura*, survival of the *A. lawii* and *C. polyanthum* remained stable over the entire course of the investigation, declining by ≤14.4% between four- and 26-months after sowing (Fig. 4). Low mortality over two dry seasons provides strong evidence of high drought tolerance for seedlings of these species and suggests that, while seasonal drought may filter some LSZ tree species from the forest restoration plantations, drought tolerance may also be fairly common. Consequently, the impact of seasonal drought on seedling composition in the plantations may be less important than the effects of factors such as limited seed dispersal for many species.

5.4. Implications for tropical forest restoration

By providing evidence to suggest that seed availability is more limiting than establishment microsites to the colonization of forest restoration plantings by late-successional LSZ trees, this investigation underscores the importance of seed limitation in the restoration of the tree community. Given that sources of seed limitations (e.g. distant seed sources, low seed production, and poor animal-dispersal) are unlikely to decrease in the foreseeable future, active intervention will be required to ensure that LSZ tree species are represented in the future forest. Enrichment planting, the interplanting of tree species into the existing forest, may augment forest species diversity by introducing absent species. Enrichment planting is most often carried out using nursery grown seedlings and saplings (Lamb et al., 2005); however, raising seedlings in a nursery is costly, labor intensive, and limits planting to just those species that are suitable for nursery propagation (Engel and Parrotta, 2001, Zahawi and Holl, 2009). Direct seeding (i.e. sowing seeds directly into restoration habitats), on the other hand, can be a more efficient and cost-effective means of restoration and enrichment planting (Cole et al., 2010, Meli et al., 2017). Although we did not explicitly focus on testing the viability of direct seed enrichment planting, successful seedling establishment from seeds sown in our investigation supports Cole et al.'s (2010) finding that direct-seed sowing is an effective means of introducing LSZ tree species into young tropical forest restoration plantations. The results of our experiment also contribute to a growing body of research indicating that large-seeded, late-successional trees are well-suited for direct-seeding under a range of early successional conditions (Hardwick et al., 1997, Camargo et al., 2002, Hooper et al., 2002, Bonilla-Moheno and Holl, 2009, de Souza and Engel, 2018).

This investigation also has potential implications for the selection of direct seeding methods. Although studies on deforested sites have reported increased recruitment following burial of large seeds (Hardwick et al., 1997, Doust et al., 2006); this study found that burial beneath soil or leaves did not significantly affect seed germination or the survival and growth of seedlings in the understory of 13-year-old forest restoration plantations. This suggests that direct seeding by broadcast sowing of seeds onto the leaf litter in closed-canopy plantations may be just as effective as seed burial, while also being considerably easier and less expensive to implement. By simplifying seed planting, broadcast sowing may also speed up the process of planting and reduce the time seeds spend in storage. Given the challenges of storing recalcitrant LSZ seeds (Pammenter and Berjak, 2013), seeds should be planted as soon as possible after harvesting to minimize seed damage and loss of viability.

As previously discussed, there was unusually high precipitation in the first year of this investigation and our seeds were protected from some forms of seed predation by a fence. Therefore, prior to the use of broadcast sowing in large-scale enrichment planting, additional research should first clarify the relationship between seedbed microsites

and factors such as variable annual precipitation and seed predation, prior to the use of broadcast sowing in large-scale enrichment planting.

Although the species tested in our investigation were all putatively shade-tolerant, mature-forest species, they manifested a wide range of germination, survival, and growth responses in the restoration plantation understory. These differences underscore the need for additional seed sowing experiments, to test the suitability of desired species for direct seed enrichment planting. They also suggest the need for an objective means of comparing species suitability to facilitate species selection by forest managers. Tunjai and Elliott (2012) proposed one such method that scores species based on a combination of establishment (percent survival of seeds at one year after sowing) and height growth and then ranks those scores relative to other species. When we applied a version of this method to the results of our investigation, *C. polyanthum* and *A. lawii* were the highest ranked of the five tested species. This suggests that of the five tested species, these two species are most suitable for direct seed enrichment planting *if* the primary objective of planting is maximizing seedling establishment and growth. Enrichment planting in the restoration setting, however, may have additional objectives such as increasing diversity and habitat value and conserving rare and endangered species. Future ranking methods should incorporate factors related to these objectives as well as efficient establishment and growth to ensure that the resulting rankings accurately represent the value of species towards meeting all enrichment planting goals.

