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The role of plant functional traits in understanding forest recovery in wet tropical secondary forests
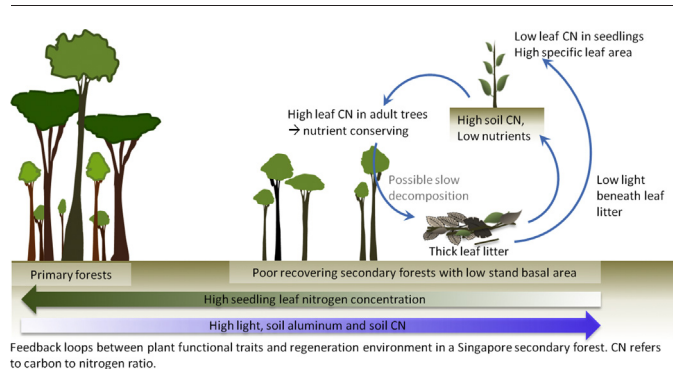
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HIGHLIGHTS

- Environmental variables were significantly correlated with seedling traits.
- Soil aluminum is lower but seedling leaf nitrogen is higher in primary forests.
- Opposing trends in sec forests as specialist species accrued high nutrients.
- Seedling traits reflect successful strategies, not necessarily forest recovery.

GRAPHICAL ABSTRACT

Feedback loops between plant functional traits and regeneration environment in a Singapore secondary forest. CN refers to carbon to nitrogen ratio.

ARTICLE INFO

ABSTRACT

Simultaneous measurement of plant functional traits and the regeneration environment should shed light on the plant-environment interactions and feedbacks as secondary forest regenerates. However, little of such work has been done in the wet tropics, and even fewer studies have examined soil nutrients. We investigated whether plant functional traits and environmental variables explain the varied recovery of secondary forests in Singapore. Our study plots included three primary forest plots and eight approximately 60-year-old secondary forest plots regenerating from intensive agricultural activities. Using 35 seedling quadrats, we asked: Q1) How do environmental variables explain the variation in seedling functional traits observed in primary and secondary forests? Q2) How do seedling traits, adult traits and environmental variables relate and explain variation in species richness and stem density in secondary forests? We found that both light and soil fertility explained the shifts in plants functional traits from poorly recovering secondary forests to primary forests. Poor forest regrowth was correlated with high soil aluminum levels and lower leaf nitrogen concentrations. Low nutrients and high aluminum saturation were also negatively correlated with seedling species richness, but not stem density, in the secondary forests. Forest recovery is probably slowed by positive feedback between slower nutrient returns from slow decaying litter and further recruitment of nutrient conserving species, as indicated by positive correlations among adult leaf CN ratio, litter depth, soil CN ratio and quadrat level CN ratio. Plant functional traits are indicative of the strategies of successful seedlings and do not necessarily relate to overall forest recovery. Hence, while some specialist plant species are able to accrue high nutrients on degraded soils with aluminum toxicity and low nutrients, species richness on these soils was poor. This underscores the need to concurrently measure environmental variables and plant traits when investigating the mechanisms driving changes during forest recovery.

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1. Introduction

With widespread anthropogenic disturbances in the tropics (Asner et al., 2009), secondary forests are fast becoming the forests of tomorrow, and their potential to support biodiversity as well as ecosystem functions hinges on their recovery. In general, secondary forests recover aboveground biomass with increasing water availability (Poorter et al., 2016) and soil fertility (Lu et al., 2002; Moran et al., 2000). However, beyond forest biomass and structure, the recovery of floristic composition is harder to predict (Chazdon, 2008; Guariguata and Ostertag, 2001; Peterson and Carson, 2008), often proceeding at varying rates (Corlett, 1995; Finegan, 1996; Letcher and Chazdon, 2009), or stagnating over time and taking hundreds of years for species composition to converge to that of pre-clearance forests (Finegan, 1996; Guariguata and Ostertag, 2001).

Key to understanding secondary succession is to study the processes that drive species replacement as the forest regenerates (Huston and Smith, 1987; Pickett and Collins, 1987). Succession is an evolving process where the changing structure and diversity of the recovering forest continuously interacts with deterministic (environmental conditions) and stochastic (dispersal events, catastrophic events, etc.) factors to influence subsequent forest dynamics (Huston and Smith, 1987). The changing tree community alters the forest environment, which in turn favors species whose suite of characteristics is best suited to those new conditions. Over time, as trees grow, the adult trees have an increasing influence on the regenerating environment and hence on the composition of recruiting species (Grime, 1998). Current theory also suggests that as tropical forests regenerate, the environment often progresses from a high resource state with high light, high nutrient input to a low resource state with low light and low nutrients. Correspondingly, fast-growing species that rapidly acquire resources are replaced over time by slow-growing, resource conserving species (Bazzaz and Pickett, 1980; Garnier et al., 2004; Huston and Smith, 1987; Odum, 1969). For example, decreasing light has been postulated as the main driver in wet tropical rainforests (Lohbeck et al., 2013, 2015; Wright et al., 2010). Other studies have documented declining or constant soil nutrients (Feldpausch et al., 2004; Hughes et al., 1999; Johnson et al., 2001) as a forest recovers, providing some support of the change from high to low resource over succession. However, past research have also shown that forests originating on soil with low fertility tend to recover more slowly from disturbances (Corlett, 1991; Lu et al., 2002), and are often colonized by species that are more nutrient conserving (Chua et al., 2013). Overall, we know very little about how resources, in particular soil nutrients, interact to affect the recruitment of plants with different characteristics. Examination of these cause and effect relationships would develop a more mechanistic understanding of secondary forests recovery.

One approach to understanding the dynamics of community assembly during succession has been through studying corresponding shifts in functional trait composition (Boukili and Chazdon, 2017; Craven et al., 2015; Lasky et al., 2014; Lohbeck et al., 2013), which reflect plants' strategies and responses to the environment (Gitay et al., 1999; Lavorel et al., 1997). For example, fast growing species generally have high specific leaf area (leaf area per mass), leaf nitrogen concentration and low leaf dry matter content (dry mass per fresh mass) to facilitate high rates of photosynthesis at low leaf construction cost, but the leaves are often short-lived. In contrast, slow-growing, resource conserving species have leaves with greater longevity and slower photosynthetic returns. This continuum of low cost, fast returns of inexpensive short-lived leaves to high cost, slow returns of costly long-lived leaves has been coined the "leaf economics spectrum" (Wright et al., 2004), and is exemplified by fast growing early successional species being replaced by late-successional species in wet tropical forests, as a response to decreasing light availability (Lohbeck et al., 2015). Similarly, wood density has been thought to be a trade-off between growth and survival – pioneer species tend to have low stem density with higher hydraulic

conductivity, trading low cost wood construction for biomechanic safety, while late successional species invest in higher wood density for protection against physical damage and herbivory (Chao et al., 2008; Chave et al., 2009; King et al., 2006). Recent work in the Neotropics has found that the leaf and stem traits are orthogonal (Baraloto et al., 2010; Fortunel Claire et al., 2012), which not only suggest a greater flexibility for different plant species to strategize resource allocation under varying environmental conditions, but also emphasizes the need to include both leaf and stem traits when investigating plants' strategies. When scaled up appropriately to community level by biomass (Grime, 1998, Garnier et al., 2004), plant functional traits and other forest stand level characteristics such as stand basal area, are measures of the whole plant community's effect on the environment (Lavorel and Garnier, 2002; Quétier et al., 2007). For example, the community-weighted means of leaf economic traits have been found to predict the litter decomposition and hence nutrient cycling in different tropical land use types (Bakker et al., 2011; Cornwell et al., 2008). A criticism of the functional traits approach is that beyond environmental filtering, functional traits could also response to biotic interactions such as competition (Cadotte and Tucker, 2017; Kraft et al., 2015). Measuring environmental variables would improve inferences on the relative strength of abiotic influences (Cadotte and Tucker, 2017), but such work is lacking in tropical rainforests, and of the few studies conducted, most were in dry tropical rainforests (Becknell and Powers, 2014; Lebrija-Trejos et al., 2010).

To help fill this knowledge gap, our study examined the relationships between selected traits in seedlings and the environmental variables that we had previously shown to affect seedling regeneration in the successional wet tropical rainforests of Singapore (Chua et al., 2016). We also measured adult traits to explore possible influence of adult traits on the regeneration environment, and vice versa. The forests in Singapore underwent intense agriculture activities from the mid-1800s to 1900s. Areas depicted by old aerial photographs as sizeable opened canopy areas in the 1950s have recovered differently today, with some still fernlands-dominated, while others are secondary forests with distinct species composition and structure. In some cases, forest recovery even appears to be arrested (Goldsmith et al., 2011) despite the contiguity of the landscape and relative accessibility of seed sources from mature secondary forests, which suggests strong environmental filtering on site. Since these forests are estimated to be fairly similar aged, about 60-year-old, our approach is not via chronosequence interpretation, but rather, to emulate conventional studies of how forest community functional composition could be influenced by environmental factors and adult trees (Allié et al., 2015; Fortunel et al., 2014; Gastauer and Meira-Neto, 2014; Jager et al., 2015). We measured nine leaf traits in the adult and seedling communities: specific leaf area (SLA), leaf dry matter content (LDMC), leaf concentration of nitrogen (LNC), phosphorus (LPC), potassium, calcium and magnesium, CN and NP ratios. Although we acknowledge that environmental filtering might be stronger at later growth stage due to certain amount of lottery effect as the seedling establishes (Busing and Brokaw, 2002), we focused on the seedling stage because environmental filtering at the seedling stage has been shown to have long-lasting effects on plant ecological strategies and the overall community assembly process (Grubb, 1977; Poorter, 2007). We also utilized the fourth-corner approach, which is less often employed by plant traits researchers (but see Blundo et al., 2015), but has been argued to be superior in testing for trait-environment correlations over more conventional methods of community weighted means correlation and species niche centroid correlation (Peres-Neto et al., 2017), and comparable with but less computing-intensive than GLM-based tests (ter Braak et al., 2017).

In summary, our study investigated the relationships among seedlings traits, adult tree traits and environmental variables, and explored how these relationships explain forest recovery in terms of seedlings species richness and stem density. Our research was guided by the following specific questions:

2.1. How do environmental variables explain the variation in seedling functional traits observed in primary and secondary forests?

