

SOIL SEED BANK CHARACTERISTICS UNDER AND OUTSIDE CANOPY OF *DACRYDIUM* LAMB. IN CHINA

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Abstract

Understanding of how canopy affects the spatiotemporal patterns of soil seed bank is of importance for addressing the problems of forest regeneration originating from seed bank. This study investigated the soil seed bank characters under and outside the canopy of genus *Dacrydium* Lamb. community in Hainan tropical rain forest. Results show that a total of 194 plant species seeds exist in soil seed bank, the seed density of M_6 (80 cm < DBH₆ ≤ 90 cm) was the highest (5370 seeds·m⁻²) in December. The distribution of seeds in four directions was significantly different. The seed densities under the canopies were greater than those of outside in three seasons. The trees with DBH between 60 and 90 cm have high seed vigor in December. It was concluded that the canopy of trees has important effects on seed accumulation and seed vigor, which is vital for forest management and regeneration, especially for taking protection measures to forest.

Introduction

The typhoon and tsunami phenomenon has made the global forest area drastically reduced, the survival of rare and endangered species become difficult (Sardans *et al.* 2017). The protective measures must be considered for the survival and reproduction of rare and endangered tree species (Bossuyt and Honnay 2008). Seed source is the foundation of the settlement, survival, reproduction, and spread of forest population (Todd *et al.* 2016). Soil seed bank characteristics not only reflect the past status of the forest, but also represent possible future development trends of the forest (Lucas-Borja *et al.* 2017). Therefore, it is of great significance to study soil seed bank characteristics for their species protection.

Previous studies have determined that the species richness and seed density of soil seed banks affect the temporal and spatial characteristics of above-ground vegetation (Skowronek *et al.* 2014). The seasonal dynamics of soil seed banks was considered to understand the ecological significance of soil seed banks in natural tropical forests (Qian *et al.* 2016). Therefore, seeds in the soil below or outside the tree canopy have a distribution pattern (Pugnaire and Lázaro 2000, Marod *et al.* 2002).

Dacrydium is the only genus of Podocarpaceae family that exists in China. The species is a third-class rate-endangered plant species under the China red data book classification (Keppel *et al.* 2011). The main objectives of this study were as follows (i) to investigate changes of soil seed bank pattern response for under and outside of tree canopies, and (ii) to determine how the seed bank responds to the DBH of the sample trees and the variation of seasons in the years.

Materials and Methods

The study was conducted at the Hainan Bawangling National Nature Reserve (109°03' - 109°17' E, 18°57' - 19°11' N), the Jianfengling National Forest Park (108°39' - 109°24' E, 18°24' - 18°58' N), and the Diaoluoshan National Forest Park (109°43' - 110°03' E, 18°43' - 18°58' N), which are the only areas in China where genus *Dacrydium* grows (Chen *et al.* 2014) (Fig. 1).

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According to survey, the range of the DBH size of the trees was from 17 to 136 cm. Therefore, samples were divided into 11 ranks according to DBH, which are $DBH_1 \leq 40$ cm (M_1), 40 cm $< DBH_2 \leq 50$ cm (M_2), 50 cm $< DBH_3 \leq 60$ cm (M_3), 60 cm $< DBH_4 \leq 70$ cm (M_4), 70 cm $< DBH_5 \leq 80$ cm (M_5), 80 cm $< DBH_6 \leq 90$ cm (M_6), 90 cm $< DBH_7 \leq 100$ cm (M_7), 100 cm $< DBH_8 \leq 110$ cm (M_8), 110 cm $< DBH_9 \leq 120$ cm (M_9), 120 cm $< DBH_{10} \leq 130$ cm (M_{10}), and 130 cm $< DBH_{11}$ (M_{11}).