Final seedling heights in our investigation were similar to those reported by Cole et al. (2010) for two-year old, late successional, LSZ tree seedlings sown into 8–10-year-old rainforest restoration plantings, but our results further indicated that most seedling growth occurred within the first nine months after sowing. With the exception of *C. polyanthum*, growth of all seedlings slowed substantially between 9 and 20 months after sowing, increasing by ≤ 3 cm in height. Our results and those of Cole et al. (2010) suggest that slow growth may be characteristic of seedlings established by direct seeding in the understory of restoration plantings. This further suggests that seedling recruitment alone may be insufficient to ensure timely maturation of desirable LSZ species in the forest restoration plantations. Research that pairs direct seeding with additional management interventions such as gap formation or seasonal watering may accelerate the rate of maturation. Moreover, similar treatments may also facilitate the recruitment and establishment of species that had relatively poor establishment in this investigation, thereby increasing their suitability for direct-seed enrichment planting.

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Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.foreco.2018.03.021.

References

Aide, T.M., Zimmerman, J.K., Pascarella, J.B., Rivera, L., Marcano-Vega, H., 2000. Forest regeneration in a chronosequence of tropical abandoned pastures: implications for restoration ecology. Restor. Ecol. 8, 328–338.

Anusarnsunthorn, V., Elliott, S., 2004. Long-term Monitoring of Biodiversity Recovery in Forest Restoration Plots in Northern Thailand (BRT 344004). Forest Restoration Research Unit, Chiang Mai University, Chiang Mai, Thailand.

Augspurger, C.K., 1984. Seedling survival of tropical tree species: interactions of dispersal distance, light-gaps, and pathogens. Ecology 65, 1705–1712.

Bertacchi, M.I.F., Amazonas, N.T., Brancalion, P.H.S., Brondani, G.E., de Oliveira, A.C.S., de Pascoa, M.A.R., Rodrigues, R.R., 2016. Establishment of tree seedlings in the understory of restoration plantations: natural regeneration and enrichment plantings. Restor. Ecol. 24, 100–108.

Blain, D., Kellman, M., 1991. The effect of water supply on tree seed germination and seedling survival in a tropical seasonal forest in Veracruz, Mexico. J. Trop. Ecol. 7, 69–83.

Bonilla-Moheno, M., Holl, K.D., 2009. Direct seeding to restore tropical mature-forest species in areas of slash-and-burn agriculture. Restor. Ecol. 18, 438–445.

Brearley, F.Q., Prajadinata, S., Kidd, P.S., Proctor, J., Suriantata, 2004. Structure and floristics of an old secondary rain forest in Central Kalimantan, Indonesia, and a comparison with adjacent primary forest. For. Ecol. Manage. 195, 385–397.

Brewer, S.W., Molly, A.H.W., 2001. Ignorant seed predators and factors affecting the seed survival of a tropical palm. Oikos 93, 32–41.

Brodie, J.F., Helmy, O.E., Brockelman, W.Y., Maron, J.L., 2009. Bushmeat poaching reduces the seed dispersal and population growth rate of a mammal-dispersed tree. Ecol. Appl. 19, 854–863.

Brown, N., 1996. A gradient of seedling growth from the centre of a tropical rain forest canopy gap. For. Ecol. Manage. 82, 239–244.

Camargo, J.L.C., Ferraz, I.D.K., Imakawa, A.M., 2002. Rehabilitation of degraded areas of Central Amazonia using direct sowing of forest tree seeds. Restor. Ecol. 10, 636–644.