Q1. How do seedling traits, adult traits and environmental variables relate and explain variation in species richness and stem density in secondary forests?

2. Materials and methods

2.1. Site description

Our study was located in the Central Catchment Nature Reserve (CCNR) (1° 22′ 32.0514″, 103° 48′ 13.0674″), Singapore. The mean temperature is 27.0 °C and the mean annual rainfall is 2353 mm, with no month having <150 mm of rain on average (National Environment Agency, Singapore). The elevations of the study sites ranged from 25 to 55 m. CCNR overlays the Bukit Timah granite, which has given rise to Typic Paleudults soils of the Rengam series (Ives, 1977). The soils are acidic and particularly low in phosphorus (Thomas, 1991). The original forests are an extension of the lowland Dipterocarp forests in the Malay Peninsula. The arrival of the British in the 19th century resulted in an influx of migrants and rapid deforestation for cash crops such as gambier (*Uncaria gambier*) and pepper (Corlett, 1992), although the precise history of our plots remains unclear. In the twentieth century, farming activities ceased as the island urbanized and the remaining forests were left to regenerate naturally, culminating in the establishment of nature reserves in 1951. Today, the nature reserve encompasses around 2000 ha of 60–100 year old secondary forests and scattered pockets of primary Dipterocarp forest surrounding four water reservoirs.

2.2. Plot selection and sampling design

Our plot selection was based on forest structural maps that were created from stereoscopic aerial photographs taken from 1950 to 1952 and a forest structural classification by Wong et al. (1994) and refined by Turner et al. (1996). We selected eight plots that were open vegetation with only herbaceous cover in 1950s, and are today at various stages of regeneration, ranging from open vegetation, forests with small pole trees to tall mature secondary forests. Three primary forest plots were also selected for comparison. At each site, we laid out a 0.25-ha octahedral plot, 50 m across (Fig. 1). In order to capture as much site heterogeneity as possible, we first randomly sampled seedling quadrats of 5 × 5 m within the plot. In each quadrat, we surveyed all seedlings ≥ 10 cm in height and <1 cm DBH. We surveyed 200 seedlings or a minimum of four and maximum of 20 quadrats from each tree plot, in total 91 seedling quadrats from all 11 tree plots. Then within each plot, based on a NMDS ordination of seedling species composition, we selected four quadrats with high species dissimilarity for environmental monitoring. An exception was made for two plots that were dominated by the fern, *Dicranopteris linearis* and had few to no seedlings, and hence environmental measurements were made in only three quadrats. To include trees that could have influenced abiotic conditions and seedling regeneration, we identified, measured and tagged all trees ≥ 10 cm diameter at breast height (DBH) within the 50 m plot, as well as in an additional outer 11 m annulus. Tree species were cross-referenced with specimens from the Singapore Herbarium (SING).

Due to data loss in the field and laboratory we ended up with 24 secondary forests quadrats and 11 primary forests quadrats for analysis (Fig. 1), which are 80% and 90% of the original quadrats established for the intensive analysis. In addition, these quadrats account for 3–3.67% of the secondary and primary forests plots sampled respectively. This sampling intensity is consistent with previous studies of seedlings in tropical forests, which sampled on average 0.2–4.0% of forest plot area (Comita et al., 2010; Comita and Hubbell, 2009; Murphy et al., 2017).

2.3. Environmental variables

In each selected seedling quadrat, we measured the regeneration environment that likely influenced the growth and performance of seedlings. We monitored from March 2012 to March 2013 the relative humidity and air temperatures of the selected seedling quadrats using iButton[R] sensors (model DS1923). Each iButton was housed in an opaque ventilated housing and staked 1 m from the ground. Temperature measurements were not used for this work as previous analysis showed that vapor pressure deficit, and not temperatures, was more important to seedling recruitment (Chua et al., 2016). Canopy structure and gap light transmission were quantified with hemispherical photographs taken with a 4.5 mm F2.8 Ex DC Circular Fisheye HSM Sigma DC Lens mounted on a Cannon digital SLR Camera. The photographs were analyzed using the Gap light analyzer software (GLA version 2.0) which calculates the percentage of canopy cover (Frazer et al., 2000). The mean daily standard deviation of vapor pressure deficit (sdV) was calculated for each seedling quadrat. It was less meaningful to use maximum vapor pressure deficit as humidity commonly exceeds 95% in Singapore. In addition, a composite of the top 5 cm organic soil was collected from beneath the leaf litter in each of the seedling quadrats using a 2-cm diameter soil probe. After removing the un-decomposed organic materials, the soils were sent to Soils Laboratory of the Smithsonian Tropical Research Institute and analyzed for resin-extractable phosphorus (P), exchangeable cations with $BaCl_2$, pH, total carbon (TC) and total nitrogen (TN). Finally, we took measurements of the mean leaf litter depth with a stiff measuring tape. Refer to Table 1 for the list of abbreviation used for traits and environmental variables in our study.

A complete description of the plot sampling design and measurement of environmental variables is detailed in Chua et al. (2016).

2.4. Plant traits

Seedling leaf traits were collected from both primary and secondary forests (for Q1) while adult leaf traits were collected only from secondary forest plots (for Q2). For seedlings, leaves were collected from species which accounted for on average 90% of the seedling density in each quadrat (3–16 species, which constituted 68–100% of the individual seedling stems in each quadrats). Since many of the seedlings had few leaves, we collected one leaf per seedling and composited leaf specimens from an average of three individuals (ranging from 1 to 7) per species per site. All species except one had at least three individuals sampled. Similarly, for adult trees, the leaves collected accounted for on average 88% of the plot basal area (16–21 species per plot, which constituted 77–96% of the total tree basal area in the nine secondary forest plots). We focused only on large trees with DBH ≥ 10 cm and trees with numerous coppicing stems, such that the total basal area per individual tree was ≥78.5 cm^2 (equivalent of basal area of tree with DBH ≥ 10 cm). Leaves from the outer crown of the trees were collected using a long pruner, slingshots, or by catapulting an abrasive fishing line and sawing a thin shoot/branch with the line. At least three leaves from each adult tree and at least three to six individuals per species were collected, except for three species where there were less than two individuals in the entire survey area. The cut end of the leaves/shoot was wrapped in damp tissues and the specimens were put in Ziploc bags and transported in a cooler box to the laboratory.

In both seedlings and adult trees, we measured nine leaf traits: specific leaf area (SLA), leaf dry matter content (LDMC), leaf concentration of nitrogen (LNC), phosphorus (LPC), potassium, calcium and magnesium, CN and NP ratios. Leaf area and mass measurement were done on entire leaves, including the petioles (Cornelissen et al., 2003). The fresh weight of the leaves was measured within six to 24 h, during which they were stored in damp paper towels and in a refrigerator. Given the wet local climate with frequent thunderstorms at the time of collection, the leaves should have been near saturated fresh weights at the point of measurement. Leaf areas of individual leaf were
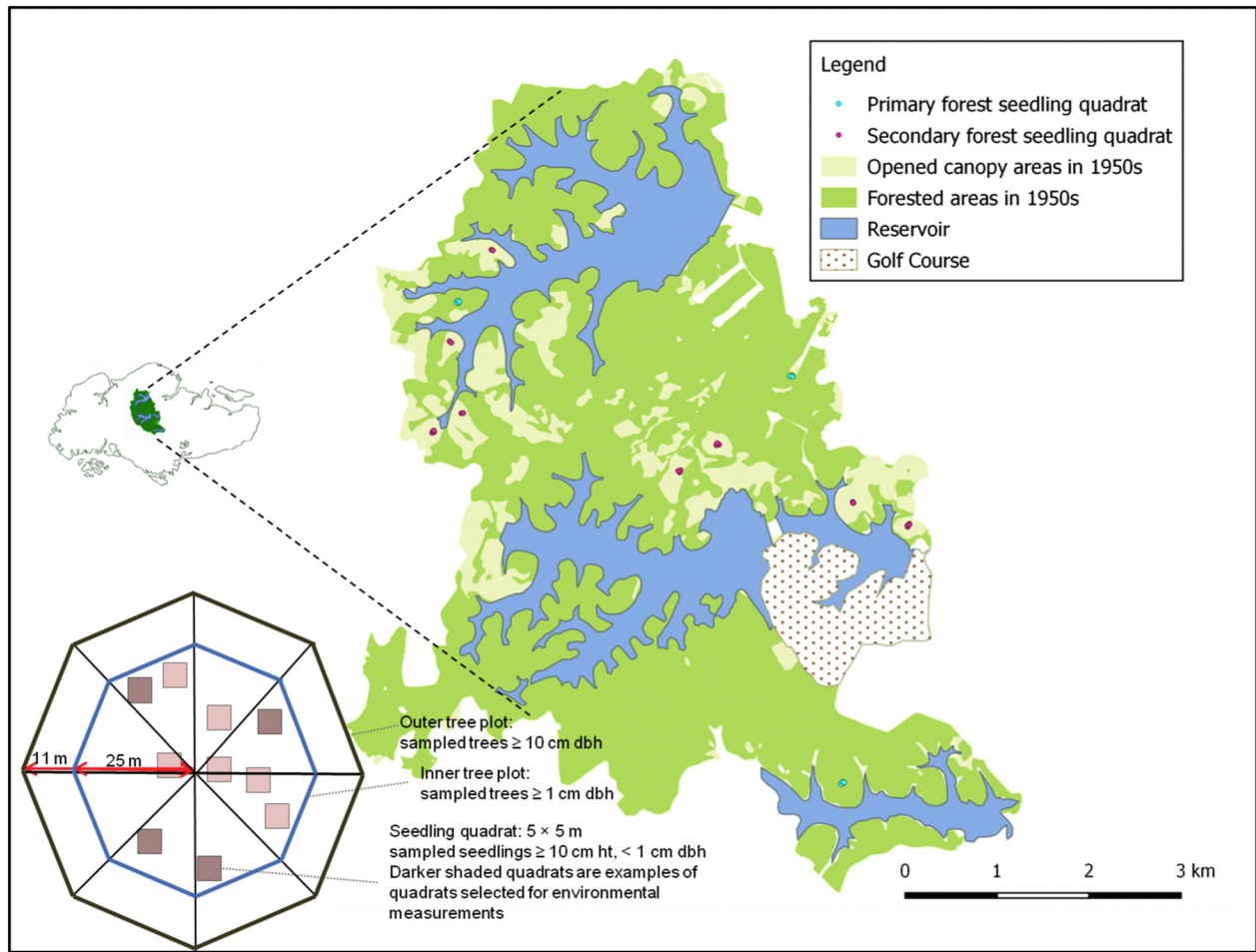
Fig. 1. Location of seedling quadrats within primary and secondary forest plots in the Central Catchment Nature Reserve, Singapore.

measured using a desktop scanner and analyzed with the pixel-counting software ImageJ (http://rsbweb.nih.gov/ij/). The leaves were oven dried for at least 72 h at 60 °C prior to measuring the dry masses.

