Influence of herbivory, water, light and nutrient availability in shaping tropical tree distribution patterns

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“*Cada quien es víctima de su propio invento*”
-Colombian proverb-

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Abstract

One of the most prominent patterns in lowland tropical forests is an increase of tree species diversity with rainfall. At the same time the change of forest composition, beta diversity, is extremely high along tropical rainfall gradients. The controls of species distribution patterns, which underlie patterns of alpha and beta diversity in such species-rich communities, remain a central question in ecology. With global climate change, rainfall intensity and seasonality are projected to vary, with unknown consequences for tropical forest communities. It is imperative to understand what determines tree distribution patterns, to be able to make reliable projections how climate change will affect tropical forest diversity.

Apart from direct effects of water availability, other factors that co- vary with rainfall have been hypothesized to indirectly influence species distributions across rainfall gradients: While wet origin species have been shown to be excluded from dry forests by drought (physiological tolerance hypothesis), the causes for the exclusion of dry origin species from wet forests are less clear. It has been hypothesized that high pest pressure through insect herbivores and pathogens (pest pressure gradient hypothesis), low light or low nutrient availability (light and nutrient availability distribution hypotheses, respectively) play an important role. Underlying these hypotheses are two assumptions: First that environmental conditions change, with wet forests having higher pest pressure and lower light and nutrient availability than dry forests; second that responses of species to these factors vary, with dry forest species being less defended and more nutrient- and light-demanding than wet forest species, which excludes them from wet forests.

In this thesis, the role of pest pressure, water, light and nutrient availability for species distributions was assessed experimentally along a pronounced rainfall gradient in Panama. Along a distance of only 65 km, annual rainfall increases twofold from 1600 to more than 3000 mm/year, accompanied by a decrease of dry season length. Along this gradient, two reciprocal transplant experiments as well as a feeding trial were performed, using species with contrasting distributions (dry vs. wet). The focus was set to the earliest regeneration stage, since this is believed to be the most susceptible one and the one when discrimination into origins takes place. The first transplant experiment included a pest exclosure treatment and aimed to disentangle the effects of pests, water and light availability for the distribution of 26 species. In the feeding trial, feeding rejection behavior of a generalist caterpillar was compared for seedlings and saplings of 50 species, to assess their integrated herbivore defenses. In the second transplant experiment, first-year seedlings of 26 species were transplanted to six sites spanning the gradient, and the influence of (natural) water, light and nutrients for seedling performance and consequent distribution was analyzed.

In congruence with previous studies, this thesis supported the physiological tolerance hypothesis. In contrast, at early regeneration stages the pest pressure gradient hypothesis was not supported. However, at later life stages, wet origin species had higher defenses, suggesting that responses change with ontogeny and that pests get more important after longer time periods. Light and nutrient availability did not consistently decrease with rainfall, refuting the first assumption of the respective hypotheses. Additionally, species of contrasting origins did not respond differentially to light and nutrients, and responses in general were weak, refuting the light and nutrient availability distribution hypotheses. Indeed, species did not exhibit any home advantage in terms of survival within their respective home range during the earliest regeneration stages (seedlings up to one year old), suggesting that discrimination into origins occurs at later life stages. However, wet origin species exhibited a home advantage through higher growth rates. This is related to a stress tolerance-growth trade-off, with drought tolerance achieved at the cost of lower growth rates. Dry origin species may be overgrown by wet origin species and excluded from wet forests through this mechanism, but only after longer time periods. While wet origin species clearly suffered more from drought than dry origin species, the dry seasons in this study were not strong enough to lead to their exclusion from dry sites. Strong dry seasons like during El Niño events may play a decisive role.

In summary, discrimination into origins does not occur within the first year of establishment, but needs longer time periods. Inherently lower growth rates as well as lower defenses of dry origin species eventually lead to their exclusion from wet forests, while periodic strong dry seasons are the most important mechanism for the exclusion of wet origin species from dry forests. Increases in frequency and strength of extreme dry seasons due to climate change will likely have direct influences on species distributions, with unknown consequences for tropical forest communities.

Zusammenfassung

Eins der auffälligsten Muster in tropischen Regenwäldern ist eine Zunahme der Baumartenzahlen mit zunehmendem Niederschlag, begleitet von einem starken Wechsel in der Identität dieser Arten (Beta-Diversität). Die Erklärung von Verbreitungsmustern von Arten, welchen die Alpha- und Beta-Diversität zugrunde liegt, ist eine der Hauptaufgaben in der Ökologie. Durch Klimawandel bedingt wird es starke Änderungen in der Intensität und Saisonalität von Niederschlägen geben, mit unbekannten Auswirkungen auf tropische Pflanzengesellschaften. Es ist deshalb dringlich zu verstehen, was die Verbreitungsmuster von tropischen Baumarten beeinflusst, um zuverlässige Prognosen machen zu können, wie sich Klimawandel auf Diversitätsmuster in tropischen Wäldern auswirken wird.

Außer dem direkten Einfluss von Niederschlag auf Verbreitungsmuster wurden andere Faktoren, die mit Niederschlag einhergehen, als indirekte Ursache vermutet. Der Ausschluss feuchtangepasster Arten (die in feuchten Wäldern vorkommen) aus trockenen Wäldern durch Trockenheit (Physiologische-Toleranz-Hypothese) ist durch viele Studien belegt worden. Die Ursachen für das Fehlen trockenangepasster Arten (die in trockenen Wäldern vorkommen) in feuchten Wäldern blieben dagegen weitgehend ungeklärt. Es gibt mehrere Hypothesen, welche Faktoren trockenangepasste Arten ausschließen, unter anderem hoher Fraßdruck (Pflanzenschädlings-Verbreitungs-Hypothese) und geringe Licht- und Nährstoffverfügbarkeit (Licht- und Nährstoffverfügbarkeits-Verbreitungs-Hypothesen) in feuchten Wäldern. Grundlage dieser Hypothesen sind jeweils zwei Voraussetzungen: erstens, dass feuchte Wälder mehr Schädlinge und weniger Licht- und Nährstoffverfügbarkeit haben, und zweitens, dass trockenangepasste Arten schlechter verteidigt und licht- und nährstoffbedürftiger sind als feuchtangepasste Arten, was zu ihrem Ausschluss aus feuchten Wäldern führt.

In dieser Arbeit wurden die Physiologische-Toleranz-, Schädlings-, Licht- und Nährstoffverfügbarkeits- Hypothesen entlang eines starken Niederschlagsgradienten in Panama untersucht. Der Niederschlag verdoppelt sich hier entlang einer Strecke von nur 65 km von 1600 auf mehr als 3000 mm/Jahr, was mit einer Verkürzung der Trockenzeit einhergeht. Es wurden zwei reziproke Umpflanzungsversuche, sowie ein Fütterungsversuch durchgeführt, unter Verwendung von Arten kontrastierender

Ursprünge (trocken vs. feucht). Der Fokus lag dabei auf den frühen Regenerationsstadien, da diese am empfindlichsten und die sind von denen man annimmt, dass die Trennung in trocken- und feuchtangepasste Arten entsteht. Der erste Umpflanzungsversuch enthielt eine Behandlung bei der Schädlinge ausgeschlossen wurden, und hatte zum Ziel, den Einfluss von Wasser, Schädlingen und Licht auf das Verbreitungsmuster von 26 Arten zu untersuchen. Im Fütterungsversuch wurde das Fraßverhalten einer generalistischen Raupe bei Keimlingen und Schösslingen von 50 Arten betrachtet, um deren Herbivorenabwehr zu quantifizieren. Im zweiten Umpflanzungsversuch wurden Keimlinge von 26 Arten auf sechs Flächen entlang des Gradienten ausgepflanzt und der Einfluss von (natürlich vorkommendem) Wasser, Licht und Nährstoffen auf Wachstum und Überleben, und wie sich diese auf Verbreitungsmuster auswirken, untersucht.

Wie in früheren Studien schon gezeigt, konnte die Physiologische-Toleranz-Hypothese in dieser Arbeit unterstützt werden. Die Schädlings-Verbreitungs-Hypothese dagegen konnte für die frühen Regenerationsstadien nicht unterstützt werden. Allerdings hatten bei älteren Keimlingen feuchtangepasste Arten höhere Herbivorenabwehr, was schließen lässt, dass sich diese mit der Ontogenie ändern, und dass der Einfluss von Herbivoren im späteren Alter an Bedeutung zunimmt. Weder Licht- noch Nährstoffverfügbarkeit waren in feuchten Wäldern geringer, was die erste Annahme der jeweiligen Hypothesen widerlegt. Zusätzlich zeigten trocken- und feuchtangepasste Arten keine unterschiedlichen Antworten auf Licht und Nährstoffe, was die Licht- und Nährstoffverfügbarkeits-Hypothesen weiter in Frage stellt. Bei frühen Regenerationsstadien (ein Jahr alte Keimlinge) hatte keine der Artengruppen einen Überlebensvorteil in ihrem Verbreitungsgebiet, was schließen lässt, dass die Unterteilung in trocken- und feuchtangepasste Arten zu einem späteren Zeitpunkt geschieht. Allerdings hatten feuchtangepasste Arten einen Heimvorteil in Bezug auf höhere Wachstumsraten, was auf ein Trade-off zwischen Trockentoleranz und Wachstum zurückzuführen ist. Trockenangepasste Arten könnten demnach von feuchtangepassten Arten auskonkurriert und nach längeren Zeiträumen aus feuchten Wäldern ausgeschlossen werden. Während feuchtangepasste Arten wie erwartet stärker unter Trockenheit litten als trockenangepasste Arten, waren die Trockenzeiten, die während dieser Studie herrschten, nicht stark genug um diese aus trockenen Wäldern auszuschließen. Stärkere Trockenzeiten, wie zum Beispiel während El Niño Ereignissen, spielen wahrscheinlich eine entscheidende Rolle dafür.