Caughlin, T.T., Ferfuson, J.M., Lichstein, J.W., Zuidema, P.A., Bunyavejchewin, S., Levey, D.J., 2014. Loss of animal seed dispersal increases extinction risk in a tropical tree species due to pervasive negative density dependence across life stages. Proc. R. Soc. B: Biol. Sci. 282, 20142095.

Chang, G., Xiao, Z., Zhang, Z., 2009. Hoarding decisions by Edward's long-tailed rats (*Leopoldamys edwardsi*) and South China field mice (*Apodemus draco*): the responses to seed size and germination schedule in acorns. Behav. Process. 82, 7–11.

Chazdon, R.L., 2003. Tropical forest recovery: Legacies of human impact and natural disturbances. Perspect. Plant Ecol. Evolut. Syst. 6, 51–71.

Chazdon, R.L., 2014. Second Growth: The Promise of Tropical Forest Regeneration in an Age of Deforestation. The University of Chicago Press, Chicago.

Chua, S.C., Ramage, B.S., Ngo, K.M., Potts, M.D., Lum, S.K.Y., 2013. Slow recovery of a secondary tropical forest in Southeast Asia. For. Ecol. Manage. 308, 153–160.

Cintra, R., 1997. Leaf litter effects on seed and seedling predation of the palm *Astrocaryum murumuru* and the legume tree *Dipteryx micrantha* in Amazonian forest. J. Trop. Ecol. 13, 709–725.

Clark, J.S., Beckage, B., Camill, P., Cleveland, B., Hille Ris Lambers, J., Lichter, J., McLachlan, J., Mohan, J., Wyckoff, P., 1999. Interpreting recruitment limitation in forests. Am. J. Bot. 86, 1–16.

Cole, R.J., Holl, K.D., Keene, C.L., Zahawi, R.A., 2010. Direct seeding of late-successional trees to restore tropical montane forest. For. Ecol. Manage. 261, 1590–1597.

Corlett, R.T., 2017. Frugivory and seed dispersal by vertebrates in tropical and subtropical Asia: an update. Global Ecol. Conserv. 11, 1–22.

Dalling, J.W., Hubbell, S.P., 2002. Seed size, growth rate and gap microsite conditions as determinants of recruitment success of pioneer species. J. Ecol. 90, 557–568.

Daws, M.I., Garwood, N., Pritchard, H.W., 2006. Prediction of desiccation sensitivity in seeds of woody species: a probabilistic model based on two seed traits and 104 species. Ann. Bot. 97, 667–674.

Daws, M.I., Garwood, N.C., Pritchard, H.W., 2005. Traits of recalcitrant seeds in a semideciduous tropical forest in Panamá: some ecological implications. Funct. Ecol. 19, 874–885.

de Souza, D.C., Engel, V.L., 2018. Direct seeding reduces costs, but it is not promising for restoring tropical seasonal forests. Ecol. Eng. 116, 35–44.

de Souza, F.M., Batista, J.L.F., 2004. Restoration of seasonal semideciduous forests in Brazil: influence of age and restoration design on forest structure. For. Ecol. Manage. 191, 185–200.

Dirzo, R., Young, H.S., Mooney, H.A., Ceballos, G. (Eds.), 2011. Seasonally Dry Tropical Forests: Ecology and Conservation. Island Press, Washington, DC.

Doust, S.J., Erskine, P.D., Lamb, D., 2006. Direct seeding to restore rainforest species: microsite effects on the early establishment and growth of rainforest tree seedlings on degraded land in the wet tropics of Australia. For. Ecol. Manage. 234, 333–343.

Dupuy, J.M., Chazdon, R.L., 2008. Interacting effects of canopy gap, understory vegetation and leaf litter on tree seedling recruitment and composition in tropical secondary forest. For. Ecol. Manage. 255, 3716–3725.

Elliott, S., Blakesley, D., Hardwick, K., 2013. Restoring Tropical Forests: A Practical Guide. Royal Botanic Gardens, Kew.