Table 1
List of abbreviation used for traits and environmental variables.

Variable	Abbreviation	Name
Environmental	Al	Soil exchangeable aluminum
	Alsat	Soil aluminum saturation
	Ca	Soil exchangeable calcium
	CN	Soil CN ratio
	Cnopy	Canopy openness
	K	Soil exchangeable potassium
	LL	Leaf litter depth
	Mg	Soil exchangeable magnesium
	P	Soil available phosphorus (resin extracted)
	pH	Potential hydrogen
	sdV	Mean standard deviation of daily vapor pressure deficits
Plant traits[a]	BA[b]	Total basal area of plot
	CN	Leaf CN ratio
	K	Leaf potassium concentration
	LDMC	Leaf dry matter content
	LNC	Leaf nitrogen concentration
	LPC	Leaf phosphorus concentration
	Mg	Leaf magnesium concentration
	NP	NP ratio
	SLA	Specific leaf area
	Wd	Wood density/specific stem density

[a] Seedling traits end with suffix ".x" and adult traits end with suffix ".y".
[b] Plot total basal area is not a functional trait but in our MFA analysis. We included it to examine its relationship with the environmental variables.

Total carbon and total nitrogen content of the leaves were measured using a Thermo Scientific FLASH 2000 NC Analyzer. We used a Perkin Elmer Inductively Coupled Plasma to analyze for other macronutrients in the leaves, including phosphorus, magnesium, calcium and potassium. In total, leaves were collected from 55 seedling species and 27 tree species.

Using these measurements, we calculated specific leaf area (the one-sided area of the fresh leaf divided by its oven-dry mass, $mm^2\ mg^{-1}$) and leaf dry matter content (the oven-dry mass of a leaf divided by its fresh weight, $mg\ g^{-1}$). We also calculated the CN and NP stoichiometry of the leaves. Wood density data were taken from the Global Wood Density Database (Chave et al., 2009; Zanne et al., 2009). In all our analysis, species average values were used. We were not able to get leaf trait data for 15 of the 74 seedling species (due to a substantial number of single-tons and time constraints), and for these the average values of species within the genus, or the community aggregated means of that quadrat (see below) were used. These 15 species accounted on average, for 5% or less of the seedling abundance in the quadrats in which they occurred. Similarly, for three of the 27 trees species without wood density data, we used average values of species within the genus. We used species' average trait values although we recognize that intraspecific trait variation could be important for species' adaptation. However, we found that for our plots, the interspecific variation is larger than the in-traspecific variation for most adult tree traits (Table S1 and S2) and hence we decided it is appropriate to use species average trait values. As seedling leaf samples were pooled by species and plot for chemical analysis, we were only able to verify that intraspecific variation is smaller than interspecific variance for LDMC and were comparable for SLA.

All abbreviations used for the traits and environmental variables are listed in Table 1 and data of mean trait values of seedling and tree species can be found in Table S4 and S5 respectively.

2.5. Statistical analysis

Unless otherwise stated, throughout this paper, "adult traits" refer to community-weighted means (cwm) of traits of the adult trees at each plot, while "seedling traits" refer to cwm of traits of seedlings at each quadrat. Following biomass ratio theory (Grime, 1998; Garnier et al., 2004), cwm for each plot or quadrat was calculated as follow:

$$cwm = \sum_{i=1}^{n} p_i \times trait_i$$

For seedling traits, p_i was calculated based on number of individuals for each species relative to the total number of seedlings in each seedling quadrat; for adult traits, p_i was the sum of basal area of each tree species relative to the total basal area of trees in each plot. Basal area, rather than abundance was used for adult trees, as it better reflects the species' biomass, and in turn their relative contribution to the plots' functional spectrum (Garnier et al., 2004).

In our statistical analyses focused on answering Q1, to minimize the impact of small sample size and covariation between variables, we used only the environmental variables that were shown to be important from our previous work (Chua et al., 2016), namely soil phosphorus, CN ratio, exchangeable Al and Al saturation, along with soil potassium, an important plant nutrient. We included Al saturation, which is the relative quantity of Al^{3+} ions to that of other base cations (Ca^{2+}, Mg^{2+} and K^+), as it could be a more important predictor of plant responses to aluminum toxicity (Cronan and Grigal, 1995; Kinraide, 1997). We also replaced vapor pressure deficit, which was shown to be more important from previous work, with its correlate, canopy openness ($r^2 = 0.71$, $p < 0.001$), as many of the iButtons were lost or malfunctioned, and also for better comparison with most other studies that measured light, and not vapor pressure deficit. However, in our statistical analyses focused on answering Q2, as we were interested in the broad linkages between adult traits, seedling traits and the environmental variables, we included all the environmental variables mentioned above.

For Q1, due to the nested design of the seedling quadrats within the tree plots, we first applied partial mantel test to distinguish the effects of space from that of the standardized environmental variables on seedling traits. We verified that although seedling traits were significantly correlated to space ($r = 0.231^{**}$, conditioned on environmental variables), the traits were also significantly correlated to environmental variables ($r = 0.262^*$, conditioned on x- and y-coordinates). We then used a combination of the RLQ and the fourth-corner method (Dray and Legendre, 2008; ter Braak et al., 2012; Dray et al., 2014) to analyze the relationship of environmental variables and seedling traits. We carried out the analysis using relative abundance of seedlings species. RLQ is an extension of co-inertia analysis that searches simultaneously for linear combinations of variables in the species × trait matrix (Q) and the site × environmental matrix (R) that maximize covariance and weighting per site × species matrix (L). We then used the fourth corner method to test the correlation between 1) the RLQ sample scores (Axis R1/Axis R2, corresponding to the environmental gradients) and species trait (table Q); and 2) the RLQ species scores (Axis Q1/Axis Q2, corresponding to the trait variables) and the environmental variables (table R). The fourth corner method is more precise than simple regression as it explicitly tests for significant relationships between the environmental variables with species abundance, and that between species abundance and plant traits. Significances of the correlations were tested by comparing the observed test statistic to its predicted distribution under null hypotheses. Following Dray and Legendre (2008) and Dray et al. (2014), we used two null models, Model 2 and Model 4. Model 2

permutes the rows of table L (or rows of R) and tests for the non-random distribution of species with fixed traits. Model 4 permutes the columns of L (or rows of Q) and tests for the influence of traits on the species composition under given environmental conditions. The global statistical significance of the correlation is significant if the larger of the two p values from both models is lower than $\alpha = 0.05$ (ter Braak et al., 2012). The false discovery rate method (Benjamini and Hochberg, 1995) was used to adjust p values for multiple testing. We used the ade4 R package (Dray and Dufour, 2007) for both the RLQ and fourth corner method.

To answer Q2, we performed Multiple Factor Analysis (MFA) (Escofier and Pagès, 1994; Borcard et al., 2011) to explore the overall correlation among seedling traits, adult traits and environmental variables within secondary forests only. Total basal area of the plot was used as a proxy for stand structure and was also included as an adult trait. MFA is identical to a PCA performed on all three set of variables simultaneously with each of the three data subsets weighted. The RV coefficient which ranges from 0 to 1 is used to assess the correlation of the three sets of variables and is tested by permutations (Borcard et al., 2011; Josse et al., 2008). To account for the spatial nestedness of the seedling quadrats within tree plots, we permutated the blocks of seedling quadrats at the plot level.

We used the R package FactoMineR (Lê et al., 2008) for the MFA analysis.

All statistical analyses were done using R statistical software v3.3.1. (R Core Team, 2014).

3. Results

3.1. Summary of environmental conditions observed

Within the 35 seedling quadrats surveyed, the percent of canopy openness ranged from below 1% in the least disturbed primary forest to >6% under the fern foliage of the open (class O) vegetation plots. Fluctuation of vapor pressure deficits were the highest in the open plots (0.36–0.53 kPa), moderate in the secondary forests (0.35–0.16 kPa) to lowest in the primary forests (0.08–0.22 kPa). For soil nutrients among the seedling quadrats, soil resin-extractable P ranged 0.65–19.17 mg P/kg, while soil available K was 0.09–0.31 $cmol_c$/kg. Soil CN ratio ranged 16–36, Al ranged 2.70–8.80 $cmol_c$/kg and AlSat ranged 76.22–91.15%.

3.2. Q1. How do environmental factors explain the variation in seedling functional traits observed in primary and secondary forests?

Although there was a large overlap of trait range between secondary and primary forest species (Fig. 2c), we found that the environmental variables in the secondary and primary forests were significantly correlated with overall seedling traits when analyzed using species relative abundance ($p = 0.0085$ for Model 2 and $p = 0.021$ for Model 4 of the fourth-corner test). The first two RLQ axes explained 73.1% and 17.0% respectively of the cross-covariance between the seedling traits and the environmental variables (Fig. 2). The first RLQ axis captured a transition of seedling quadrats in secondary forests to seedling quadrats in primary forest of decreasing disturbance (Fig. 2a), which was also significantly correlated to seedling species richness (Spearman rho = 0.555, $p < 0.005$, Fig. S1).