Zusammenfassend geschieht die Unterteilung in trocken- und feuchtangepasste Arten nicht während des ersten Jahres der Besiedlung, sondern braucht längere Zeiträume. Geringere Wachstumsraten sowie Herbivorenabwehr trockenangepasster Arten führen nach längerer Zeit zu deren Ausschluss aus feuchten Wäldern, während extreme Trockenzeiten den wichtigsten Mechanismus für den Ausschluss feuchtangepasster Arten aus trockenen Wäldern darstellen. Eine Häufung extremer Trockenzeiten, die durch Klimawandel erwartet werden, wird einen großen Einfluss auf Verbreitungsmuster von Arten haben, mit ungeahnten Konsequenzen für tropische Wälder.

Synopsis

Background

Tropical forests and the importance of understanding species distribution patterns

Tropical forests deliver important ecosystem services like water balance regulation and carbon storage, absorbing around a third of manmade carbon emission (Soepadmo 1993, Pan et al. 2011, Suryatmojo et al. 2013). However, tropical forests are threatened by climate change (Condit 1998, Sala et al. 2000, Malhi et al. 2009, Wright et al. 2009, Zelazowski et al. 2011). Pronounced changes of precipitation patterns in the tropics are projected with global change, with a strong drying trend especially in the neotropics (Hulme and Viner 1998, Hidalgo et al. 2013). Additionally, projected temperature increases will enhance evapotranspiration leading to decreased soil water availability (Hulme and Viner 1998, Delire et al. 2008). Decreased water availability associated with global change is projected to have pervasive consequences for tropical forests, leading to tree species composition changes and species loss (Condit et al. 1996, Coley 1998, Lewis 2006, Cook and Vizy 2008, Malhi et al. 2009, Sommer et al. 2010, Zelazowski et al. 2011). However, these projections have to be regarded with caution, given the difficulty of predicting future rainfall from "noisy" rainfall data, based on the high inter-annual variation in rainfall (Hulme and Viner 1998). Additionally, other factors which may be influenced by changes of water availability also have important influence on tree species composition and distribution. It is therefore of crucial importance to understand how this system works to be able to make predictions of the influence of climate change on species composition and diversity.

In tropical forests, tree species distributions and diversity vary along environmental gradients (Givnish 1999). One of the most common patterns is an increase of tree species diversity along rainfall gradients, accompanied by community composition changes (Gentry 1988, Wright 1992, Pyke et al. 2001, ter Steege et al. 2003). Understanding the processes underlying the observed gradients in forest composition is crucial to improve projections of consequences of climate change for tropical forests, and to inform mitigation and adaptation strategies.

Most studies analyzing tree species distribution patterns have focused on direct influences of abiotic factors such as water, light, nutrient and temperature. However, there is increasing evidence that interactions with the biotic environment like pest pressure, competition, pollination and seed dispersal are as important as abiotic factors in determining species distributions (Kitajima and Poorter 2008, see also Kubisch et al. 2014). This thesis aims to improve our understanding of how pest pressure and abiotic factors interact with species specific intrinsic factors related to growth, herbivore defenses, drought and shade tolerance, and how these factors shape tree species distribution patterns across tropical rainfall gradients. As a general guideline of the interplay of these factors, a conceptual framework was developed (see Figure 1). The most important factors influencing species distributions as shown in Figure 1 will be introduced in the following section.

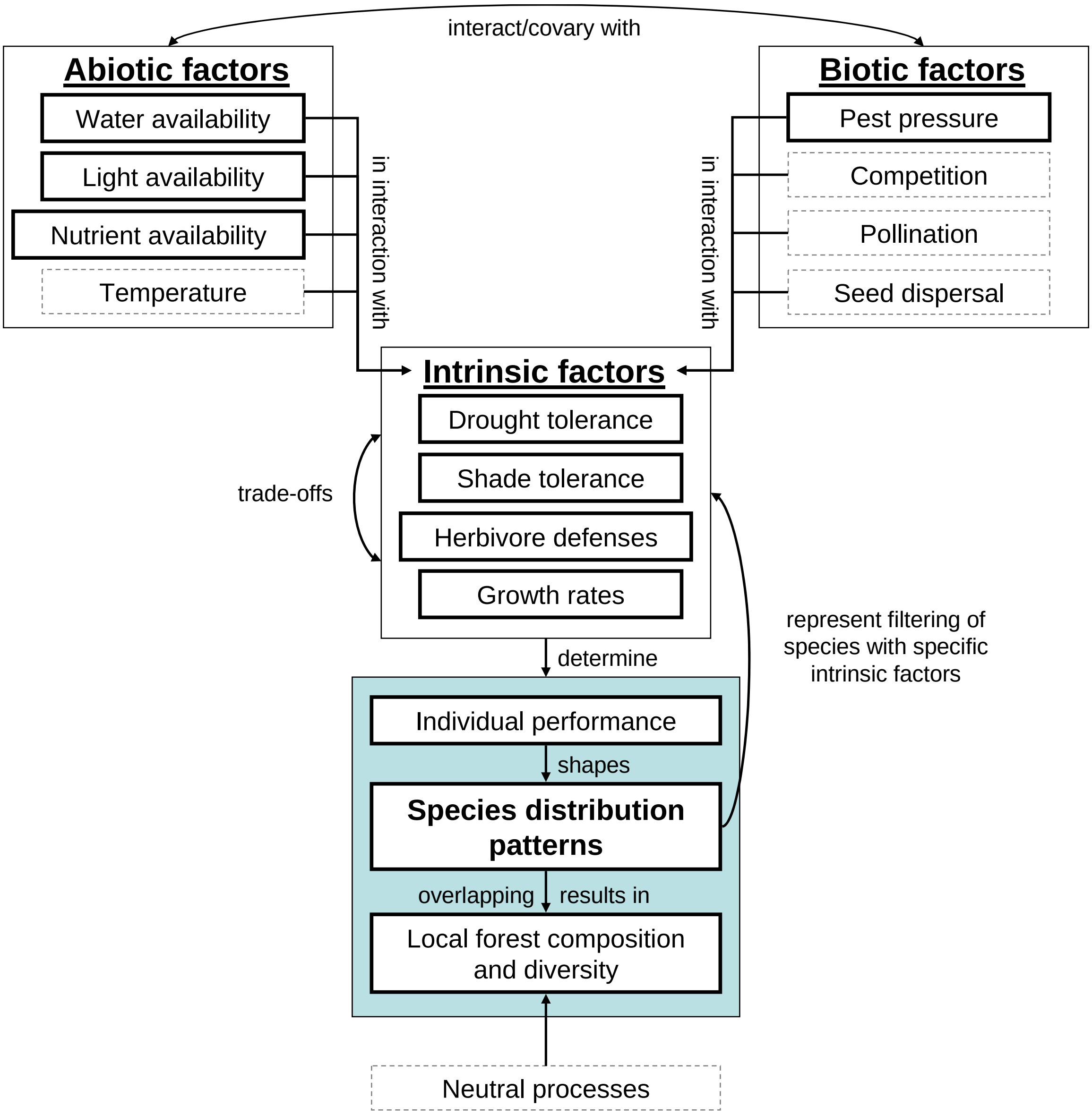

Figure 1: Conceptual frame work showing potential mechanisms of how abiotic and biotic factors interact with species-specific intrinsic factors (adaptations to the environment) to determine species performance and consequent distribution patterns, which in turn determine the local forest composition and diversity. The arrow going from species distribution patterns to intrinsic factors represents the filtering of a species pool with intrinsic factors favorable for the adverse conditions in each local forest site. Species-specific intrinsic factors may limit each other by trade-offs, so that e.g. a species may not be both drought- and shade tolerant. In addition to species distribution patterns based on their ability to colonize niches (“niche theory”), neutral processes may shape local forest composition and diversity. Abiotic factors in the study area co-vary, and may also interact with biotic factors (e.g. wetter forests are expected to be darker, pest pressure is expected to increase with rainfall). Dashed boxes represent factors that were not analyzed or discussed in this thesis.

Factors influencing species distribution patterns

Abiotic factors influencing plant species performance are mainly water, light and nutrient availability as well as temperature. These factors often co-vary and interact with each other, making it difficult to distinguish between effects of single factors. Additionally, they may influence biotic factors such as plant-animal interactions, i.e. pest pressure, pollination and seed dispersal, as well as plant-plant interactions, i.e. competition.

Plant species may have adapted to their specific environmental conditions and should therefore have a "home advantage" compared to "foreign" species adapted to a different set of environmental variables (Kawecki and Ebert 2004). Following Figure 1, such adaptations of organisms to their environment will be termed "intrinsic factors". For example, plant species may have evolved drought and shade tolerance or defense mechanisms as an answer to low water and light availability or herbivory, respectively. A filtering of species with unsuitable intrinsic factors, additional to evolutionary adaptations, may also result in species compositions with distinct "home advantage". These species specific intrinsic factors interact with the abiotic and biotic environment to determine the individual performance and consequent establishment success of a plant in a specific site. Differential establishment success leads to different distribution patterns of species which underlie the species diversity of a specific site.

Under limited resources, investing in adaptations to an environmental stress leaves less resources for other processes, resulting in trade-offs between different intrinsic factors (see Orians and Solbrig 1977, Smith and Huston 1989). For example higher drought and/or shade tolerance may come at the cost of lower growth rates, or shade tolerant species may have limited drought tolerance (Smith and Huston 1989). A possible mechanism for the existence of these trade-offs is thought to be related to differential investment of limited resources to different organs or systems (Orians and Solbrig 1977). For example, a plant can invest either in above- or in belowground biomass. In the first case, the plant would increase its leaf area index and be more shade tolerant; in the second case, the plant would increase its root mass and be able to reach deeper water layers, and therefore be more drought tolerant (Orians and Solbrig 1977). However, the mechanisms behind these trade-offs are complicated, and little empirical support has been found for these hypothesized trade-offs.