Elliott, S., Navakitbumrung, P., Kuarak, C., Zangkum, S., Anusarnsunthorn, V., Blakesley, D., 2003. Selecting framework tree species for restoring seasonally dry tropical forests in northern Thailand based on field performance. For. Ecol. Manage. 184, 177–191.

Elliott, S., Navakitbumrung, P., Zangkum, S., Kuarak, C., Kerby, J., Blakesley, D., Anusarnsunthorn, V., 2000. Performance of six native tree species, planted to restore degraded forestland in northern Thailand and their response to fertiliser. In: Elliott, S., Kerby, J., Blakesley, D., Hardwick, K., Woods, K., Anusarnsunthorn, V. (Eds.),

Forest Restoration for Wildlife Conservation. International Tropical Timber Organisation and The Forest Restoration Research Unit, Chiang Mai University, Thailand, pp. 244–255.

Elliott, S., Promkutkaew, S., Maxwell, J.F., 1994. Flowering and seed production phenology of dry tropical forest trees in northern Thailand. In: International Symposium on Genetic Conservation and Production of Tropical Forest Tree Seed. ASEAN-Canada Forest Tree Seed Centre, Saraburi, Thailand, pp. 52–61.

Engel, V.L., Parrotta, J.A., 2001. An evaluation of direct seeding for reforestation of degraded lands in central São Paulo state, Brazil. For. Ecol. Manage. 152, 169–181.

Engelbrecht, B.J.M., Dalling, J.W., Pearson, T.H.R., Wolf, R.L., Gálvez, D.A., Koehler, T., Tyree, M.T., Kursar, T.A., 2006. Short dry spells in the wet season increase mortality of tropical pioneer seedlings. Oecologia 148, 258–269.

Ferreira, W.N., de Lacerda, C.F., da Costa, R.C., Filho, S.M., 2015. Effect of water stress on seedling growth in two species with different abundances: the importance of Stress Resistance Syndrome in seasonally dry tropical forest. Acta Botanica Brasilica 29, 375–382.

FORRU, 2005. How to Plant a Forest: The Principles and Practice of Restoring Tropical Forests. Forest Restoration Research Unit, Biology Department, Science Faculty, Chiang Mai University, Chiang Mai.

Gerhardt, K., 1996. Effects of root competition and canopy openness on survival and growth of tree seedlings in a tropical seasonal dry forest. For. Ecol. Manage. 82, 33–48.

Goosem, S.P., Tucker, N., 2012. Repairing the Rainforest - Theory and Practice of Rainforest Re-establishment in North Queensland's Wet Tropics, second ed. Wet Tropics Management Authority, Cairns, Australia.

Hardwick, K., Healey, J., Elliott, S., Garwood, N., Anusarnsunthorn, V., 1997. Understanding and assisting natural regeneration processes in degraded seasonal evergreen forests in northern Thailand. For. Ecol. Manage. 99, 203–214.

Harper, J.L., 1977. Population Biology of Plants. Academic Press, New York, New York.

Hooper, E., Condit, R., Legendre, P., 2002. Responses of 20 native tree species to reforestation strategies for abandoned farmland in Panama. Ecol. Appl. 12, 1626–1641.

Kitajima, K., 2000. Ecology of seedling regeneration. In: Fenner, M. (Ed.), Ecology of Seedling Regeneration. CABI Publishing, Wallingford, UK, pp. 331–359.

Kitamura, S., Yumoto, T., Poonswad, P., Chuailua, P., Plongmai, K., Maruhashi, T., Noma, N., 2002. Interactions between fleshy fruits and frugivores in a tropical seasonal forest in Thailand. Oecologia 133, 559–572.

Lamb, D., 2011. Regreening the Bare Hills: Tropical Forest Restoration in the Asia-Pacific Region. Springer, Dordrecht, Netherlands.

Lamb, D., Erskine, P.D., Parrotta, J.A., 2005. Restoration of degraded tropical forest landscapes. Science 310, 1628–1632.