Fourth-corner analyses also showed that along this secondary to primary forest axis, decreasing light quantity, exchangeable aluminum and aluminum saturation were significantly correlated to increasing seedling LNC (adjusted p = 0.05, Fig. 3a and b). The second RLQ axis corresponded to seedling quadrats with high leaf dry matter content and leaf NP ratio that were found in habitats with low soil potassium and available phosphorus (Fig. 2b and e). However this relationship was not significant.
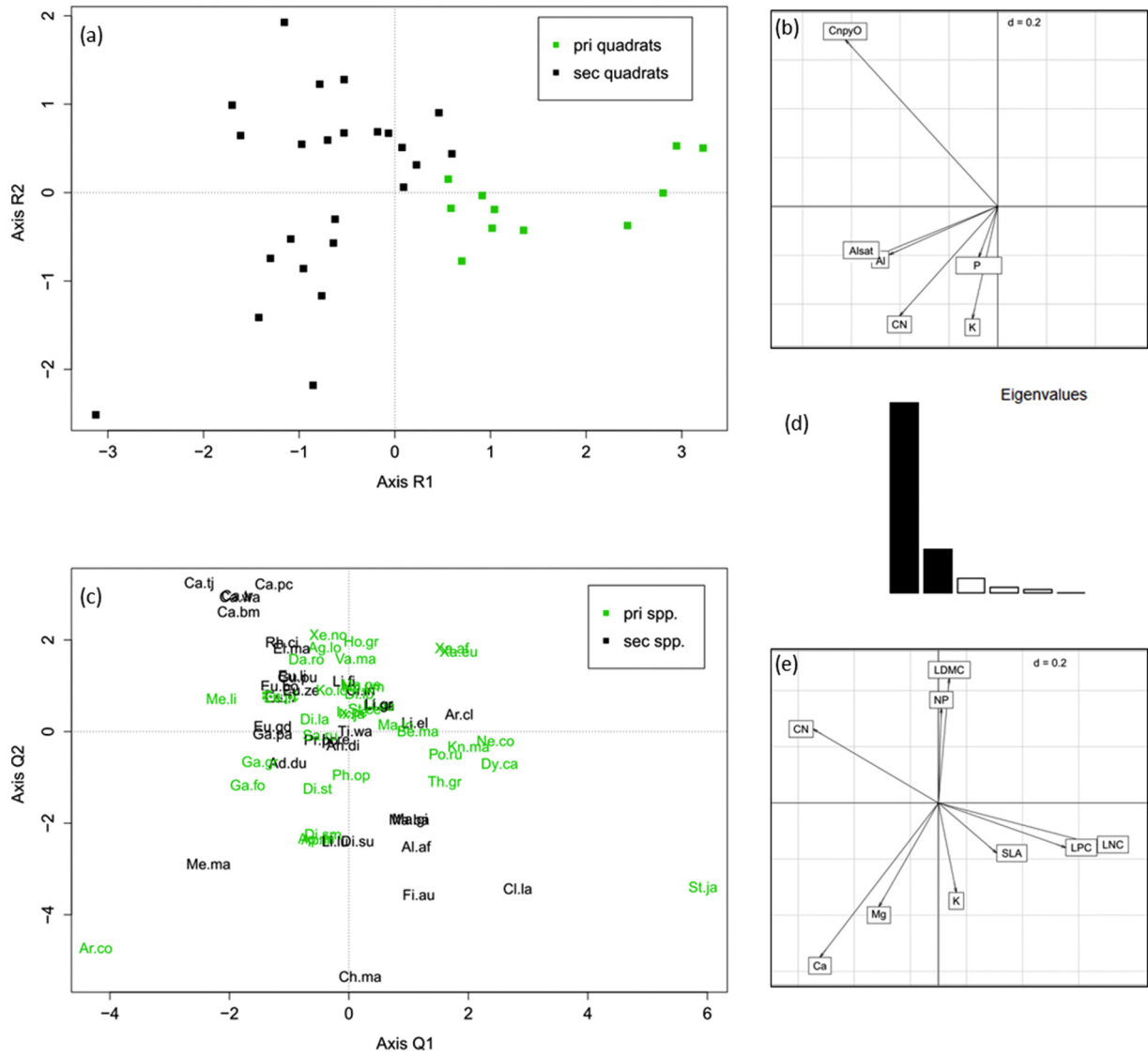
Fig. 2. Results of RLQ analysis: (a) distribution of the seedling quadrats by the R (environmental variables) row scores, (b) coefficients for the environmental variables, (c) distribution of species by Q (seedling traits) row scores, (d) the eigenvalues of the first two axes, as indicated in black. See Table S3 for species names corresponding to the species code, and (e) coefficients for traits. Note that values of "d" give the grid size for (b) and (e). The categorization to primary and secondary forest species was based on Tree Flora of Malaya Volume 1–4 (Ng, 1978, 1989; Whitmore, 1972, 1973), Wayside trees of Malaya Volume 1 and 2 (Corner, 1988) and online web resources (Slik, 2009) (Table S3).
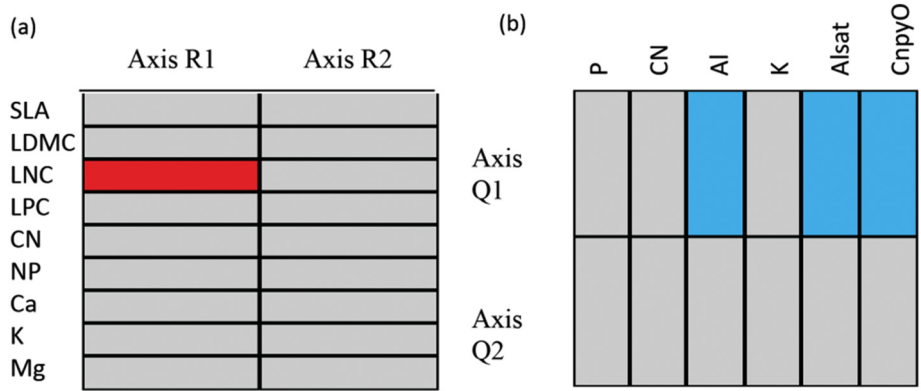

Fig. 3. Results of fourth-corner tests between (a) seedling traits and the first two RLQ axes for environmental gradients (Axis R 1/Axis R 2), (b) environmental variables and the first two RLQ axes for seedling traits (Axis Q 1 and Axis Q 2) at adjusted $p = 0.05$. Significant positive associations are represented by red cells and significant negative associations by blue cells. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

3.3. Q2. How do seedling traits, adult traits and environmental variables relate and explain variation in species richness and stem density in secondary forests?

3.3.1. MFA traits and environmental factors correlations

The MFA results showed that only the adult tree traits and the environment variables were significantly correlated (RV = 0.462, $p \leq 0.000$) (Table 2). The correlation between the seedling traits and the environmental variables was marginally significant (RV = 0.239, $p = 0.065$), while seedling traits and adult traits were not significantly correlated.

The first three axes of the MFA explained >65% of the total variance of the combined datasets (Fig. S2 and Fig. 4). The first MFA Axis (31.39%) captured a resource gradient that is characterized at one end by high Al saturation and low soil Ca, Mg, light availability and less fluctuating vapor pressure deficits. Interestingly, the low resource axis correlated to *increasing leaf Mg, K, P and N in both seedling and adult trees*, and to increasing adult SLA and leaf Ca (Fig. S2). Although seedling traits had borderline significant correlation with environmental variables, the positive relationship between the seedling leaf nutrients with Al saturation and exchangeable Al (see second MFA axis) (Fig. S2a and 4b) is interesting as it differed from the RLQ results obtained using data from both primary and secondary plots. The second MFA Axis (20.28%) captured *opposing responses* of seedlings and adult traits to increasing exchangeable Al, leaf litter depth and soil CN and K – seedling SLA and leaf foliar nutrients increased, while leaf CN deceased; conversely adult SLA, foliar nitrogen and calcium concentrations decreased, while leaf CN increased. The third MFA axis (13.85%) captured the correlation of decreasing soil pH and CN, with increasing soil P, Mg, exchangeable Al, adult wood density and plot basal area (Fig. S2 and Fig. 4). Seedling SLA, adult trees CN ratio, soil CN ratio and leaf litter depth were also positively correlated to one another. Simple bivariate plots showed that of these correlations, the adult tree CN ratio was strongly correlated to leaf litter depth (Pearson $r^2 = 0.67$, $p < 0.001$), which in turn was strongly correlated to soil CN ratio (Pearson $r^2 = 0.54$, $p < 0.01$) (See Fig. S3).

3.3.2. Species richness and density association

Our results show a trend that seedling species richness was lower at quadrats with higher seedling foliar nutrients (Fig. 4a) Seedling species richness was also negatively correlated with high soil CN ratio, high leaf litter depth, high seedling SLA and low stand basal area (Fig. 4c). Seedling quadrats with high Al saturation and low available phosphorus also tend to have the lowest species richness (4b). Due to the success of individual species, trends in seedling stem density deviated slightly from that of species richness described above (Fig. 4d, e and f). We conducted the same analysis using sapling species richness and found similar results (Fig. S4).

4. Discussion

We investigated the extent to which plant functional traits and environmental variables explained the varied recovery status of secondary forests in Singapore. Due to the plots' similar age, we did not attempt to trace trends over succession, but rather identified potential environmental filters and corresponding plant traits that explained the observed differences in the secondary forest plots. As determined by the

partial mantel test, both space, which is indicative of dispersal effects, as well as the regeneration environment, play a role in structuring the seedling trait spaces. Overall, we found indications that plants are likely more nutrient constrained in the secondary forests as compared to the primary forests, which differ from results of other studies (Boukili and Chazdon, 2017). Increase in light availability and soil aluminum levels from the primary to secondary forest plots were the most important environmental factors that differentiated plant functional spaces. These factors were significantly correlated to decreasing seedlings LNC. Within the secondary forests, low soil nutrients and high aluminum saturation best explained the poor forest recovery, measured in terms of species richness and stem density of seedlings and saplings. In contrast the measured seedling traits did not exhibit any trend with measures of structure and diversity. This differed from the overall trends we found when analyzing both the primary and secondary forest plots, and can be attributed to the ability of the few specialists of degraded soil to accrue surprisingly high foliar nutrients despite the high aluminum saturation and low nutrients soils.