In this thesis, I focus on early life stages including the seed germination and seedling stage, since these are considered to be most vulnerable to abiotic stressors and the ones during which forest composition is determined (Harper 1977, Daws et al. 2005). I further concentrate my efforts on analyzing the effects of pest pressure, water, light and nutrient availability in shaping species distributions, with a special emphasis on pest pressure. Other factors that influence performance and regeneration success of plants could additionally influence distribution patterns, but were not covered in this thesis:

Temperature normally is an important factor for plant performance, but in tropical forests, it is relatively constant throughout the year and varies little across rainfall gradients (see e.g. Figure 3). Accordingly, it has previously been shown to play a minor role in shaping species distribution patterns along tropical rainfall gradients (Amissah et al. 2014). Alternatively to deterministic factors, species diversity and distribution have been hypothesized to be driven by stochastic processes (Hubbell 2001). As a null model against the “niche theory”, the “neutral theory of biodiversity” postulates that every species has the same chance to establish and that diversity patterns are driven by stochastic processes like random dispersal. This has been highly debated (Harpole 2010, Rosindell et al. 2011), and it would go far beyond the scope of this thesis to discuss this further. Competition between seedlings has been shown to have little influence for recruitment in the understory of tropical forests (Wright 2002, Paine et al. 2008), probably because the effects of competition in low resource habitats (in terms of nutrients, but also water and light availability) is lower than the direct effects of the low resource availability on plant recruitment (Grime 1977). In tropical forests, a lower degree of specialization than previously thought has been found for mutualistic networks, i.e. pollination and seed dispersal (Schleuning et al. 2012), meaning that these are less important for the distribution of species along rainfall gradients.

Several hypotheses for the relation of herbivory, water light and nutrient availability on species distributions have been postulated (Huston 1994, Coley and Barone 1996, Givnish 1999, ter Steege et al. 2003, Brenes-Arguedas et al. 2008, 2009, 2011, Baltzer and Davies 2012, Sterck et al. 2014). However, experimental evidence is still scarce, even for the basic assumptions underlying these hypotheses. Hereafter, I show an overview of potential mechanisms behind these hypotheses.

Water availability

Among the most prominent patterns in tropical forests are relations of tree species distributions (Bongers et al. 1999, Engelbrecht et al. 2007, Condit et al. 2013), changes of forest community composition (Pyke et al. 2001), and increases of species richness with rainfall (Gentry 1988, Wright 1992, ter Steege et al. 2003). Not only species numbers but also species identities change along rainfall gradients, leading to a high species turnover (Condit et al. 2002). Although these patterns have been widely documented, the underlying mechanisms remain controversial. Direct effects of water availability on species performance have been thought to limit species distributions, by excluding wet forest species, which are drought-sensitive, from dry forests (physiological tolerance hypothesis, Currie et al. 2004). Empirical evidence supports the physiological tolerance hypothesis for tropical forests. Along rainfall gradients, strong positive correlations between drought tolerance or traits conferring drought tolerance and occurrence towards the dry end of the gradient were found using reciprocal transplant experiments along rainfall gradients (Engelbrecht et al. 2007, Baltzer et al. 2008), showing that wet forest species are excluded by limited water availability (i.e. drought) from dry forests (Brenes-Arguedas et al. 2009, Baltzer and Davies 2012). Although the physiological tolerance hypothesis has received a lot of support, on its own it is not enough to explain the high species turnover observed (Engelbrecht et al. 2007). Especially, it fails to explain why dry origin species are not able to colonize wet forests.

Other factors co-varying with rainfall, including herbivory, light and nutrient availability, have been hypothesized to additionally shape species distribution patterns (Givnish 1999). These factors, which are thought to exclude dry origin species from wet forests, are explained in the following sections.

Herbivory

Herbivory is an important factor in tropical forests; up to 11% of the yearly biomass production is consumed (Coley and Barone 1996). This amount is enough to significantly reduce plant fitness and performance (Dirzo 1984, Marquis 1984, Eichhorn et al. 2010). Gradients in pest pressure therefore may influence species ranges. Two contrasting hypotheses regarding the role of pests in shaping species distributions have been brought up, and will be explained subsequently:

The pest pressure gradient hypothesis, or at least its underlying assumptions, has been around for a long time (Gillett 1962, Givnish 1999, Leigh et al. 2004, Gilbert 2005), but it was firstly named as such by Baltzer and Davies (2012). According to it, higher pest pressure (by insect herbivores and pathogens) in wetter forests excludes less defended dry forest species, limiting their distribution in wet forests. The pest pressure gradient hypothesis is based on two underlying assumptions: Firstly, that herbivore pressure increases with moisture, and secondly, that herbivore defenses increase with moisture, as a consequence of selection pressure and/or environmental filtering. Dry distribution species need to invest less in defenses, as pest pressure is lower in their distribution range. Additionally, dry distribution species are often deciduous, leading to a lower evolutionary pressure to defend the leaves, in contrast to the long-lasting leaves of (broadleaf) evergreen, wet distribution species (Coley and Barone 1996).

The opposite to this assumptions, however, has also been hypothesized: Traits conferring drought resistance like thicker, tougher leaves may also confer herbivore defense, resulting in dry origin species being better defended than wet origin species (Givnish 1999). Limiting water availability could also lead to higher costs of tissue loss towards drier forests, and thus increase the need of higher defenses in dry forest species (Givnish (1999); resource allocation theory based on the resource water). More generally speaking, every kind of stress tolerance is expected to be related to lower growth rates (stress tolerance-growth trade-off), which lead to a lower ability to recover from herbivory; as a consequence, stress-tolerant species may be expected to be better defended against herbivory (Grime 1977, Smith and Huston 1989). Pest pressure also has been hypothesized to decrease with moisture: As a consequence of higher defenses in wet forests, herbivores need a longer development time. The resulting longer exposure to predators and parasitoids is expected to result in lower herbivore abundance in wet forests (Connahs et al. 2011).

In summary, the influence of herbivory in shaping species distributions along rainfall gradients remains uncertain.

Light availability

Light availability directly influences plant performance in terms of survival, recruitment and growth (Wright 1992, Kobe 1999, Davidson et al. 2002, Montgomery and Chazdon 2002, Bloor and Grubb 2003, Balderrama and Chazdon 2005, Rüger et al. 2009, 2011), as well as indirectly in interaction with water availability (Burslem et al. 1996, Barone 1998, de Gouvenain et al. 2007, Brenes-Arguedas et al. 2011). In general, higher light availability results in higher performance, unless it gets too high (over 50 % full daylight; Poorter 1999), or unless water availability is limiting, resulting in lower performance with increasing light availability (Brenes-Arguedas et al. 2011). In the understory of tropical forests, light availability seldom exceeds 10 % daylight (Coomes and Grubb 2000) and is often much lower (Chazdon and Pearcy 1991, Poorter 1999).

Light availability in tropical forests has been hypothesized to decrease along rainfall gradients (Smith and Huston 1989, Wright 1992, Markesteijn 2010). Higher rainfall correlates with higher Leaf Area Index (LAI), number of canopy strata and basal area of trees (Murphy and Lugo 1986), higher cloudiness (Wright and Van Schaik 1994, Graham et al. 2003, Santiago and Mulkey 2005) and lower proportion of deciduous species (Condit et al. 2000, Parker et al. 2005), all factors that may reduce understory light availability (Brenes-Arguedas et al. 2011). Up to now, only few studies have actually compared understory light availabilities between dry and wet forests. However, none of these studies were designed to specifically compare light availabilities in different forests, and have methodological problems which may bias the results. Most of the studies found indeed lower light availabilities with increasing rainfall (Coomes and Grubb 2000, Santiago et al. 2004, Parker et al. 2005, Brenes-Arguedas et al. 2011, Peña-Claros et al. 2012), except for one which found a positive relation (Engelbrecht 1998), and one which found no relation (Harms et al. 2004).

As light availability decreases with increasing rainfall, it has also been hypothesized that wet forest species have adapted to it and are more shade tolerant than dry forest species (Givnish 1988, Smith and Huston 1989). Lower shade tolerance of dry origin species could also result from a trade-off between drought and shade tolerance (Smith and Huston 1989, Huston 1994). The consequent partitioning along light availability gradients is thought to result in the exclusion of light-demanding dry forest species from wet forests (de Gouvenain et al. 2007, Kitajima and Poorter 2008, but see

Lieberman et al. 1995). Hereafter, this is called the light availability distribution hypothesis. However, empirical evidence shows that wet forest species are not more shade tolerant than dry forest species (Brenes-Arguedas et al. 2011) and the influence of light in shaping species distribution patterns remains unclear.

Another possible mechanism of how light availability influences species distributions is by increasing negative effects of drought (Brenes-Arguedas et al. 2011): High light microsites or gaps have been hypothesized to have lower water availabilities due to increased irradiance, temperature and wind, which enhance evaporation (Poorter and Hayashida-Oliver 2000). High light in dry areas therefore may increase drought stress for drought-intolerant, wet origin species, excluding them.

Light availability also may interact with herbivory: In shady environments, it is more costly for a plant to replace lost biomass, so that defenses should be higher (Coley and Barone (1996); resource allocation theory (Barbour et al. 1980) based on the resource light). Therefore, high defenses of wet distribution species may be influenced by lower light availabilities in wet forests.