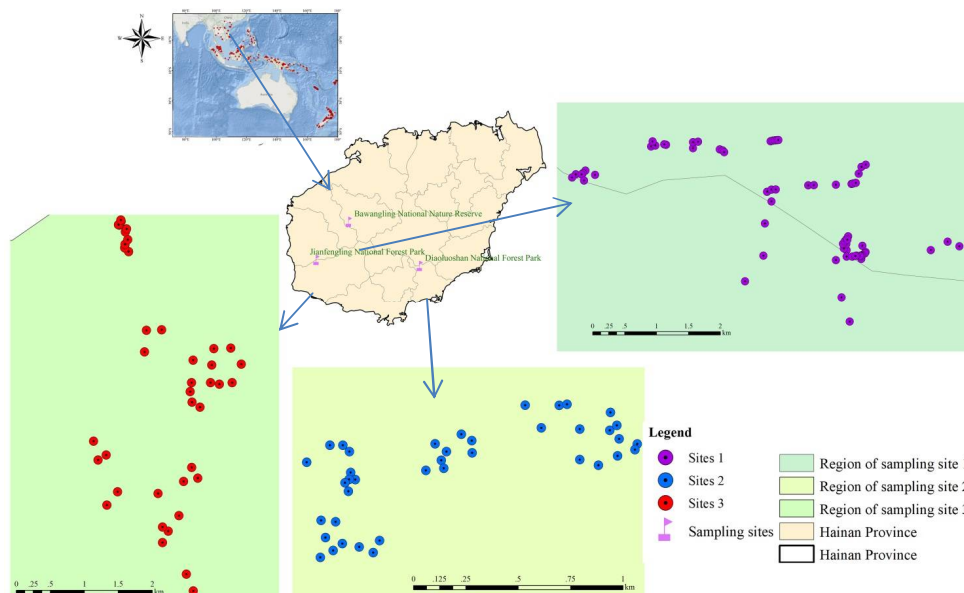


Fig. 1. Location of the study area comprising of the Hainan Bawangling National Nature Reserve, the Jianfengling National Forest Park, and the Diaoluoshan National Forest Park in China and the spatial distribution of the sample plots. The natural population of genus *Dacrydium* is distributed sparsely and along the ridge line without regularity.

Taking mother tree of every rank as the center, the survey plots (20 m \times 20 m) were set up and characteristics of each site were investigated (Schwab and Kiehl 2017). Taking sample trees as the center point, the soil samples were collected under and outside the canopies of these mother trees. The radius of the crown was less than 6 m, and maintain stability with the DBH increasing. ‘Under the canopies’ refers to samples 2 , 4 , 6 m from the center of tree, and ‘outside the canopies’ refers to samples 2 , 4 , 6 m from the edge of the trees, and then, taking the average of three sites as the final value, as the aim being to represent two different micro-sites. We set each soil seed bank plot in east, west, south and north direction of mother tree. Soil sample size was 10 cm \times 10 cm.

Depending on the flowering, seed germination, maturation and drop characteristics of *Dacrydium*, seed bank samples were investigated in April (after flowering), September (before seed maturity), and December (after seed maturity) of each year from 2013 to 2017. Three *Dacrydium* trees in each site were selected, a square metal box of 10 cm \times 10 cm was used to obtain samples from a depth of 10 cm, and then the samples were taken to the laboratory of the research station. Each sample was divided into three layers: the litter layer, the first soil layer (0 - 5 cm) and the second soil layer (6 - 10 cm) (Danny *et al.* 2015). The soil samples were air-dried and then sieved using a 1 mm screen. All roots, litter, and other debris in soil were picked-out.

Samples from the surface of the plastic basins were taken back, air-dried and sieved again. Seeds were extracted, and their viabilities were tested by TTC (Hui and Wang 2009). Thus, the number of viable (including germinated), or dead seeds per soil sample was determined. Only viable seeds were considered for assessing the composition and density of the soil seed bank.

Analyzing soil seed bank data in different time and space using the ANOVA, significant differences among them were compared, and spatial distribution characteristic of soil seed bank was inferred using geostatistics (Hui and Wang 2009).

Results and Discussion

The result showed the seed density have an obviously change in seasons and site-to-site. The difference in soil seed density at the same location among three seasons was significant ($p < 0.05$). Soil seed density was the highest in December, minimum in April and the least in September. That of M_1 , M_2 , M_6 and M_9 in April were lower than that in September. The seed density of M_6 was the highest ($5370 \text{ seeds}\cdot\text{m}^{-2}$) in December, and that of M_1 was the lowest (328) in April in that times (Fig. 2).