4.1. Q1. Environmental variables structuring traits in secondary and primary forests

4.1.1. Role of light and soil aluminum

Previous studies, largely from the Neotropics, have emphasized that decreasing light availability explains the transition from leaf traits focused on resource acquisition to leaf traits focused on resource conservation over succession in the wet tropical forests (Boukili and Chazdon, 2017; Craven et al., 2015; Lohbeck et al., 2015, 2013). However, few studies have measured soil nutrients to determine their relative importance on plant functional strategies in regenerating forests. We quantified light conditions and soil nutrients in our study plots and found that >60 years after abandonment, both light and soil fertility together explained the shifts in plants functional traits from poorly recovering secondary forests with low species richness to the least disturbed primary forests with high species richness. Among the soil factors, we found that increasing aluminum levels from the primary to secondary forests could explain in part the poor species richness in secondary forests. Phytotoxic aluminum is prevalent in acidic tropical soils, as low pH increases the dissolution of aluminum, which primarily affects root functions and uptake of nutrients (Rengel, 2004; Kochian et al., 2005). Interestingly, research has found that native tropical rainforest plants on acidic soils could be Al-tolerant, Al-accumulator or even Al-stimulated in terms of growth (Masunaga et al., 1998; Osaki et al., 1997; Watanabe et al., 2006). However, Singapore forests soils are inherently very acidic, with pH 3.86–4.00 in our primary forest soils and pH 3.68–4.16 in the secondary forest soils, compared to soil pH largely >4.0 in other regional studies (Hamzah et al., 2009; Jamaluddin, 2013; Proctor et al., 1983; Zaidey et al., 2010). When the canopy cover is poor, as in the case of secondary forests, there could be increased susceptibility of soil base cations being rain-leached and hence increases the aluminum saturation in these very acidic soils. This proposition is supported by the positive correlation of aluminum levels with canopy opening in seedling quadrats (Fig. 2 and Fig. 3).

4.1.2. Secondary forests were more nutrient constrained

We also found evidence that seedlings in secondary forests were more nutrient constrained than that in the primary forests. This is supported by an *increase* of seedling LNC and (to a lesser extent) LPC from the poorer recovering secondary forests to the primary forests, while quadrats with high seedling LDMC and foliar NP ratio (more P than N limited) were equally prevalent in both primary and secondary forests (Fig. 2). This result differs from a study in Costa Rica, which found prevalence of resource acquisitive traits in secondary forests (oldest being 42 years old) and conservative traits in old growth forests (Boukili and Chazdon, 2017). It also contradicts findings of shifts in resource acquisitive to resource conservation traits over succession (Craven et al.,

Table 2
Correlations between seedling traits, adult traits and environmental variables in the secondary forests using Multiple Factor Analysis (MFA) Upper diagonals contained permutational-based *p* values. Lower diagonals contained RV coefficients.

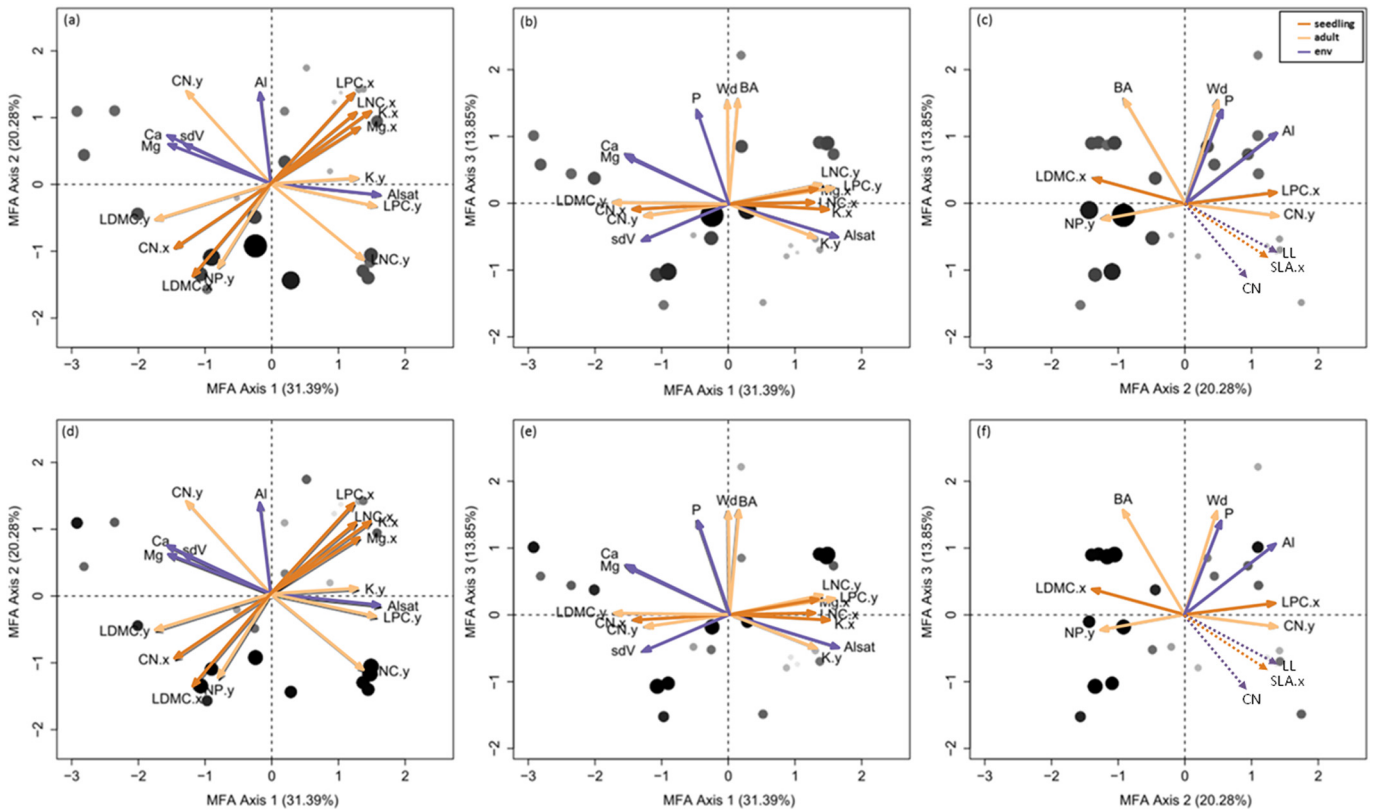|	Seedling traits	Adult traits	Env. variables
Seedling traits		0.098	0.065
Adult traits	0.387		0.000
Env. variables	0.239	0.462	

Fig. 4. Global PCA biplot showing the distribution of seedling quadrats by the traits and environmental variables, as correlated to the first thre MFA axes. Top three panels (a – c) show point size scaled by seedling species richness and the bottom three panels (d – e) and by seedling density (increase size and shade correspond to higher species richness/density). Only variables that were significantly related to the respective MFA axis at $p = 0.001$ and selected variables that were significant at $p = 0.05$ (dotted arrows) are shown here. Refer to Fig. S2 for the location of other variables. Seedling traits end with suffix ".x" and adult traits end with suffix ".y". Abbreviations used are listed in Table 1.

2015; Lohbeck et al., 2015, 2013). These studies posit that the growing biomass depletes nutrients from the soil as the forests recover. However in our study, soil available P was similar in primary forests and secondary forest plots and did not correlate to plant traits, possibly because soil available P was inherently very low in our primary forests (this study and Turner, 2012). Instead, aluminum levels strongly correlated to decreasing seedling LNC, and to a lesser extend seedling LPC, which suggests that aluminum toxicity could be important, although future studies would be needed for verification. Increasing soil CN, which could slow microbial breakdown of organic matter and nutrient returns (Robertson and Groffman, 2007 and see below), was also weakly correlated with increasing seedling LNC.

4.2. Q2. Relating traits-environmental variables to recovery of secondary forests

Despite deliberate measurement of the regeneration environment, the seedling-environmental relationship, was unexpectedly only marginally significant (Table 2). Part of the reason could be that we only had 8 secondary forest tree plots, and the seedling quadrats were permutated at the plot level, which constrained the permutational-based p in the MFA analysis. We also postulated that the opposing trait response to light and soil factors was an important cause, as elaborated below. The correlation between the adult traits and the environment was the strongest probably due to the strong influence of soil on leaf traits and biomass growth, as represented by basal area and wood density. Tree biomass had less influence on canopy cover and sdV, likely because thick leaf litter had a stronger influence on the microclimate at some sites, as discussed below. Not unexpectedly, the seedling-adult traits were not correlated probably because of changing adult and seedling species composition as the forest regenerates, and that seedlings

and adult plants were responding to different aboveground environment (e.g. light).

4.2.1. Degraded soil specialists accrued high nutrients – role of aluminum accumulators

The most intriguing result is that seedling (albeit of marginal correlation with the environmental variables) and adult foliar nutrients increased with increasing Al saturation, which opposed trends found in the analysis using primary and secondary plots data. A recent study by Russell et al. (2017) proposed a mechanism for how certain secondary forest species on infertile soils could accrue cations in their biomass through accumulating Al, which increased soil pH to liberate previously occluded soil cations. This mechanism is likely to occur in our most infertile secondary forests. For example, *Melastoma malabathricum,* a common pioneer species of degraded soils in Singapore (Corlett, 1991; Sim et al., 1992), has been found to be a hyperaccumulator of aluminum. It was found that aluminum stimulated its uptake of phosphorus and hence growth (Osaki et al., 1997), as well as reduced iron toxicity to the plant (Watanabe et al., 2001). *M. malabathricum* seedlings are also found in relatively high abundance in two of our sites with high aluminum saturation. Other notably species in our plots that could assimilate relatively high foliar N and P under comparatively low soil nutrients conditions were *Dillenia suffruticosa* and *Champereia manillana*, which were found in high density in plots that have high aluminum saturation and low phosphorus. However, fewer species could thrive on soil with high aluminum levels, as indicated by the largely poor species richness of seedling quadrats in plots with both high exchangeable Al and Al saturation (Fig. 4b and Fig. S2), although those species could be in high abundance (Fig. 4d). There is also some indication that high soil P could improve seedling species richness despite high exchangeable Al

(Fig. 4c). Similar trends were observed even in the sapling stage (Fig. S4.).