Nutrient availability

Another factor that may co-vary with rainfall is the availability of nutrients. Under high precipitation regimes, leaching of nutrients is expected to result in a lower soil nutrient availability in wet forests compared to dry forests (Radulovich and Sollins 1991, Austin and Vitousek 1998, Schuur and Matson 2001, Santiago et al. 2005). Evidence for lower nutrients with increasing rainfall has been found for some rainfall gradients (Vitousek and Sanford 1986, Swaine 1996, Swaine and Becker 1999, Schuur and Matson 2001), but not for others (Turner and Engelbrecht 2011), showing that this correlation is not ubiquitous and also depends strongly on the geology and the nutrient considered (Jones et al. 2013).

Higher nutrient availability influences seedling performance directly by increasing growth rates (e.g. Alvarez-Clare et al. 2013, Yavitt and Wright 2008). Therefore, a partitioning along a nutrient availability gradient could explain the distinct distribution of species along rainfall gradients. Wet forest species adapted to lower nutrient availability are expected to have an advantage in nutrient-poor wet forests, whereas dry forest species adapted to nutrient-rich soils have an advantage in dry forests (Brenes-Arguedas et al. 2008). Dry forest species therefore are excluded from wet forests due to low soil nutrient contents (nutrient availability distribution hypothesis). Underlying this mechanism are trade-offs between nutrient acquisition or retention ability and growth. It has been hypothesized that plants growing in nutrient-poor soils invest more in underground biomass than in aboveground biomass (Chapin III 1980, Aerts and Chapin III 1999). Consequently, they have improved nutrient acquisition, but lower growth rates. In contrast, species from nutrient-rich soils may invest more in photosynthetic tissue and have higher growth rates, but are limited in their nutrient acquisition, which limits their performance and distribution in nutrient-poor sites. However, such a trade-off is not ubiquitous and other mechanisms such as a higher root length per root mass or better nutrient retention have also been found (Aerts 1999, Aerts and Chapin III 1999).

Plants from infertile soils additionally have been expected to have lower allocation plasticity; this results in low growth rates even in fertile environments, due to constrains in allocating in organs other than roots, which are needed for nutrient absorption. Therefore, they should have inherently lower growth rates (Grime 1977, Chapin III 1980). As a consequence, species from nutrient-poor soils would be outperformed in nutrient-rich environments, leading to their exclusion (Aerts 1999).

It is difficult to distinguish between the correlated effects of water and nutrient availability (Swaine 1996, ter Steege et al. 2003, Brenes-Arguedas et al. 2008, Kitajima and Poorter 2008). Although some studies have discarded soil types and soil fertility as major factors limiting species distributions (Chust et al. 2006, Brenes-Arguedas et al. 2008), others have found a weak effect of edaphic gradients on species distributions (Toledo et al. 2012), and again others have found strong correlations between species distribution patterns and soil fertility (John et al. 2007, Condit et al. 2013). As nutrient availability significantly influences plant performance directly and indirectly (see below), and the existing evidence remains conflictive, it is important to analyze its role in shaping species distributions.

Herbivory and soil nutrient availability may interact and further shape species distribution patterns: Higher availability of soil nutrient content is generally reflected in higher leaf nutrient content (Vitousek and Sanford 1986, Schuur and Matson 2001, Ordoñez et al. 2009, Han et al. 2011). High leaf nutrient content has been shown to increase damage by herbivores (Santiago et al. 2012). This could potentially increase the need for species growing in fertile sites to develop better herbivore defenses. On the other hand, it has also been suggested that lower soil nutrient contents in wet forests may select for species with higher defenses (Santiago et al. 2004). Biomass loss should have a greater impact for a plant growing in an infertile environment compared to a plant in a fertile environment, which can re-grow fast. Additionally, most plants in infertile environments are evergreen to reduce nutrient loss through leaf shedding (Aerts and Chapin III 1999), and therefore face herbivory for a longer period of time (Coley and Barone 1996), increasing their need for defenses. If wetter forests are more infertile than drier forests, higher defenses in wet forest species may result from lower soil nutrient content (Santiago et al. 2004). The interaction between nutrient availability and herbivores has been shown to shape species distributions along edaphic gradients (Fine et al. 2004), and it could also shape distributions and species richness along rainfall gradients.

The most limiting nutrient in lowland tropical forests is phosphorus (Vitousek 1984). This is in contrast to mountain tropical forests and other ecosystems, where nitrogen is the limiting nutrient (Vitousek and Sanford 1986, Aerts and Chapin III 1999). Therefore, in this thesis I concentrate on the influence of soil phosphorus concentrations on species performance and distributions.

Hypotheses and objectives

The aim of this thesis was to test the role of herbivory, both by insect herbivores and pathogens, on tree species performance and distribution along tropical rainfall gradients. Additionally, I considered the factors water, light and nutrient availability, since they have also been hypothesized to play an important role in shaping species distributions. The study was conducted at the Isthmus of Panama - a narrow strip of land that lies between the Caribbean Sea and the Pacific Ocean. I used this Isthmus as a model system, since it has a strong rainfall gradient from the comparatively dry Pacific to the wetter Caribbean, along a relatively short distance (Figure 2). Most tree species in this gradient have a distinct distribution and can be categorized into dry and wet origin species.

The following four hypotheses were tested:

1. Physiological tolerance hypothesis: Plant species of wet origin are less drought tolerant than species of dry origin. Drought therefore directly excludes wet origin species from dry forests.
2. Pest pressure gradient hypothesis: Plant species associated with wet forests exhibit higher defenses against herbivores than species associated with dry forests due to increasing herbivore pressure. Higher defenses in wetter forests are the footprint of higher herbivore pressure leading to environmental filtering from herbivores and/or of adaptation to higher selection pressure from herbivores.
3. Light availability distribution hypothesis: Low understory light availability in wet forests excludes light-demanding dry origin species. Higher light requirements in dry than in wet forest species are based on a trade-off between drought and shade tolerance, and/or because they are adapted to higher light conditions in the understory of dry forests.
4. Nutrient availability distribution hypothesis: Low soil nutrient content in wet forest excludes nutrient-demanding dry origin species. Dry origin species are more nutrient-demanding than wet origin species as a consequence of environmental filtering from low nutrient sites or as a consequence of evolutionary adaptation.

To test these hypotheses, three experiments including one feeding trial and two transplant studies, one with seeds and one with seedlings, were conducted.

Research area and study sites

The Panama Canal Watershed was used as a model system for these studies (Figure 2). The area was suitable due to a strong rainfall gradient that exists along the Panama Canal, ranging from 1600 mm/year on the Pacific dry side, to over 3000 mm/year on the Caribbean wet side of the Isthmus, along a distance of only 65 km (see Figure 2 and Figure 3). One dry and one wet season occur during the year. Dry season length (i.e. days with rainfall lower than evapotranspiration) correlates negatively with annual rainfall, and ranges from 147 days on the Pacific coast to 117 days on the Caribbean coast (Engelbrecht et al. 2007). During the wet season, perhumid conditions prevail both in the dry and the wet side of the gradient (Figure 3). Mean annual temperature averages 27°C and is constant throughout the year, as well as across the region (see Figure 3).