Leishman, M.R., Wright, I.J., Moles, A.T., Westoby, M., 2000. The evolutionary ecology of seed size. In: Fenner, M. (Ed.), Seeds: the Ecology of Regeneration in Plant Communities. pp. 31–57.

Lieberman, D., Li, M., 1992. Seedling recruitment patterns in a tropical dry forest in Ghana. J. Veg. Sci. 3, 375–382.

Makana, J.R., Thomas, S.C., 2004. Dispersal limits natural recruitment of African mahoganies. Oikos 106, 67–72.

Makana, J.R., Thomas, S.C., 2005. Effects of light gaps and litter removal on the seedling performance of six African timber species. Biotropica 37, 227–237.

Marod, D., Kutintara, U., Tanaka, H., Nakashizuka, T., 2002. The effects of drought and fire on seed and seedling dynamics in a tropical seasonal forest in Thailand. Plant Ecol. 161, 41–57.

Maxwell, J.F., Elliott, S., 2001. Vegetation and Vascular Flora of Doi Sutep-Pui National Park, Northern Thailand. The Biodiversity Research and Training Program, Bangkok.

McLaren, K.P., McDonald, M.A., 2003. The effects of moisture and shade on seed germination and seedling survival in a tropical dry forest in Jamaica. For. Ecol. Manage. 183, 61–75.

Meli, P., Isernhagen, I., Brancalion, P.H.S., Isernhagen, E.C.C., Behling, M., Rodrigues, R.R., 2017. Optimizing seeding density of fast-growing native trees for restoring the Brazilian Atlantic Forest. Restor. Ecol. 26, 212–219.

Molofsky, J., Augspurger, C.K., 1992. The effect of leaf litter on early seedling establishment in a tropical forest. Ecology 73, 68–77.

Murphy, P.G., Lugo, A.E., 1986. Ecology of tropical dry forest. Annu. Rev. Ecol. Syst. 17, 67–88.

Nair, K.K.N., Mohanan, C., Matthew, G., 2005. Plantation technology for selected indigenous trees in the Indian peninsula. Bois et Forets des Tropiques 285, 17–23.

Neef, A., Chamsai, L., Hammer, M., Wannitpradit, A., Sangkapitux, C., Xyooj, Y., Sirisupluxuna, P., Spreer, W., 2004. Water tenure in highland watersheds of northern Thailand: tragedy of the commons or successful management of complexity? In: Gerold, G., Fremerey, M., Guhardja, E. (Eds.), Land Use, Nature Conservation and the

Stability of Rainforest Margins in Southeast Asia. Springer, Berlin, Heidelberg, pp. 367–390.

Osuri, A.M., Sankaran, M., 2016. Seed size predicts community composition and carbon storage potential of tree communities in rain forest fragments in India's Western Ghats. J. Appl. Ecol. 53, 837–845.

Pammenter, N.W., Berjak, P., 2013. Physiology of desiccation-sensitive (recalcitrant) seeds and the implications for cryopreservation. Int. J. Plant Sci. 175, 21–28.

Peterson, C.J., Facelli, J.M., 1992. Contrasting germination and seedling growth of *Betula allghaniensis* and *Rhus typhina* subjected to various amounts and types of plant litter. Am. J. Bot. 79, 1209–1216.

Poorter, L., Markesteijn, L., 2008. Seedling traits determine drought tolerance of tropical tree species. Biotropica 40, 321–331.

Pritchard, H.W., Daws, M.I., Fletcher, B.J., Gaméné, C.S., Msanga, H.P., Omondi, W., 2004. Ecological correlates of seed desiccation tolerance in tropical African dryland trees. Am. J. Bot. 91, 863–870.

Putz, F.E., 1983. Treefall pits and mounds, buried seeds, and the importance of soil disturbance to pioneer trees on Barro Colorado Island, Panama. Ecology 64, 1069–1074.

R Development Core Team, 2013. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria.