4.2.2. Poor forest recovery with high soil CN and thick leaf litter

Our results also shows that poorer seedling species richness and density correlated with increasing soil CN ratio and leaf litter depth (Fig. 4), which agrees with findings from other studies (Facelli and Pickett, 1991; Metcalfe and Grubb, 1997; Goldsmith et al., 2011, but see Baker and Murray, 2010). In our study, the correspondingly high soil and adult CN ratios (Fig. S2 and Fig. 4c) suggests that slow decomposition has probably led to the thick leaf litter. The soil CN ratios of our secondary forests (19.70–32.08) were much higher than other humid rainforests (Kauffman et al., 1998), including primary and regenerating Dipterocarp forests (See Table 5 in Sim et al., 1992, Abdu et al., 2007; Hamzah et al., 2009; Zaidey et al., 2010). High soil CN ratio slows microbial breakdown of organic matter and nutrient returns (Robertson and Groffman, 2007); while nutrient efficient plants also tend to produce leaves and leaf litter with high CN ratio that are slow to decompose. Past research had shown that litter decomposition rates in secondary forests could be similar to primary forests (Barlow et al., 2007) or slower (Mesquita et al., 1998; Parsons and Congdon, 2008), due to different soil conditions and regenerating species. The corresponding low basal area to high soil CN ratio is also indicative of slower forest growth at these low nutrient sites. Interestingly, the seedling CN ratio showed a weak decreasing relationship with increasing soil CN ratios (Fig. S2). This is likely because seedlings were responding to low light conditions under the thick leaf litter by producing leaves with larger leaf area relative to weight, as indicated by the correlation of seedling SLA with leaf litter depth (Fig. 4c). Increase in SLA would usually lead to a decrease in leaf CN and LDMC in order to minimize construction cost. It is likely that few species could optimize leaf construction for both low light under the leaf litter and low nutrients environment, leading to the observed low species richness at those sites (Fig. 4c).

5. Conclusions

Aluminum saturation appears to play a larger role than previously suspected in tropical succession on acidic, degraded soil. Although tropical plants have evolved on acidic and high aluminum soils, we found that 60 years after regenerating from intensive agricultural activities, poor forest regrowth was correlated with high soil aluminum levels, which appears to affect nutrient uptake in plants, and result in lower leaf nitrogen content in the poorly recovering forests as compared to primary forests. Succession is thus slowed by a positive feedback loop of slower nutrient returns from slow decaying litter and further recruitment of nutrient conserving species. Low nutrients and high aluminum saturation were negatively correlated with species richness recovery in the secondary forests. Resource conserving traits are part of a set of co-occurring life history traits that include slower plant growth and greater longevity (Poorter, 2007; Westoby et al., 2002; Wright et al., 2004). This would lead to slower species replacement and hence slower rates of succession, which explains the slow recovery seen in Singapore secondary forests. Future studies on transplanting primary forest seedlings to the degraded secondary forests would help to elucidate the extent that soil conditions, vs. dispersal and biotic competition, limit the establishment of primary forest species. Finally, our research shows the importance of measuring environmental variables along with the plant functional traits to elucidate possible mechanisms driving forest succession, as plant functional traits reflect the strategies of successful seedlings and the selected traits might not necessarily relate to the overall forest recovery. In our case, soil nutrients were not a constraint to the successful seedlings at infertile sites, but soil nutrients did hinder the recovery of species richness.

Data accessibility

Plant leaf trait data will be deposited with the Plant Trait Database.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.scitotenv.2018.05.397.

References

Abdu, A., Tanaka, S., Jusop, S., Ibrahim, Z., Hattori, D., Majid, N.M., Sakurai, K., 2007. Soil characteristics under rehabilitation of degraded forestland in Perak, Peninsular Malaysia. Pedol. Jpn. 51, 76–88.

Allié, E., Pélissier, R., Engel, J., Petronelli, P., Freycon, V., Deblauwe, V., Soucémarianadin, L., Weigel, J., Baraloto, C., 2015. Pervasive local-scale tree-soil habitat association in a tropical forest community. PLoS One 10, e0141488. https://doi.org/10.1371/journal.pone.0141488.

Asner, G.P., Rudel, T.K., Aide, T.M., Defries, R., Emerson, R., 2009. A contemporary assessment of change in humid tropical forests. Conserv. Biol. 23:1386–1395. https://doi.org/10.1523-1739.2009.01333.x.

Baker, A.C., Murray, B.R., 2010. Relationships between leaf-litter traits and the emergence and early growth of invasive Pinus radiata seedlings: leaf litter and pine seedlings. Weed Res. 50:586–596. https://doi.org/10.1111/j.1365-3180.2010.00805.x.

Bakker, M.A., Carreño-Rocabado, G., Poorter, L., 2011. Leaf economics traits predict litter decomposition of tropical plants and differ among land use types. Funct. Ecol. 25: 473–483. https://doi.org/10.1111/j.1365-2435.2010.01802.x.

Baraloto, C., Paine, C.E.T., Poorter, L., Beauchene, J., Bonal, D., Domenach, A.-M., Hérault, B., Patiño, S., Roggy, J.-C., Chave, J., 2010. Decoupled leaf and stem economics in rain forest trees. Ecol. Lett. 13:1338–1347. https://doi.org/10.1111/j.1461-0248.2010.01517.x.

Barlow, J., Gardner, T.A., Ferreira, L.V., Peres, C.A., 2007. Litter fall and decomposition in primary, secondary and plantation forests in the Brazilian Amazon. For. Ecol. Manag. 247:91–97. https://doi.org/10.1016/j.foreco.2007.04.017.

Bazzaz, F.A., Pickett, S.T.A., 1980. Physiological ecology of tropical succession: a comparative review. Annu. Rev. Ecol. Syst. 11:287–310. https://doi.org/10.1146/annurev.es.11.110180.001443.

Becknell, J.M., Powers, J.S., 2014. Stand age and soils as drivers of plant functional traits and aboveground biomass in secondary tropical dry forest. Can. J. For. Res. 44: 604–613. https://doi.org/10.1139/cjfr-2013-0331.

Benjamini, Y., Hochberg, Y., 1995. Controlling the false discovery rate: a practical and powerful approach to multiple testing. J. R. Stat. Soc. Ser. B Methodol. 57, 289–300.

Blundo, C., Malizia, L.R., González-Espinosa, M., 2015. Distribution of functional traits in subtropical trees across environmental and forest use gradients. Acta Oecol. 69: 96–104. https://doi.org/10.1016/j.actao.2015.09.008.

Borcard, D., Gillet, F., Legendre, P., 2011. Numerical Ecology with R. Springer Science & Business Media.

Boukili, V.K., Chazdon, R.L., 2017. Environmental filtering, local site factors and landscape context drive changes in functional trait composition during tropical forest succession. Perspect. Plant Ecol. Evol. Syst. 24:37–47. https://doi.org/10.1016/j.ppees.2016.11.003.

Busing, R.T., Brokaw, N., 2002. Tree species diversity in temperate and tropical forest gaps: the role of lottery recruitment. Folia Geobot. 37, 33–43.

Cadotte, M.W., Tucker, C.M., 2017. Should environmental filtering be abandoned? Trends Ecol. Evol. 32:429–437. https://doi.org/10.1016/j.tree.2017.03.004.

Chao, K.-J., Phillips, O.L., Gloor, E., Monteagudo, A., Torres-Lezama, A., Martínez, R.V., 2008. Growth and wood density predict tree mortality in Amazon forests. J. Ecol. 96: 281–292. https://doi.org/10.1111/j.1365-2745.2007.01343.x.

Chave, J., Coomes, D., Jansen, S., Lewis, S.L., Swenson, N.G., Zanne, A.E., 2009. Towards a worldwide wood economics spectrum. Ecol. Lett. 12:351–366. https://doi.org/10.1111/j.1461-0248.2009.01285.x.

Chazdon, R.L., 2008. Chance and determinism in tropical forest succession. Tropical Forest Community Ecology. Wiley-Blackwell Publishing, Oxford, pp. 384–408.

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. Manag. 308:153–160. https://doi.org/10.1016/j.foreco.2013.07.053.

Chua, S.C., Ramage, B.S., Potts, M.D., 2016. Soil degradation and feedback processes affect long-term recovery of tropical secondary forests. J. Veg. Sci. 27:800–811. https://doi.org/10.1111/jvs.12406.

Claire, Fortunel, Fine Paul, V.A., Christopher, Baraloto, Jim, Dalling, 2012. Leaf, stem and root tissue strategies across 758 Neotropical tree species. Funct. Ecol. 26: 1153–1161. https://doi.org/10.1111/j.1365-2435.2012.02020.x.

Comita, L.S., Hubbell, S.P., 2009. Local neighborhood and species' shade tolerance influence survival in a diverse seedling bank. Ecology 90, 328–334.

Comita, L.S., Thompson, J., Uriarte, M., Jonckheere, I., Canham, C.D., Zimmerman, J.K., 2010. Interactive effects of land use history and natural disturbance on seedling dynamics in a subtropical forest. Ecol. Appl. 20, 1270–1284.

Corlett, R.T., 1991. Plant succession on degraded land in Singapore. For. Sci. 4, 151–161.

Corlett, R.T., 1992. The ecological transformation of Singapore, 1819–1990. J. Biogeogr. 19: 411. https://doi.org/10.2307/2845569.

Corlett, R.T., 1995. Tropical secondary forests. Prog. Phys. Geogr. 19:159–172. https://doi.org/10.1177/030913339501900201.

Cornelissen, J.H.C., Lavorel, S., Garnier, E., Díaz, S., Buchmann, N., Gurvich, D.E., Reich, P.B., ter Steege, H., Morgan, H.D., van der Heijden, M.G.A., Pausas, J.G., Poorter, H., 2003. A handbook of protocols for standardised and easy measurement of plant functional traits worldwide. Aust. J. Bot. 51, 335–380.

Corner, E.J.H., 1988. Wayside Trees of Malaya: In Two Volumes. 3rd ed. The Malayan Nature Society, Kuala Lumpur.

Cornwell, W.K., Cornelissen, J.H.C., Amatangelo, K., Dorrepaal, E., Eviner, V.T., Godoy, O., Hobbie, S.E., Hoorens, B., Kurokawa, H., Pérez-Harguindeguy, N., Quested, H.M., Santiago, L.S., Wardle, D.A., Wright, I.J., Aerts, R., Allison, S.D., van Bodegom, P., Brovkin, V., Chatain, A., Callaghan, T.V., Díaz, S., Garnier, E., Gurvich, D.E., Kazakou, E., Klein, J.A., Read, J., Reich, P.B., Soudzilovskaia, N.A., Vaieretti, M.V., Westoby, M., 2008. Plant species traits are the predominant control on litter decomposition rates within biomes worldwide. Ecol. Lett. 11:1065–1071. https://doi.org/10.1111/j.1461-0248.2008.01219.x.