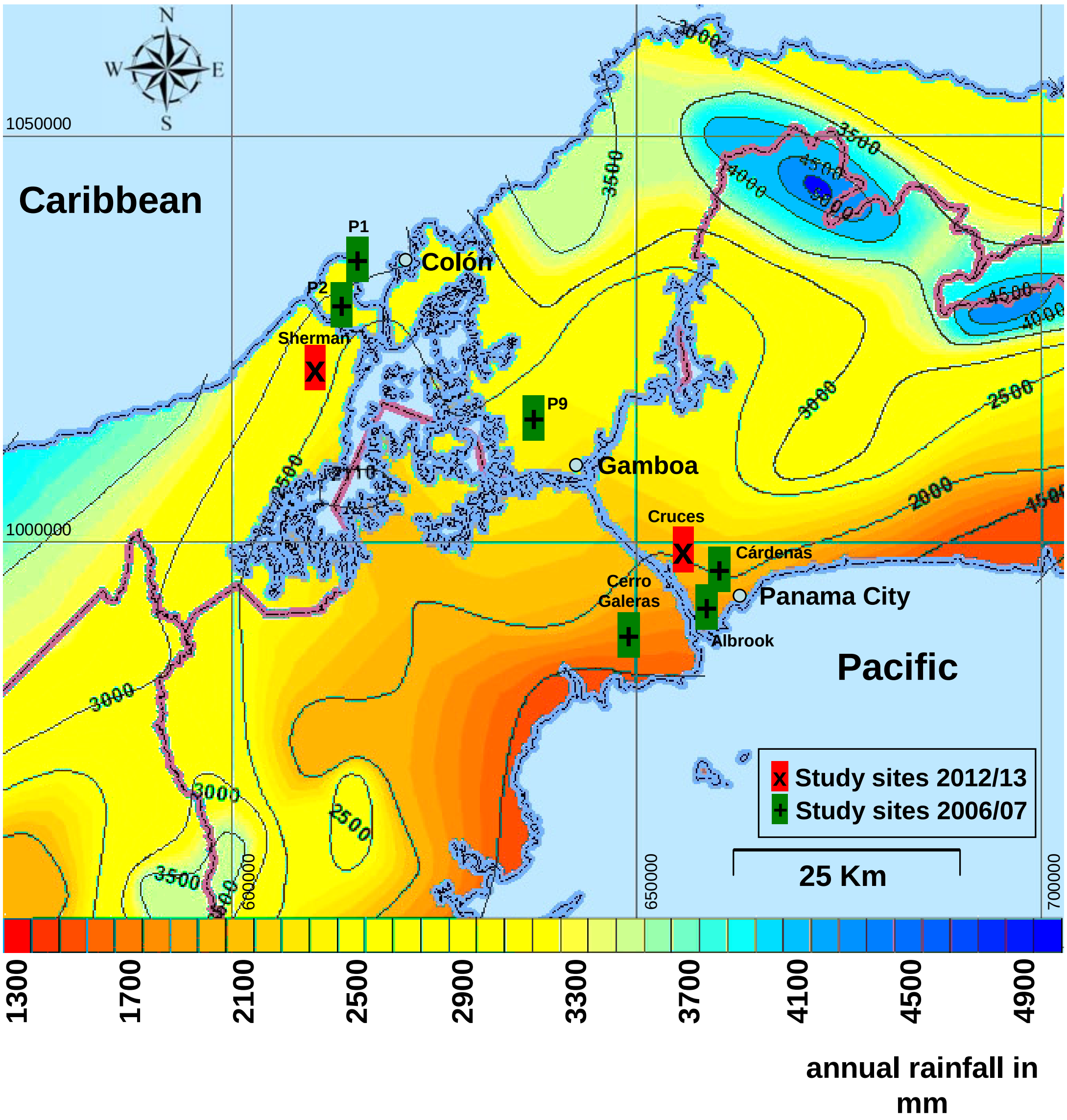

Figure 2: Map of the study area showing the study sites and rainfall gradient. The rainfall gradient ranges from around 1600 mm/year in the Pacific to over 3000 mm/year in the Caribbean side of the Isthmus. The seed transplant experiment in manuscript 1 was carried out in 2011/2012 at a wet and a dry forest site (marked with the red “x”). The seedling transplant experiment in manuscript 3 was carried out in 2006/2007 at six sites spanning the rainfall gradient (marked with a green “+”). The leaves for the feeding experiment in manuscript 2 were collected in forests along the whole gradient, without restriction to specific sites.

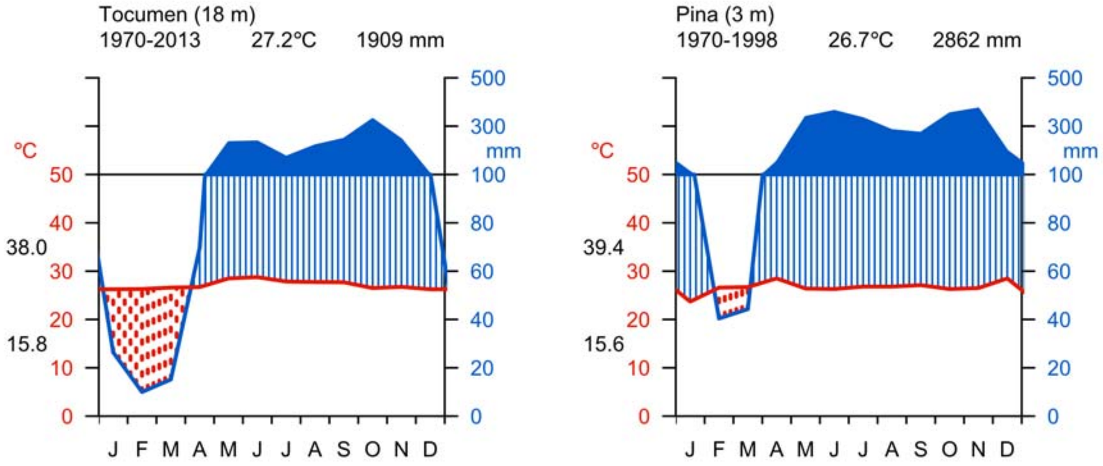

Figure 3: Climate diagrams of Tocumen (left), located near the Pacific Ocean, and Piña (right), located near the Caribbean. During the year there is one dry season (when potential evapotranspiration exceeds rainfall; dotted), ranging from January to April, and one wet season from May to December with perhumid conditions (> 100 mm/month, blue), during which water availability is not limiting. Note that yearly rainfall is higher in the Caribbean compared to the Pacific side, and that the dry season is longer and stronger in the Pacific side of the Isthmus. Graphs were created with the package climatol in R (Guijarro 2013) using data of the Panamanian "Empresa de Transmisión Eléctrica S.A." (Gerencia de Hidrometeorología ETESA 2015).

Most of the area lies below 300 m above sea level (Condit et al. 2001), but this study was conducted on sites between 30 and 200 m above sea level. The present-day forest found along the Canal Watershed is mostly secondary or mature secondary lowland tropical forest (Pyke et al. 2001, Wright and Samaniego 2008). The geology is extremely complex, including sedimentary, volcanic and metamorphic bedrock (Stewart et al. 1980, Pyke et al. 2001). Accordingly, soil nutrient availability covers a great range (Turner and Engelbrecht 2011). The vegetation of the study area ranges from tropical wet over tropical moist to tropical dry forest according to the Holdridge life zone system (Holdridge 1947).

On 72 plots spanning the rainfall and nutrient gradient, all tree species have been mapped and identified to species (Condit et al. 2013). This was the basis for most of the distribution and association indices along rainfall and nutrient gradients used in this study. Along the rainfall gradient species number increase, with sites in the drier end having around 45 species, while sites at the wetter end reach values of 150 species per hectare (Pyke et al. 2001, Condit et al. 2013). At the same time, β-diversity is very high: While adjacent sites still share around 70% of the species, this number declines continuously along the gradient, with sites 50 km apart sharing only around 10% of their species (Condit et al. 2002). This variation is much higher than in Amazonian forests in Peru and Ecuador, and was attributed to the high variation of soils and rainfall in Panama (Condit et al. 2002). As a consequence, many studies have used this rainfall gradient as a model system to analyze the role of water, herbivores, light and nutrients for species distribution patterns and diversity gradients (Bunker and Carson 2005, Engelbrecht et al. 2007, Brenes-Arguedas et al. 2008, 2009, 2011, Kursar et al. 2009, Comita and Engelbrecht 2013, Condit et al. 2013, Jones et al. 2013, Spear et al. 2015).

Manuscripts of this thesis

This thesis includes three manuscripts, each presenting a different experiment. The first experiment, a seed transplant trial, was designed to test the physiological tolerance, pest pressure, and light availability hypothesis (hypotheses 1, 2 and 3). The second experiment was a feeding trial with caterpillars of a generalist herbivore, aiming to test the pest pressure gradient hypothesis (hypothesis 2). In the third experiment a seedling transplant study was performed to test the physiological tolerance, light availability and nutrient availability hypothesis (hypotheses 1, 3 and 4). In the following section, these studies are presented in more detail.

Manuscript 1: Seed transplant experiment

In this experiment we analyzed the role of drought, pest pressure (combined insect herbivore and pathogen damage) and light availability on the distribution of tree species along the rainfall gradient (hypotheses 1, 2 and 3). In a reciprocal transplant experiment we seeded seeds of species with contrasting distributions in a dry and a wet forest site, so that in every site we had species within their home range and “foreign” species. The dry forest site was set up in the “Parque Nacional Camino de Cruces”, and the wet forest site in the “Parque Nacional San Lorenzo” (see Figure 2). Half of the seeds and emerging seedlings were treated with a combined insecticide-fungicide treatment to exclude herbivory so that within each site we had 60 paired plots with each pair containing one control and one pest exclosure. We measured water and light availability in the plots and monitored performance in terms of germination, survival and absolute height growth of the species. Establishment success, the proportion of seedlings remaining at the end of the experiment, as well as growth, germination, dry and wet season survival were related to site (dry vs. wet), origin of the species (dry vs. wet), treatment (control vs. exclosure) and light availability.

Establishment success after one year did not reflect species distribution patterns, showing that the discrimination of the species into dry and wet origin does not occur within the first year of seedling establishment. However, in the wet forest, wet origin species had a home advantage over dry forest species through higher growth rates. At the same time, drought limited survival of wet origin species in the dry forest, supporting the physiological tolerance hypothesis. Together these processes sort species over longer time frames, and exclude species outside their respective home range. Although we found pronounced negative effects of herbivory on the seedlings, dry origin species were not affected stronger than wet origin species in the wet site. Therefore, this study did not support the pest pressure gradient hypothesis, at least at the seed-to-seedling transition stage. We found some positive effects of light availability on the seedlings. However, dry origin species did not benefit more from higher light availability compared to wet origin species, as needed for the light availability hypothesis. This study therefore did not support the light availability hypothesis at early life stages.

Manuscript 2: Feeding trial

In this experiment, we aimed to corroborate the second assumption of the pest pressure gradient hypothesis (hypothesis 2): Wet forest tree species have higher herbivore defenses than dry forest species, as an adaptation to higher herbivore pressure in wet forests. We conducted a feeding trial with caterpillars of a generalist herbivore (*Spodoptera frugiperda*) to determine feeding rejection behavior of plants with known distribution along the rainfall gradient. In each trial, the caterpillars had the choice (dual choice test) between leaf disks of a focal species and a standard species (*Ixora coccinea*). The leaves for the experiment were collected from seedlings and saplings in forests along the Canal, spanning the whole rainfall gradient. A rejection index was calculated based on the ratio of consumed leaf area between the focal and the standard species. This feeding rejection represented the integrated herbivore defenses of plants and was related to plant species distribution patterns along the rainfall gradient, to test the hypothesis that wet forest species are better defended against insect herbivores than dry forest species. We also related the feeding rejection to shade tolerance and associations to soil nutrients of the species, to control for the effects of light and nutrient availability on defenses, as well as to several mechanical defensive traits.

The main result of this manuscript was that the rejection of the caterpillars increased with the association of the tree species to wetter forests. This shows that wet origin species were better defended than dry origin species and supports the second assumption of the pest pressure gradient hypothesis. Rejection also increased with increasing shade tolerance of the species. However, shade tolerance and association to moisture were not correlated, showing that higher defenses of wet origin species were independent of the shade tolerance of the species. Association to nutrients did not influence rejection, supporting that defenses and not nutrient content of the leaves were responsible for the rejection behavior of the caterpillars. None of the mechanical defensive traits analyzed influenced rejection, suggesting that chemical and not mechanical defenses were more important in this system.