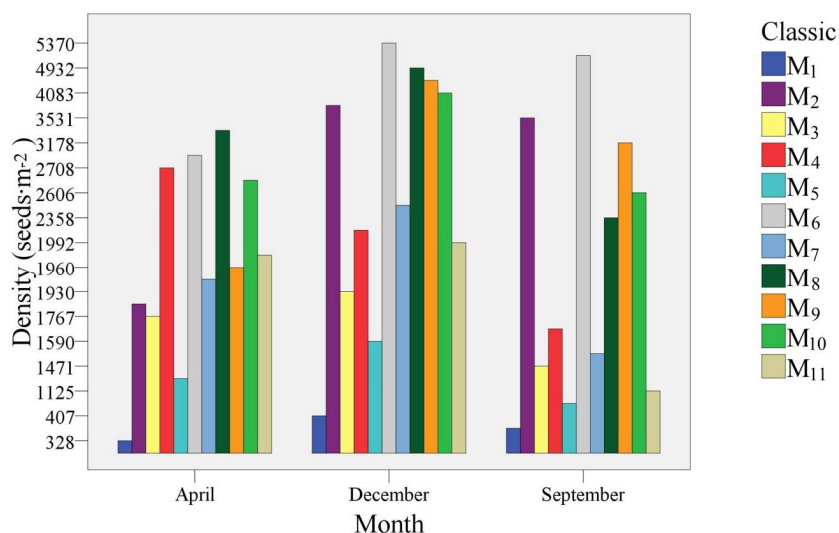


Fig. 2. Seed density of soil seed bank in different seasons. M_1 : The sample trees that $\text{DBH}_1 \leq 40 \text{ cm}$; M_2 : that $40 \text{ cm} < \text{DBH}_2 \leq 50 \text{ cm}$; M_3 : that $50 \text{ cm} < \text{DBH}_3 \leq 60 \text{ cm}$; M_4 : that $60 \text{ cm} < \text{DBH}_4 \leq 70 \text{ cm}$; M_5 : that $70 \text{ cm} < \text{DBH}_5 \leq 80 \text{ cm}$; M_6 : that $80 \text{ cm} < \text{DBH}_6 \leq 90 \text{ cm}$; M_7 : that $90 \text{ cm} < \text{DBH}_7 \leq 100 \text{ cm}$; M_8 : that $100 \text{ cm} < \text{DBH}_8 \leq 110 \text{ cm}$; M_9 : that $110 \text{ cm} < \text{DBH}_9 \leq 120 \text{ cm}$; M_{10} : that $120 \text{ cm} < \text{DBH}_{10} \leq 130 \text{ cm}$ and M_{11} : that $130 \text{ cm} < \text{DBH}_{11}$.

The statistical results showed that the distribution of seeds in all directions was significantly different ($p < 0.05$) (Table 1). The seed density in the east seed bank of M_1 mother tree was $341 \text{ seeds}\cdot\text{m}^{-2}$, accounting for 31.12% of the total number of seeds; that was 193 (17.59%), 314 (28.69%), 247 (22.60%) $\text{seeds}\cdot\text{m}^{-2}$ in the westward, southward, northward direction, respectively. The order of seed bank densities from most to least in four directions was east > south > north > west. The average seed density of 11 samples was also significantly different, M_6 had the biggest seed density (3 328) among them, and M_1 had the smallest one.

Compared to other models, after calculating several times, the result showed that an exponential model was chosen for all sites (Table 2). The highest coefficient of determination for the model fitting effect was 0.76 (M_6). The sill ($C_0 + C$) of *Dacrydium* with DBH size that $80 \text{ cm} < \text{DBH}$

$6 \leq 90$ cm (M_6) was big (8.094), the extent of spatial heterogeneity of distribution of *Dacrydium* with DBH size between 80 and 90 cm was higher than that of trees with DBH sizes within other ranges.

Table 1. Seed density of soil seed bank in different directions (mean value \pm standard deviation).

Number	Density (seeds·m ⁻²)				Average
	East	West	South	North	
M ₁	341±82	193±78	314±121	247±142	274±26
M ₂	3150±269	1921±207	1429±170	2950±366	2363±253
M ₃	2321±173	1048±69	1216±184	583±159	1292±196
M ₄	332±29	1384±112	3514±178	1439±265	1667±121
M ₅	103±75	274±19	634±157	2143±275	789±206
M ₆	1759±384	235±230	1349±719	9969±704	3328±609
M ₇	242±41	727±98	3535±113	1367±228	1468±140
M ₈	1145±70	3573±54	3496±44	2531±95	2686±166
M ₉	1883±44	1316±179	4444±216	2033±152	2419±248
M ₁₀	2477±60	1908±233	1561±163	3365±550	2328±252
M ₁₁	307±74	1181±188	825±564	2788±274	1276±451

Table 2. Spatial pattern variation functions parameter of the soil seed bank of different mother trees.

Classic	Theoretical model	Nugget (C ₀)	Sill (C ₀ +C)	Spatial variability [C/(C ₀ +C)]	Range (A/m)	(R ²)	(RSS)
M ₁	Exponential	1.014	3.382	0.700	19.6	0.56	2.923
M ₂	"	1.893	5.965	0.683	18.2	0.61	2.571
M ₃	"	2.975	8.001	0.628	17.8	0.63	2.145
M ₄	"	2.325	8.973	0.741	17.4	0.59	1.867
M ₅	"	2.801	10.382	0.730	16.9	0.73	1.541
M ₆	"	2.552	10.094	0.747	16.5	0.76	1.207
M ₇	"	2.784	9.842	0.717	17.2	0.72	2.546
M ₈	"	3.932	8.644	0.545	18.1	0.68	2.854
M ₉	"	3.402	8.072	0.579	17.6	0.67	3.061
M ₁₀	"	2.17	4.907	0.558	17.4	0.58	2.583
M ₁₁	"	2.454	7.973	0.692	18.5	0.62	2.116

RSS: Residual sum of squares; R²: Coefficient of determination.