Craven, D., Hall, J.S., Berlyn, G.P., Ashton, M.S., van Breugel, M., 2015. Changing gears during succession: shifting functional strategies in young tropical secondary forests. Oecologia 179:293–305. https://doi.org/10.1007/s00442-015-3339-x.

Cronan, C.S., Grigal, D.F., 1995. Use of calcium/aluminum ratios as indicators of stress in forest ecosystems. J. Environ. Qual. 24:209–226. https://doi.org/10.2134/jeq1995.00472425002400020002x.

Dray, S., Dufour, A.-B., 2007. The ade4 package: implementing the duality diagram for ecologists. J. Stat. Softw. 22, 1–20.

Dray, S., Legendre, P., 2008. Testing the species traits-environment relationships: the fourth-corner problem revisited. Ecology 89, 3400–3412.

Dray, S., Choler, P., Dolédec, S., Peres-Neto, P.R., Thuiller, W., Pavoine, S., ter Braak, C.J.F., 2014. Combining the fourth-corner and the RLQ methods for assessing trait responses to environmental variation. Ecology 95:14–21. https://doi.org/10.1890/13-0196.1.

Escofier, B., Pagès, J., 1994. Multiple factor analysis (AFMULT package). Comput. Stat. Data Anal. 18:121–140. https://doi.org/10.1016/0167-9473(94)90135-X.

Facelli, J.M., Pickett, S.T.A., 1991. Plant litter: its dynamics and effects on plant community structure. Bot. Rev. 57, 1–32.

Feldpausch, T.R., Rondon, M.A., Fernandes, E.C., Riha, S.J., Wandelli, E., 2004. Carbon and nutrient accumulation in secondary forests regenerating on pastures in Central Amazonia. Ecol. Appl. 14, 164–176.

Finegan, B., 1996. Pattern and process in neotropical secondary rain forests: the first 100 years of succession. Trends Ecol. Evol. 11:119–124. https://doi.org/10.1016/0169-5347(96)81090-1.

Fortunel, C., Paine, C.E.T., Fine, P.V.A., Kraft, N.J.B., Baraloto, C., 2014. Environmental factors predict community functional composition in Amazonian forests. J. Ecol. 102: 145–155. https://doi.org/10.1111/1365-2745.12160.

Frazer, G.W., Trofymow, J.A., Lertzman, K.P., 2000. Canopy openness and leaf area in chronosequences of coastal temperate rainforests. Can. J. For. Res. 30:239–256. https://doi.org/10.1139/x99-201.

Garnier, E., Cortez, J., Billès, G., Navas, M.-L., Roumet, C., Debussche, M., Laurent, G., Blanchard, A., Aubry, D., Bellmann, A., 2004. Plant functional markers capture ecosystem properties during secondary succession. Ecology 85, 2630–2637.

Gastauer, M., Meira-Neto, J.A.A., 2014. Interactions, environmental sorting and chance: phylostructure of a tropical forest assembly. Folia Geobot. 49:443–459. https://doi.org/10.1007/s12224-013-9181-1.

Gitay, H., Noble, I.R., Connell, J.H., 1999. Deriving functional types for rain-forest trees. J. Veg. Sci. 10:641–650. https://doi.org/10.2307/3237079.

Goldsmith, G.R., Comita, L.S., Chua, S.C., 2011. Evidence for arrested succession within a tropical forest fragment in Singapore. J. Trop. Ecol. 27:323–326. https://doi.org/10.1017/S0266467411000010.

Grime, J.P., 1998. Benefits of plant diversity to ecosystems: immediate, filter and founder effects. J. Ecol. 86, 902–910.

Grubb, P.J., 1977. The maintenance of species-richness in plant communities: the importance of the regeneration niche. Biol. Rev. 52:107–145. https://doi.org/10.1111/j.1469-185X.1977.tb01347.x.

Guariguata, M.R., Ostertag, R., 2001. Neotropical secondary forest succession: changes in structural and functional characteristics. For. Ecol. Manag. 148, 185–206.

Hamzah, M.Z., Arifin, A., Zaidey, A.K., Azirim, A.N., Zahari, I., Hazandy, A.H., Affendy, H., Wasli, M.E., Shamshuddi, J., Muhamad, M.N., 2009. Characterizing soil nutrient status and growth performance of planted dipterocap and non-dipterocarp species on degraded forest land in Peninsular Malaysia. J. Appl. Sci. 9:4215–4223. https://doi.org/10.3923/jas.2009.4215.4223.

Hughes, R.F., Kauffman, J.B., Jaramillo, V.J., 1999. Biomass, carbon, and nutrient dynamics of secondary forests in a humid tropical region of México. Ecology 80:1892–1907. https://doi.org/10.2307/176667.

Huston, M., Smith, T., 1987. Plant succession: life history and competition. Am. Nat. 130, 168–198.

Ives, D.W., 1977. Soils of the Republic of Singapore. New Zealand Soil Bureau. Dept. of Scientific and Industrial Research, Wellington, N.Z.

Jager, M.M., Richardson, S.J., Bellingham, P.J., Clearwater, M.J., Laughlin, D.C., 2015. Soil fertility induces coordinated responses of multiple independent functional traits. J. Ecol. 103:374–385. https://doi.org/10.1111/1365-2745.12366.

Jamaluddin, 2013. Assessing soil fertility status of rehabilitatd degraded tropical rainforest. Am. J. Environ. Sci. 9:280–291. https://doi.org/10.3844/ajessp.2013.280.291.

Johnson, C.M., Vieira, I.C.G., Zarin, D.J., Frizano, J., Johnson, A.H., 2001. Carbon and nutrient storage in primary and secondary forests in eastern Amazônia. For. Ecol. Manag. 147: 245–252. https://doi.org/10.1016/S0378-1127(00)00466-7.

Josse, J., Pagès, J., Husson, F., 2008. Testing the significance of the RV coefficient. Comput. Stat. Data Anal. 53:82–91. https://doi.org/10.1016/j.csda.2008.06.012.

Kauffman, S., Sombroek, W., Mantel, S., 1998. Soils of rainforests characterization and major constraints of dominant forest soils in the humid tropics. Soils of Tropical Forest Ecosystems. Springer, Berlin, Heidelberg:pp. 9–20 https://doi.org/10.1007/978-3-662-03649-5_1.

King, D.A., Davies, S.J., Tan, S., Noor, N.S.M., 2006. The role of wood density and stem support costs in the growth and mortality of tropical trees. J. Ecol. 94:670–680. https://doi.org/10.1111/j.1365-2745.2006.01112.x.

Kinraide, T.B., 1997. Reconsidering the rhizotoxicity of hydroxyl, sulphate, and fluoride complexes of aluminium. J. Exp. Bot. 48:1115–1124. https://doi.org/10.1093/jxb/48.5.1115.

Kochian, L.V., Piñeros, M.A., Hoekenga, O.A., 2005. The physiology, genetics and molecular biology of plant aluminum resistance and toxicity. Plant Soil 274:175–195. https://doi.org/10.1007/s11104-004-1158-7.

Kraft, N.J.B., Adler, P.B., Godoy, O., James, E.C., Fuller, S., Levine, J.M., 2015. Community assembly, coexistence and the environmental filtering metaphor. Funct. Ecol. 29: 592–599. https://doi.org/10.1111/1365-2435.12345.

Lasky, J.R., Uriarte, M., Boukili, V.K., Chazdon, R.L., 2014. Trait-mediated assembly processes predict successional changes in community diversity of tropical forests. Proc. Natl. Acad. Sci. U. S. A. 111, 5616–5621.

Lavorel, S., Garnier, E., 2002. Predicting changes in community composition and ecosystem functioning from plant traits: revisiting the holy grail. Funct. Ecol. 16:545–556. https://doi.org/10.1046/j.1365-2435.2002.00664.x.

Lavorel, S., McIntyre, S., Landsberg, J., Forbes, T.D., 1997. Plant functional classifications: from general groups to specific groups based on response to disturbance. Trees 12, 474–478.

Lê, S., Josse, J., Husson, F., 2008. FactoMineR: a package for multivariate analysis. J. Stat. Softw. 25:1–18. https://doi.org/10.18637/jss.v025.i01.

Lebrija-Trejos, E., Pérez-García, E.A., Meave, J.A., Bongers, F., Poorter, L., 2010. Functional traits and environmental filtering drive community assembly in a species-rich tropical system. Ecology 91, 386–398.

Letcher, S.G., Chazdon, R.L., 2009. Rapid recovery of biomass, species richness, and species composition in a forest chronosequence in northeastern Costa Rica. Biotropica 41, 608–617.

Lohbeck, M., Poorter, L., Lebrija-Trejos, E., Martinez-Ramos, M., Meave, J.A., Paz, H., Perez-Garcia, E.A., Eunice Romero-Perez, I., Tauro, A., Bongers, F., 2013. Successional changes in functional composition contrast for dry and wet tropical forest. Ecology 94, 1211–1216.

Lohbeck, M., Lebrija-Trejos, E., Martínez-Ramos, M., Meave, J.A., Poorter, L., Bongers, F., 2015. Functional trait strategies of trees in dry and wet tropical forests are similar but differ in their consequences for succession. PLoS One 10, e0123741. https://doi.org/10.1371/journal.pone.0123741.

Lu, D., Moran, E., Mausel, P., 2002. Linking Amazonian secondary succession forest growth to soil properties. Land Degrad. Dev. 13:331–343. https://doi.org/10.1002/ldr.516.

Masunaga, T., Kubota, D., Hotta, M., Wakatsuki, T., 1998. Mineral composition of leaves and bark in aluminum accumulators in a tropical rain forest in Indonesia. Soil Sci. Plant Nutr. 44:347–358. https://doi.org/10.1080/00380768.1998.10414456.

Mesquita, R. de C.G., Workman, S.W., Neely, C.L., 1998. Slow litter decomposition in a Cecropia-dominated secondary forest of central Amazonia. Soil Biol. Biochem. 30: 167–175. https://doi.org/10.1016/S0038-0717(97)00105-3.