Manuscript 3: Seedling transplant experiment

In this experiment, we analyzed performance of tree species as a function of water, light and nutrient availability, to see which factors are the most constraining for seedlings in tropical forests. We then related performance to species distribution patterns to see if these abiotic factors are responsible for discriminating into dry and wet origin species (hypotheses 1, 3 and 4). A transplant experiment at six sites (see Figure 2) was conducted transplanting seedlings of species with known distribution patterns along the rainfall gradient, so that in every site all species were available. Environmental parameters gravimetric soil moisture, light and nutrient availability were monitored during one dry and one wet season, totaling one year. Survival and relative leaf area and height growth were measured, and the influence of abiotic factors on these performance parameters analyzed.

Drought was the major constrain to seedling growth and survival especially during the dry season. Performance was higher in the wet season than in the dry season and increased with species drought resistance. Light availability had a positive effect on growth unless under dry conditions, when it enhanced the negative effects of drought. Soil phosphorus enhanced performance when water availability was high, but reduced it under drought, most probably due to an interaction with mycorrhiza. These results indicate that soil moisture availability across space and time is a dominant factor limiting seedling regeneration even in moist tropical forests, while light and nutrient availability play a minor role. However, we found no indication of a home advantage of species under the conditions they were associated with, indication that performance difference at the seedling stage do not drive species distribution patterns at the regional scale, but that other life stages are driving distribution patterns. Additionally, strong dry seasons like during an El Niño event may be the trigger that excludes wet origin species from dry sites.

General discussion

In the following sections, I discuss the results of my work in the light of the physiological tolerance, pest pressure gradient, light availability and nutrient availability hypotheses. I further depict some interesting side-results that go beyond the presented hypotheses and have influences on differential species performance and/or distribution.

Physiological tolerance hypothesis

According to the physiological tolerance hypothesis, dry origin species should be more drought-tolerant than wet origin species, resulting in wet origin species being excluded from dry forests due to drought. This effect should be especially strong during the dry season.

Along the rainfall gradient I analyzed, soil water availability mimicked rainfall patterns: I found strong correlations between rainfall and gravimetric soil water content (manuscript 3), and plots in the wet Caribbean were significantly moister than in the dry Pacific (manuscript 1). Soil water content was significantly lower in the dry compared to the wet season (manuscript 1 and 3). Drought significantly reduced species performance in the dry season (manuscript 1 and 3). However, it also reduced survival during the wet season (manuscript 3), highlighting the importance of water availability even within the humid tropics (Comita and Engelbrecht 2013).

Dry origin species have been shown to be more drought-tolerant than wet origin species (Engelbrecht et al. 2007), and this was also valid for the species used in this study (manuscript 3).

In both transplant experiments (manuscript 1 and 3), water availability played a key role for species performance. During the dry season, survival of dry origin species was higher than for wet origin species (manuscript 1 and 3) especially in the dry side (manuscript 1). This indicates that drought tolerance of dry origin species enhanced their performance during drought. For wet origin species, dry season survival was lower in the dry compared to the wet site (manuscript 1). This underlines that reduced water availability in the dry season limits wet origin species, as expected from the physiological tolerance hypothesis.

However, the overall establishment success, i.e. the proportion of seedlings remaining after one year, did not differ between dry and wet origin species (manuscript 1). Therefore, the home advantage found for dry origin species in the dry season was not strong enough to exclude wet origin species from dry sites, at least during the initial year of seedling establishment. Species performance was also not affected significantly by the origin of the species in interaction with neither soil moisture nor drought tolerance (manuscript 3). This indicates that although water availability limited performance of the species, it did not discriminate between the origins. The year in which the seedling transplant experiment was conducted was a relatively humid year. The dry season was characterized by being shorter than the long-term average, and by short wet spells in between, which may have been strong enough to prevent differential performance between origins. This picture could change after several dry seasons or with stronger and longer dry seasons like during an El Niño event. These events occur in irregular intervals more or less every four years, and lead to extreme drought in some tropical regions including Panama (Cane and Zebiak 1985, Malhi and Wright 2004). Strong inter-annual performance differences in the dry season have been found in years with differing rainfall regimes (Comita and Engelbrecht 2013). While in the initial years after establishment the discrimination of the species into dry and wet origin species may not be visible, stronger and longer dry seasons may be the filter that finally excludes wet origin species from dry sites.

Overall, these results are consistent with the physiological tolerance hypothesis, and add to the increasing amount of studies that strongly support the influence of drought and drought resistance for species distributions along moisture availability gradients (Engelbrecht et al. 2005, 2007, Brenes-Arguedas et al. 2009, Comita and Engelbrecht 2009, 2013, Kursar et al. 2009, Baltzer and Davies 2012, Condit et al. 2013). Understanding these processes will get more important under ongoing climate change, which is expected to increase drought and highlights the importance of long-term studies to understand the processes shaping species distribution patterns.

Pest pressure gradient hypothesis

According to the pest pressure gradient hypothesis, insect herbivores and pathogens benefit from more humid conditions in wet forests, leading to a gradient in pest pressure that positively correlates with rainfall. Plants growing in wet forests have adapted to the higher pest pressure and are highly defended. Plants growing in dry forests face lower pest pressure and do not have to invest so much in defenses, but are excluded from wet forests due to higher pest pressure.

In the seed transplant experiment (manuscript 1), exclusion of pests strongly increased germination and survival. This highlights that pests are a major factor influencing species performance in tropical forests. Wet species origins also had higher defenses than dry origin species (manuscript 2), as expected from the pest pressure gradient hypothesis. While the feeding trial also supported that shade tolerant species have higher defenses due to higher costs of replacing biomass in shady conditions (resource allocation theory (Barbour et al. 1980) based on the resource light Coley and Barone 1996), shade tolerance and species' origin were not correlated. Association of the species to soil nutrients, indicative of leaf nutrient content, did not influence the feeding rejection behavior of the caterpillars. Therefore, defenses were directly determined by pest pressure, independently from species' shade tolerance and leaf nutrient content. By showing higher defenses in wet forest species, the feeding trial also indirectly supports the idea that wet forests have higher pest pressure.

However, the results of the feeding trial in the laboratory were not confirmed by the results of the field experiment: while pests had a strong negative influence on performance, when comparing the effects of pests separately for the two sites and origins, the results of the seed transplant experiment (manuscript 1) did not support the pest pressure hypothesis. We had expected that dry origin species would suffer from pests especially in the wet site. However, the results showed exactly the opposite, with dry origin suffering from pests in their home range and not in the wet site. Also contrary to the expectations, wet origin species suffered strongly from pests, both in the dry and the wet site. These results can be explained by differential specialization of pests along the rainfall gradient. Growing in the wet forest, dry origin species may have escaped specialized pests in their home range (compare enemy release hypothesis, see Williamson 1997, Heger and Jeschke 2014). But regardless of the mechanism behind our results, higher pest pressure in wet forests did not exclude dry origin species from wet sites, as would have been necessary to support the pest pressure gradient hypothesis.

Previous studies analyzing the pest pressure gradient hypothesis also have found contradicting results. In a transplant experiment along the Isthmus of Panama, Brenes-Arguedas et al. (2009) found evidence for higher pest pressure in wetter forests: Both total leaf damage and leaf damage attributable to leaf chewing insects were higher in the rainier compared to the drier site, corroborating the first assumption of the pest pressure hypothesis. Mortality attributed to systemic pathogen attack was also higher in the wet compared to the dry site. Across the same rainfall gradient, Spear et al. (2015) also found a higher risk of infection by pathogens in the wetter compared to the drier forest. In contrast, another transplant experiment across a rainfall gradient in South-East Asia found no gradient in pest pressure, and pest pressure overall was very weak (Baltzer and Davies 2012). This challenges the view that higher pest pressure in wetter forests is a general pattern.

Neither leaf chewing insect nor pathogen attack were higher for dry origin compared to wet origin species in the transplant experiment of Brenes-Arguedas et al. (2009), refuting the second assumption of the pest pressure gradient hypothesis. Similarly, total herbivory of species with contrasting distribution patterns was not consistent with wet forest species having higher defenses in the transplant experiment in South-East Asia: in aseasonal forests, both origins had similar amounts of herbivory (Baltzer and Davies 2012). Provenance of the species did not significantly influence pest damage and mortality in both experiments. In the transplant experiment by Spear et al. (2015), the risk of being attacked by pathogens did not differ between species' origins, again challenging the idea that wet forest species are better defended. However, the mortality rate after being infected by pathogens was significantly higher for dry than for wet origin species, regardless of the site. The authors argued that this represents a potential mechanism of how pests influence species distribution patterns: not by differences in defenses, but by differences in tolerance to pest attack.

In summary, although pests strongly affected performance, and wet origin species had higher defenses, this study could not corroborate the pest pressure gradient hypothesis. Similarly, other studies have found contradicting results regarding the role of pest pressure in shaping species distribution patterns along rainfall gradients. While they may still play a role, they are certainly not the major factor and other processes must be more important.

Light availability hypothesis

The light availability hypothesis states that wet forests cast deeper shades than dry forests, due to differences in forest structure and cloud cover. Accordingly, wet origin species are more shade tolerant than dry origin species, and dry origin species are excluded from wet forests due to low light availabilities.

The wet site in the seed transplant experiment (manuscript 1) had indeed lower light availability than the dry site, but only during the dry season. This suggests that higher light availability in the dry site was based on a higher proportion of deciduousness in the dry forest. The first assumption of the light availability hypothesis, deeper shade in wet forests, was confirmed by the seed transplant experiment. However, in the seedling transplant experiment (manuscript 3), the correlation between rainfall and light availability was not significant (dry season) or only very weak (wet season), and the wettest site was also the one with the highest light availability values both in the dry and the wet season. This could be a consequence of the high soil nutrient concentrations in the wettest site, which increase the benefits of leaf shedding even under wet conditions, since rebuilding new leaves when resources are abundant is less costly. Leaf shedding increases light availability in the understory, as discussed before. Geology therefore may be at least as important as rainfall in determining the amount of light reaching the forest understory (Toledo et al. 2011). The combination of these results challenges the notion that lower light availability in wet forests is a general pattern.