Average seed densities of genus *Dacrydium* in M₁ - M₁₁ was 1 095, 9 451, 5 168, 6 669, 3 155, 13 312, 5 872, 10 745, 9 676, 9 312, 5 102, respectively (Table 2). This result indicated that seed densities of soil seed bank in *Dacrydium* community had a "Parabolic shape" changes with the DBH of trees increased significantly ($p < 0.05$). When DBH size smaller than 90 cm, the seed densities increased with the size of DBH increase, when DBH size between 100 and 130 cm, the seed densities decreased with the size of DBH increase. Seed density between under and outside the canopy did not change significantly with different DBH size, the trend of seed density in the layers was decreasing with the depth of soil increase, and the seed density not changed significantly with the increase of DBH in different seasons.

There were considerable differences in the viable seeds number in three seasons with a DBH sequence ($p < 0.01$) (Fig. 3). The results showed that the seed vigor rate in December was the

highest, followed by that in September, and in April. The highest seed viability was M_4 in December; the lowest of that was M_1 in April. In December, M_4 had the highest seed viability (41%), followed by M_1 (32%) and then M_5 (31%). In September, M_6 had the highest seed vigor (26%), followed by M_4 (23%) and then M_9 (19%). In April, M_5 had the highest seed vigor (16%), followed by M_3 (15%) and then M_{11} (11%).

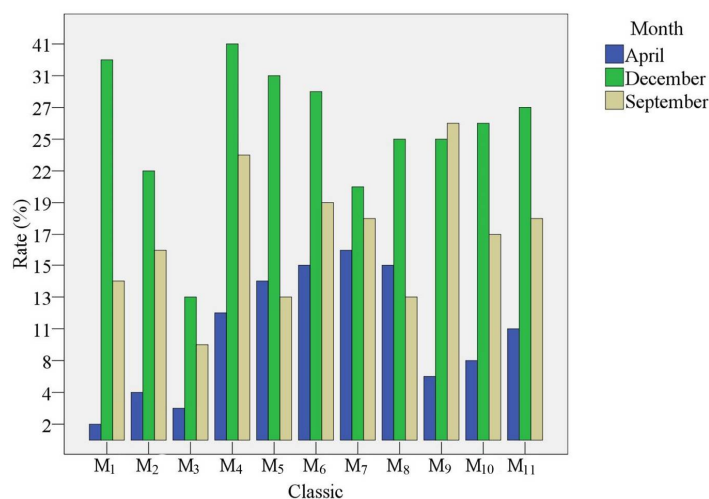


Fig. 3. Dynamics of seed vigor in soil seed banks.

The peak seed density occurred in December when seed production was completed (Fig. 4). Differences in the seed densities between under and outside the canopies were not significant for $DBH \leq 40$ cm (M_1) plantation sites for three seasons. That of M_{10} site in December, as indicated by the microsite-season interactions. They had a great difference in amount of seed density between under and outside canopies in the 6 - 10 cm layer in December, which was less than that in 0 - 5 cm and litter layer in December. It has same situation in litter layer and 6 - 10 cm layer was in September, while the seed densities under the canopies were smaller than that of outside in 0 - 5 cm layer in M_5 , M_7 , M_8 , M_9 , M_{11} , having no seed in under and outside the canopies in 6 - 10 cm layer in April, the seed densities under the canopies were smaller than that of outside in 0 - 5 cm layer in M_1 , M_2 , M_8 , the situation in litter layer in M_3 and M_7 was the same. The seed densities of under and outside canopies were the largest in M_6 , which was more than 10,000.

Seed density under the canopies was totally different than those of outside in three seasons, indicating that canopy has effect on the composition, richness, and density of soil seed bank (Tessema *et al.* 2017). This might be caused by the seed gravity effect for the genus *Dacrydium*, which allowed the seeds fall from the canopy into the soil directly when it was matured, and the seeds can be transported to outside of canopy in the case of windy as a carrier of transport. The temporal and spatial distribution patterns of soil seed banks were affected by that of the DBH size of trees, the seasons of seed collected, wind direction, and canopy size, which result in soil seed bank having an observable spatial distribution pattern.