Metcalfe, D.J., Grubb, P.J., 1997. The responses to shade of seedlings of very small-seeded tree and shrub species from tropical rain forest in Singapore. Funct. Ecol. 11:215–221. https://doi.org/10.1046/j.1365-2435.1997.00070.x.

Moran, E.F., Brondizio, E.S., Tucker, J.M., da Silva-Forsberg, M.C., McCracken, S., Falesi, I., 2000. Effects of soil fertility and land-use on forest succession in Amazônia. For. Ecol. Manag. 139:93–108. https://doi.org/10.1016/S0378-1127(99)00337-0.

Murphy, S.J., Wiegand, T., Comita, L.S., 2017. Distance-dependent seedling mortality and long-term spacing dynamics in a neotropical forest community. Ecol. Lett. 20: 1469–1478. https://doi.org/10.1111/ele.12856.

Ng, F., 1978. Tree Flora of Malaya: A Manual for Foresters. Vol. 3. Longman Malaysia, Kuala Lumpur.

Ng, F., 1989. Tree Flora of Malaya: A Manual for Foresters. Vol. 4. Longman Malaysia, Kuala Lumpur.

Odum, E.P., 1969. The strategy of ecosystem development. Science 164:262–270. https://doi.org/10.1126/science.164.3877.262.

Osaki, M., Watanabe, T., Tadano, T., 1997. Beneficial effect of aluminum on growth of plants adapted to low pH soils. Soil Sci. Plant Nutr. 43:551–563. https://doi.org/10.1080/00380768.1997.10414782.

Parsons, S.A., Congdon, R.A., 2008. Plant litter decomposition and nutrient cycling in North Queensland tropical rain-forest communities of differing successional status. J. Trop. Ecol. 24. https://doi.org/10.1017/S0266467408004963.

Peres-Neto, P.R., Dray, S., ter Braak, C.J.F., 2017. Linking trait variation to the environment: critical issues with community-weighted mean correlation resolved by the fourth-corner approach. Ecography 40:806–816. https://doi.org/10.1111/ecog.02302.

Peterson, C.J., Carson, W.P., 2008. Processes constraining woody species succession on abandoned pastures in the tropics: on the relevance of temperate models of

succession. Tropical Forest Community Ecology. Wiley-Blackwell Publishing, Oxford, pp. 384–408.

Pickett, S.T.A., Collins, S.L., 1987. Models, mechanisms and pathways of succession. N. Y. Bot. Gard. 53, 335–371.

Poorter, L., 2007. Are species adapted to their regeneration niche, adult niche, or both? Am. Nat. 169, 433–442.

Poorter, L., Bongers, F., Aide, T.M., Almeyda Zambrano, A.M., Balvanera, P., Becknell, J.M., Boukili, V., Brancalion, P.H.S., Broadbent, E.N., Chazdon, R.L., Craven, D., de Almeida-Cortez, J.S., Cabral, G.A.L., de Jong, B.H.J., Denslow, J.S., Dent, D.H., Dewalt, S.J., Dupuy, J.M., Durán, S.M., Espírito-Santo, M.M., Fandino, M.C., César, R.G., Hall, J.S., Hernandez-Stefanoni, J.L., Jakovac, C.C., Junqueira, A.B., Kennard, D., Letcher, S.G., Licona, J.-C., Lohbeck, M., Marín-Spiotta, E., Martínez-Ramos, M., Massoca, P., Meave, J.A., Mesquita, R., Mora, F., Muñoz, R., Muscarella, R., Nunes, Y.R.F., Ochoa-Gaona, S., de Oliveira, A.A., Orihuela-Belmonte, E., Peña-Claros, M., Pérez-García, E.A., Piotto, D., Powers, J.S., Rodríguez-Velázquez, J., Romero-Pérez, I.E., Ruíz, J., Saldarriaga, J.G., Sanchez-Azofeifa, A., Schwartz, N.B., Steininger, M.K., Swenson, N.G., Toledo, M., Uriarte, M., van Breugel, M., van der Wal, H., Veloso, M.D.M., Vester, H.F.M., Vicentini, A., Vieira, I.C.G., Bentos, T.V., Williamson, G.B., Rozendaal, D.M.A., 2016. Biomass resilience of Neotropical secondary forests. Nature 530:211–214. https://doi.org/10.1038/nature16512.

Proctor, J., Anderson, J.M., Chai, P., Vallack, H.W., Journal, S., Mar, N., Andersont, J.M., Chait, P., 1983. Ecological Studies in Four Contrasting Lowland Rain Forests in Gunung Mulu National Park, Published by: British Ecological Society Ecological Studies In Four Contrasting Lowland Rain Forests In Gunung Mulu National Park, Sarawak. Br. Ecol. Stud. 71, 237–260.

Quétier, F., Thébault, A., Lavorel, S., 2007. Plant traits in a state and transition framework as markers of ecosystem response to land-use change. Ecol. Monogr. 77, 33–52.

ter Braak, C.J.F., Cormont, A., Dray, S., 2012. Improved testing of species traits–environment relationships in the fourth-corner problem. Ecology 93:1525–1526. https://doi.org/10.1890/12-0126.1.

ter Braak, C.J.F., Peres-Neto, P., Dray, S., 2017. A critical issue in model-based inference for studying trait-based community assembly and a solution. PeerJ 5, e2885. https://doi.org/10.7717/peerj.2885.

R Core Team, 2014. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria.

Rengel, Z., 2004. Aluminium cycling in the soil-plant-animal-human continuum. Biometals 17:669–689. https://doi.org/10.1007/s10534-004-1201-4.

Robertson, G.P., Groffman, P.M., 2007. Nitrogen transformations. In: Paul, E.A. (Ed.), Soil Microbiology, Ecology, and Biochemistry. Academic Press, Amsterdam; Boston, pp. 341–347.

Russell, A.E., Hall, S.J., Raich, J.W., 2017. Tropical tree species traits drive soil cation dynamics via effects on pH: a proposed conceptual framework. Ecol. Monogr. 87: 685–701. https://doi.org/10.1002/ecm.1274.

Sim, J.W.S., Tan, H.T.W., Turner, I.M., 1992. Adinandra belukar: an anthropogenic heath forest in Singapore. Vegetatio 102:125–137. https://doi.org/10.1007/BF00044729.

Slik, J.W.F., 2009. Plants of Southeast Asia [WWW Document]. URL. http://www.asianplant.net/, Accessed date: 21 January 2017.

Thomas, G.S.P., 1991. Geology and geomorphology. In: Chia, L.S., Rahman, A., Tay, B.H. (Eds.), The Biophysical Environment of Singapore. Singapore University Press, Singapore, pp. 50–88.

Turner, B.L., 2012. Soil Classification in the 2 ha Forest Dynamics Plot in Primary Forest. Bukit Timah Nature Reserve, Singapore.

Turner, I.M., Wong, Y.K., Chew, P.T., Ibrahim, A.B., 1996. Rapid assessment of tropical rain forest successional status using aerial photographs. Biol. Conserv. 77 (177 ± 83).

Watanabe, T., Osaki, M., Tadano, T., 2001. Al uptake kinetics in roots of *Melastoma malabathricum* L. – an Al accumulator plant. Plant Soil 231:283–291. https://doi.org/10.1023/A:1010365607325.

Watanabe, T., Jansen, S., Osaki, M., 2006. Al–Fe interactions and growth enhancement in Melastoma malabathricum and Miscanthus sinensis dominating acid sulphate soils. Plant Cell Environ. 29, 2124–2132.

Westoby, M., Falster, D.S., Moles, A.T., Vesk, P.A., Wright, I.J., 2002. Plant ecological strategies: some leading dimensions of variation between species. Annu. Rev. Ecol. Syst. 33: 125–159. https://doi.org/10.1146/annurev.ecolsys.33.010802.150452.

Whitmore, T., 1972. Tree Flora of Malaya: A Manual for Foresters. Vol. 1. Longman Malaysia, Kuala Lumpur.

Whitmore, T., 1973. Tree Flora of Malaya: A Manual for Foresters. Vol. 2. Longman Malaysia, Kuala Lumpur.

Wong, Y.K., Chew, P.T., Ibrahim, A.B., 1994. The tree communities of the central catchment nature reserve, Singapore. Gard. Bull. Singap. 46, 37–78.

Wright, I.J., Reich, P.B., Westoby, M., Ackerly, D.D., Baruch, Z., Bongers, F., Cavender-Bares, J., Chapin, T., Cornelissen, J.H.C., Diemer, M., Flexas, J., Garnier, E., Groom, P.K., Gulias, J., Hikosaka, K., Lamont, B.B., Lee, T., Lee, W., Lusk, C., Midgley, J.J., Navas, M.-L., Niinemets, Ü., Oleksyn, J., Osada, N., Poorter, H., Poot, P., Prior, L., Pyankov, V.I., Roumet, C., Thomas, S.C., Tjoelker, M.G., Veneklaas, E.J., Villar, R., 2004. The worldwide leaf economics spectrum. Nature 428:821–827. https://doi.org/10.1038/nature02403.

Wright, S.J., Kitajima, K., Kraft, N.J.B., Reich, P.B., Wright, I.J., Bunker, D.E., Condit, R., Dalling, J.W., Davies, S.J., Díaz, S., Engelbrecht, B.M.J., Harms, K.E., Hubbell, S.P., Marks, C.O., Ruiz-Jaen, M.C., Salvador, C.M., Zanne, A.E., 2010. Functional traits and the growth–mortality trade-off in tropical trees. Ecology 91:3664–3674. https://doi.org/10.1890/09-2335.1.

Zaidey, A.K., Arifin, A., Zahari, I., Hazandy, A.H., Zaki, M.H., Affendy, H., Wasli, M.E., Hafiz, Y.K., Shamshuddi, J., Muhamad, M.N., 2010. Characterizing soil properties of lowland and hill dipterocarp forests at Peninsular Malaysia. Int. J. Soil Sci. 5:112–130. https://doi.org/10.3923/ijss.2010.112.130.

Zanne, A.E., Lopez-Gonzalez, G., Coomes, D.A., Ilic, J., Jansen, S., Lewis, S.L., Miller, R.B., Swenson, N.G., Wiemann, M.C., Chave, J., 2009. Data from: Towards a worldwide wood economics spectrum. Dryad Digital Repository https://doi.org/10.5061/dryad.234.