In the seedling transplant experiment (manuscript 3), light availability had no differential effect on species with different origin. This indicates that dry origin species did not benefit more from higher light availabilities than wet origin species, as would have been expected from the second assumption of the light availability hypothesis. In the seed transplant experiment (manuscript 1), the overall establishment success was even slightly higher for wet origin species than for dry origin species with increasing light. However, none of the experiments supported that wet forest species are more shade tolerant. Together with the findings that wet forests are not necessarily darker than dry forests, this study does not support the light availability hypothesis.

Other studies comparing light availabilities between forests with differing rainfall do not lead to conclusive results. While some found a decrease of light availability with rainfall (Coomes and Grubb 2000, Santiago et al. 2004, Parker et al. 2005, Brenes-Arguedas et al. 2011, Peña-Claros et al. 2012), others found an increase or no pattern (Engelbrecht 1998, Harms et al. 2004). Additionally to rainfall, other factors like soil nutrients and texture are controlling forest structure, and lead to changes in light availability in the understory (Toledo et al. 2011). However, none of the studies was specifically designed to measure light availabilities along rainfall gradients (see also discussion in Brenes-Arguedas et al. 2011). The results of this study, although again not designed to capture the degree of variation in light within forests, adds to the others to show that a decrease of light availability with rainfall is not a general pattern.

The only study specifically testing the light availability distribution hypothesis did not find evidence that light shapes species distribution patterns (Brenes-Arguedas et al. 2011). While their wetter study sites had lower light and a lower variation in light availability than their drier sites, wet origin species were not more shade tolerant than dry origin species.

Although not consistent with the light availability distribution hypothesis, Brenes-Arguedas et al. (2011) describe a different mechanism of how light availability may influence species distributions: Under dry conditions high light exacerbated the negative effects of drought, a result found also in the seedling transplant experiment (manuscript 3). Therefore, high light in dry sites may, additionally to drought, filter drought-sensitive species.

Nutrient availability hypothesis

According to the nutrient availability hypothesis, wet forests have lower soil nutrient availability than dry forests, mostly due to higher leaching of nutrients with increasing rainfall. Accordingly, dry origin species are more nutrient-demanding than wet origin species, since they are used to higher nutrient concentrations. Therefore, they are excluded from wet forests due to low nutrient availability.

Soil phosphorus concentrations in the seedling transplant experiment (manuscript 3) decreased with increasing rainfall, as expected. However, the correlation was weak, and the wettest site was also the one with the highest soil phosphorus concentrations. Due to the complex geology of the Isthmus of Panama, young, phosphorus-rich bedrock patches may mask the leaching effect under higher rainfall. Panama therefore represents a special situation, and challenges the notion that low soil nutrients in wetter forests are a general pattern.

Dry origin species did not benefit more from higher soil phosphorus concentrations in the seedling transplant experiment (manuscript 3), and consequently were not more nutrient-demanding than wet origin species. This study therefore did not support the idea that low phosphorus in wet forests excludes dry origin species from wet sites.

Lower soil nutrients with increasing rainfall have been found along gradients in Hawaii and Ghana (Vitousek and Sanford 1986, Swaine 1996, Swaine and Becker 1999, Schuur and Matson 2001). Soil nutrients have been shown to significantly influence species performance (e.g. Alvarez-Clare et al. 2013, Yavitt and Wright 2008) and distribution (Condit et al. 2013). However, the only study specifically testing for the influence of nutrients on species distribution along rainfall gradients, did not find support for the hypothesis (Brenes-Arguedas et al. 2008). Dry origin species were outperformed by wet origin species in both low and high nutrient soils, indicating that dry origin species had no advantage under fertile conditions and consequently that they were not more nutrient-demanding (Brenes-Arguedas et al. 2008). This thesis and the study of Brenes-Arguedas et al. (2008) together offer no support for the nutrient availability distribution hypothesis.

Additional mechanisms that influence performance and distributions

Two additional factors influencing plant performance and/or distribution, which go beyond the tested hypotheses, were found in this study: Intrinsic differences in growth between dry and wet origin species, related to a drought tolerance-growth trade-off, may exclude dry origin species from wet forests. Differences in allocation patterns and/or interactions with mycorrhiza may increase performance under drought in nutrient-poor sites. These mechanisms are depicted in detail below.

Drought tolerance-growth trade-off

Dry origin species had lower growth rates than wet origin species both based on height (seed transplant experiment, manuscript 1) and leaf area (seedling transplant experiment, manuscript 3), a result found also in other studies (Baltzer et al. 2007, Brenes-Arguedas et al. 2008, 2009, Baltzer and Davies 2012). This indicates that lower growth rate of dry origin species is an intrinsic factor, and that dry origin species can not grow faster even when resources are abundant. Through this growth difference, wet origin species may over time outperform and exclude dry origin species from wet forests.

Dry origin species have been shown to be more drought-tolerant (Baltzer et al. 2007, 2008, Engelbrecht et al. 2007); lower growth rates could therefore be related to a drought tolerance-growth trade-off. According to the resource availability hypothesis, plants adapted to survive and reproduce under drought, i.e. drought-tolerant plants, are not able to grow fast even when resources are abundant (Smith and Huston 1989), due to trade-offs in e.g. investment in below- vs. aboveground biomass. Direct empirical evidence for a drought tolerance-growth trade-off remains surprisingly scarce. Support for a trade-off between drought survival and maximum growth rates or shoot growth rate across species was found e.g. by O’Brien et al. (2015), Polley et al. (2002) and Wikberg and Ögren (2004), in tropical tree seedlings, tropical and subtropical woody legumes, and in willows, respectively. Additionally, traits that confer drought tolerance like denser wood, small vessels and high non-structural carbohydrate concentrations are often related to low growth rates (Poorter et al. 2010, O’Brien et al. 2015). However, the only rigorous experimental test for this trade-off, which was conducted in eight desert grasses, did not find evidence for a trade-off

(Fernandez and Reynolds 2000). This is an important factor that has to be considered in future studies.

Differences in allocation patterns and interactions with mycorrhiza

In the seedling transplant experiment (manuscript 3), I found a puzzling result for the influence of soil phosphorus on seedling performance: During the wet season, soil phosphorus had no effect on performance, while during the dry season, it had a negative effect. Looking more carefully, I found that during the dry season the negative effect of phosphorus occurred only in the driest plots, while in the wet plots it had a positive effect. This suggests that phosphorus enhanced the negative effects of drought. Two mechanisms, alone or in combination, may be driving this pattern:

Plants, including tropical seedlings, growing in fertile environments usually shift biomass allocation towards aboveground parts at the expense of lower biomass allocation to roots, resulting in a lower root-shoot ratio (Chapin III 1980, Paz 2003, Wright et al. 2011, Poorter et al. 2012). Plants in high phosphorus sites may therefore have had a lower root/shoot ratio at the onset of the dry season and consequently poorer access to water than plants growing in less fertile sites.

Interactions with mycorrhiza may additionally have exacerbated the negative effects of drought under high phosphorus. It is long known that most plants in tropical forests develop mycorrhiza, predominantly of the vesicular-arbuscular type (Janos 1980, Read 1991, Herre et al. 2005). Mycorrhiza has positive effects on plant growth and survival (Janos 1980), due to increased nutrient supply of especially phosphorus, but also increased access to water for the plant (Read 1991, Ruiz-Lozano and Azcón 1995). The fungal partner of this mutualistic interaction benefits from carbohydrates produced by the plant. According to the functional equilibrium model (Johnson 2010), the benefit for each partner is related to the abundance of resources: under higher soil fertility conditions, plants develop less mycorrhiza (Treseder 2004). However, as mycorrhiza also enhances water availability of the plants (Read 1991, Ruiz-Lozano and Azcón 1995), reduced investment into mycorrhiza under fertile conditions may in turn render the seedlings more susceptible to drought. During the wet season, when water is not limiting, this may not influence plant performance. However, when the dry season starts, plants growing in fertile sites that did not invest in mycorrhiza may suffer stronger water shortage than plants that had developed more mycorrhiza due to limited soil fertility. Therefore, the observed negative effect of soil phosphorus on survival under dry conditions may be governed by an indirect interaction with mycorrhiza. To our knowledge, no study about interactions between soil water and phosphorus availability and interactions with mycorrhiza has been conducted in tropical forests so far. To test if this interactions are also responsible for species distributions, it would be necessary to see if plants from different origins have differential investment in mycorrhiza. More research is needed to test these hypothesized mechanisms.

The role of ontogeny for species distribution patterns

In this study, I focused on the seed-to-seedling transition and early seedling stage, because these are considered the most vulnerable to environmental stressors and a bottleneck in population dynamics (Harper 1977). It also has been shown that drought at the seedling stage excludes wet origin species from dry forests (Engelbrecht et al. 2005, 2007, Brenes-Arguedas et al. 2009, Comita and Engelbrecht 2009, 2013, Kursar et al. 2009, Baltzer and Davies 2012, Condit et al. 2013). However, support for the exclusion of dry origin species from wet forests at this life stage, represented by a home advantage of wet forest species in their home range, is contradictory with most studies finding no support for it (Brenes-Arguedas et al. 2008, 2009, 2011, Baltzer and Davies 2012, Spear et al. 2015). Similarly, neither the seed nor the seedling transplant experiment (manuscript 1 and 3) led to the conclusion that wet origin species have a survival home advantage at the earliest regeneration stages. As by the time they reach 1 cm diameter at breast height (dbh) tropical trees exhibit the distribution patterns they have as adults (Baldeck et al. 2013), later seedling/sapling stages must be the ones when dry forest species are excluded from wet forests.