According to the result that having not obvious regular pattern existed in the distribution of seeds density of soil seed bank in four directions (Yudi *et al.* 2017). This might be caused by the wind force affection, the main direction of wind of Hainan Island is the southwest or southerly, the main wind in winter is northeast, north wind sometimes (Farjon and Filer 2013). Judging from the prevailing wind direction, seeds in the north and northeast should be more than other directions, and

seeds in the south and southwest should be less than other directions. However, it was also observed to have many seeds in the south direction, which was affected by different populations of the community, canopy shelter around each other, so that the bearing capacity of tree seed is different.

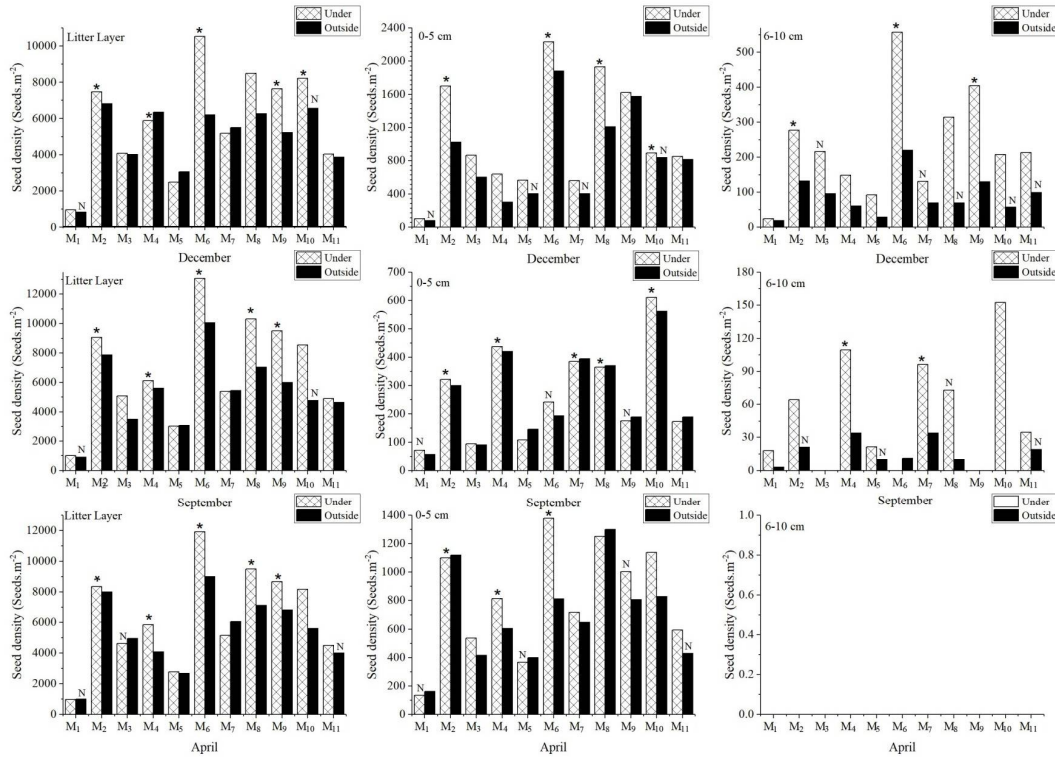


Fig. 4. Seed density under and outside the canopies in a DBH sequence of *Dacrydium* community. *: $p < 0.05$; N: Not significant for seed densities between microsites; Under: The seed density of under of the canopy's region; Outside: The seed density of outside of the canopies region.

The characters of soil seed density in season showed that the characteristics of soil seed bank in *Dacrydium* communities have an obvious correlation with season (Sousa *et al.* 2014). Many tree species produce fruit during the flowering season, or some tree species that produce fruit one after another over the seasons (Lozano *et al.* 2017). It is the best choice for one to understand about the fruit species and quantity status of the community before producing large amounts of mature seeds in September, this is also the reason why soil seed density was the least in September. After seed became maturity, the seeds of many species are gathered to the maximum together in December. It is therefore important that much exploration should be done in December to get the detailed research result for soil seed bank of the communities.

In conclusion, the soil seed bank of *Dacrydium* communities contained variety species of forest tree, shrub and herb in tropical rain forest of Hainan, China. The factors that the canopy size and DBH size of *Dacrydium* communities affect the spatiotemporal distribution of seeds in soil and seed vigor. Therefore, keeping appropriate canopy area and DBH size of trees of communities by artificial measure should be chosen for forest management to reduce natural regeneration barriers, which can improve the regeneration ability of the communities in ecosystem.

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