Reid, J.L., Holl, K.D., Zahawi, R.A., 2015. Seed dispersal limitations shift over time in tropical forest restoration. Ecol. Appl. 25, 1072–1082.

Saldanha, C.J., Nicholson, D.G., 1976. Flora of Hassan District. Amerind Publishing Company, New Delhi.

Sangsupan, H., 2017. Limitations to seedling regeneration on tropical forest restoration plantations in northern Thailand. Ph.D. dissertation. Oregon State University, Corvallis.

Schnitzer, S.A., Carson, W.P., 2001. Treefall gaps and the maintenance of species diversity in a tropical forest. Ecology 82, 913–919.

Sinhaseni, K. 2008. Natural establishment of tree seedling in forest restoration trials at Ban Mae Sa Mai, Chiang Mai Province. M.S. Thesis. Chiang Mai University, Chiang Mai, Thailand.

Svenning, J.-C., Wright, S.J., 2005. Seed limitation in a Panamanian forest. J. Ecol. 93, 853–862.

Thai Meteorological Department, 2012. Annual Weather Summary of Thailand in 2011. Thai Meteorological Department.

Thaiying, J., 2003. The Diversity of Small Mammal Communities in FORRU's Reforested Areas. Chiang Mai University, Chiang Mai, Thailand.

Tunjai, P., Elliott, S., 2012. Effects of seed traits on the success of direct seeding for restoring southern Thailand's lowland evergreen forest ecosystem. New For. 43, 319–333.

Vargas, I.N., Stevenson, P.R., 2013. Seed and establishment limitation: Effects on plant diversity in an Amazonian rain forest. Biotropica 45, 737–746.

Vazquez-Yanes, C., Orozco-Segovia, A., 1992. Effects of litter from a tropical rainforest on tree seed germination and establishment under controlled conditions. Tree Physiol. 11, 391–400.

Vazquez-Yanes, C., Orozco-Segovia, A., 1996. Physioloigcal ecology of seed dormancy and longevity. In: Mulkey, S., Chazdon, R.L., Smith, A.P. (Eds.), Tropical Forest Plant Ecophysiology. Chapman and Hall, New York, pp. 535–558.

Vieira, D.L.M., Scariot, A., 2006. Principles of natural regeneration of tropical dry forests for restoration. Restor. Ecol. 14, 11–20.

Wada, N., 1993. Dwarf bamboos affect the regeneration of zoochorous trees by providing habitats to acorn-feeding rodents. Oecologia 94, 403–407.

Whittaker, R.H., Levin, S.A., 1977. The role of mosaic phenomena in natural communities. Theor. Popul Biol. 12, 117–139.

Woods, K., Elliott, S., 2004. Direct seeding for forest restoration on abandoned agricultural land in northern Thailand. J. Trop. For. Sci. 16, 248–259.

Wotton, D.M., Kelly, D., 2011. Frugivore loss limits recruitment of large-seeded trees. Proc. R. Soc. B: Biol. Sci. 278, 3345–3354.

Wright, S.J., Stoner, K.E., Beckman, N., Corlett, R.T., Dirzo, R., Muller-Landau, H.C., Nuñez-Iturri, G., Peres, C.A., Wang, B.C., 2007. The plight of large animals in tropical forests and the consequences for plant regeneration. Biotropica 39, 289–291.

Wydhayagarn, C., Elliott, S., Wangpakapattanawong, P., 2009. Bird communities and seedling recruitment in restoring seasonally dry forest using the framework species method in northern Thailand. New For. 38, 81–97.

Yu, Y., Baskin, J.M., Baskin, C.C., Tang, Y., Cao, M., 2008. Ecology of seed germination of eight non-pioneer tree species from a tropical seasonal rain forest in southwest China. Plant Ecol. 97, 1–16.

Zahawi, R.A., Holl, K.D., 2009. Comparing the performance of tree stakes and seedlings to restore abandoned tropical pastures. Restor. Ecol. 17, 854–864.