Species responses to environmental conditions change with ontogeny (Comita et al. 2007). Differential responses of species with contrasting origin may therefore become more apparent at other life stages than the ones considered in the transplant experiments. Light requirements may increase with ontogeny, as a consequence of the cost of increasing supporting tissue with plant size (Lusk et al. 2008). Ontogenetic niche shifts towards nutrient-rich niches have been shown for temperate trees

(Bertrand et al. 2011). Defenses also change with ontogeny, with older seedlings and saplings having higher defenses than younger ones (Boege and Marquis 2005, Barton and Koricheva 2010, Mason and Donovan 2015). Indeed, while within the first year of establishment herbivory did not differentially affect establishment success of species with contrasting origins in this study (seed transplant experiment, manuscript 1), older seedlings/saplings (up to 1 m height) showed significant differences (feeding trial, manuscript 2), potentially reflecting a differential increase in defenses with ontogeny.

Processes excluding dry forest species from wetter forests may therefore act at later life stages than the first year seedlings we considered in our study (Daws et al. 2005, Comita et al. 2007), and may also accumulate over time.

Conclusions: What determines the distribution of species along tropical rainfall gradients?

Water availability clearly is the most important factor limiting species performance during the dry season, but also during the wet season. I also found indication that wet origin species suffer more from drought than dry origin species. However, differences in performance do not lead to differences in establishment success at the seed-to-seedling transition and early seedling stage or after one experimental year. Later life stages or longer time spans are needed for drought to exclude wet origin species from dry sites. Also stronger and longer dry seasons, like during El Niño events, may lead to the observed differences in species composition.

The effect of pest pressure on species distribution is less clear. Pests strongly affect species performance and wet origin species have higher defenses against insect herbivores. However, the effects of pest pressure were not strong enough to lead to an exclusion of dry origin species from wet sites, at least within the first year of seedling establishment. The effect of pest pressure may be more important for later ontogenetic stages, but more studies are needed to disentangle these effects.

This study found little support for the role of light in shaping species distributions. Light availability slightly increased performance, but only when water was not limiting; light reduced performance in the dry season, probably enhancing the negative effects of drought. Wet forests do not necessarily cast deeper shades than dry forests, as other factors like nutrient availability influence forest composition and consequent light transmittance. Additionally, dry origin species were not more light-demanding than wet origin species. Together, these results do not support that dry origin species are excluded from wet forests due to low light availability.

Overall, the role of nutrients in shaping species distributions along rainfall gradients is negligible. While there is a trend for wetter forests having lower nutrients, complex geology impedes this to be a general pattern. Also, dry origin species do not appear to be more nutrient-demanding than wet origin species. Therefore, low nutrient availability in wet forests is not the factor excluding dry origin species.

Another possible mechanism that may exclude dry origin species from wet forests are intrinsic growth differences due to a drought tolerance-growth trade-off. Drought tolerant dry origin species had consistently lower growth rates than drought intolerant wet origin species, especially in wetter forests. Wet origin species may therefore overgrow dry origin species in wet forests and lead in the long term to their exclusion.

In summary, water availability was conclusively found to be the most important factor limiting species distributions in tropical forests. A combination of drought and species-specific drought resistance excluded wet origin species from dry sites, while intrinsic performance trade-offs related to drought tolerance reduced competitive ability of dry origin species in wet forests, which in the long-term may lead to their exclusion.

Due to climate change, tropical forests will suffer from stronger and more prolonged dry seasons. This drying trend will have influences on species performance and consequent distribution through direct effects of decreased water availability, and may in the long term change forest composition and diversity. In contrast, indirect effects of drought through interaction with pests, light or nutrients will probably play a subordinate role.

References of the synopsis

Aerts, R. 1999. Interspecific competition in natural plant communities: mechanisms, trade-offs and plant-soil feedbacks. Journal of Experimental Botany 50:29–37.

Aerts, R., and F. Chapin III. 1999. The mineral nutrition of wild plants revisited: a re-evaluation of processes and patterns. Advances in Ecological Research 30:1–67.

Alvarez-Clare, S., M. C. Mack, and M. Brooks. 2013. A direct test of nitrogen and phosphorus limitation to net primary productivity in a lowland tropical wet forest. Ecology 94:1540–1551.

Amissah, L., G. M. Mohren, F. Bongers, W. D. Hawthorne, and L. Poorter. 2014. Rainfall and temperature affect tree species distribution in Ghana. Journal of Tropical Ecology 30:435–446.

Austin, A. T., and P. Vitousek. 1998. Nutrient dynamics on a precipitation gradient in Hawai'i. Oecologia 113:519–529.

Baldeck, C. A., K. E. Harms, J. B. Yavitt, R. John, B. L. Turner, R. Valencia, H. Navarrete, S. Bunyavejchewin, S. Kiratiprayoon, A. Yaacob, M. N. N. Supardi, S. J. Davies, S. P. Hubbell, G. B. Chuyong, D. Kenfack, D. W. Thomas, and J. W. Dalling. 2013. Habitat filtering across tree life stages in tropical forest communities. Proceedings of the Royal Society B: Biological Sciences 280:20130548.

Balderrama, S. I. V., and R. L. Chazdon. 2005. Light-dependent seedling survival and growth of four tree species in Costa Rican second-growth rain forests. Journal of Tropical Ecology 21:383–395.

Baltzer, J., S. Davies, S. Bunyavejchewin, and N. Noor. 2008. The role of desiccation tolerance in determining tree species distributions along the Malay-Thai Peninsula. Functional Ecology 22:221–231.

Baltzer, J. L., and S. J. Davies. 2012. Rainfall seasonality and pest pressure as determinants of tropical tree species' distributions. Ecology and Evolution 2:2682–2694.

Baltzer, J. L., S. J. Davies, N. S. M. Noor, A. R. Kassim, and J. V. LaFrankie. 2007. Geographical distributions in tropical trees: can geographical range predict performance and habitat association in co-occurring tree species? Journal of Biogeography 34:1916–1926.

Barbour, M. G., J. H. Burk, W. D. Pitts, F. S. Gilliam, and M. W. Schwartz. 1980. Terrestrial Plant Ecology. The Benjamin/Cummings Publishing Company, Inc., California, USA.

Barone, J. A. 1998. Effects of light availability and rainfall on leaf production in a moist tropical forest in central Panama. Journal of Tropical Ecology 14:309–321.

Barton, K. E., and J. Koricheva. 2010. The ontogeny of plant defense and herbivory: characterizing general patterns using meta-analysis. The American Naturalist 175:481–493.

Bertrand, R., J.-C. Gégout, and J.-D. Bontemps. 2011. Niches of temperate tree species converge towards nutrient-richer conditions over ontogeny. Oikos 120:1479–1488.

Bloor, J. M., and P. J. Grubb. 2003. Growth and mortality in high and low light: trends among 15 shade-tolerant tropical rain forest tree species. Journal of Ecology 91:77–85.

Boege, K., and R. J. Marquis. 2005. Facing herbivory as you grow up: the ontogeny of resistance in plants. Trends in Ecology & Evolution 20:441–448.

Bongers, F., L. Poorter, R. Rompaey, and M. Parren. 1999. Distribution of twelve moist forest canopy tree species in Liberia and Cote d'Ivoire: response curves to a climatic gradient. Journal of Vegetation Science 10:371–382.

Brenes-Arguedas, T., P. D. Coley, and T. A. Kursar. 2009. Pests vs. drought as determinants of plant distribution along a tropical rainfall gradient. Ecology 90:1751–1761.

Brenes-Arguedas, T., M. Rios, G. Rivas-Torres, C. Blundo, P. D. Coley, and T. A. Kursar. 2008. The effect of soil on the growth performance of tropical species with contrasting distributions. Oikos 117:1453–1460.

Brenes-Arguedas, T., A. B. Roddy, P. D. Coley, and T. A. Kursar. 2011. Do differences in understory light contribute to species distributions along a tropical rainfall gradient? Oecologia 166:443–456.

Bunker, D. E., and W. P. Carson. 2005. Drought stress and tropical forest woody seedlings: effect on community structure and composition. Journal of Ecology 93:794–806.

Burslem, D., P. Grubb, and I. Turner. 1996. Responses to simulated drought and elevated nutrient supply among shade-tolerant tree seedlings of lowland tropical forest in Singapore. Biotropica 28:636–648.

Cane, M. A., and S. E. Zebiak. 1985. A theory for El Niño and the Southern Oscillation. Science 228:1085–1087.

Chapin III, F. S. 1980. The mineral nutrition of wild plants. Annual Review of Ecology and Systematics 11:233–260.

Chazdon, R. L., and R. W. Pearcy. 1991. The importance of sunflecks for forest understory plants. BioScience:760–766.

Chust, G., J. Chave, R. Condit, S. Aguilar, S. Lao, and R. Pérez. 2006. Determinants and spatial modeling of tree β-diversity in a tropical forest landscape in Panama. Journal of Vegetation Science 17:83–92.

Coley, P. D. 1998. Possible effects of climate change on plant/herbivore interactions in moist tropical forests. Climatic Change 39:455–472.

Coley, P. D., and J. A. Barone. 1996. Herbivory and plant defenses in tropical forests. Annual Review of Ecology and Systematics 27:305–335.

Comita, L. S., R. Condit, and S. P. Hubbell. 2007. Developmental changes in habitat associations of tropical trees. Journal of Ecology 95:482–492.

Comita, L. S., and B. M. Engelbrecht. 2009. Seasonal and spatial variation in water availability drive habitat associations in a tropical forest. Ecology 90:2755–2765.

Comita, L. S., and B. M. Engelbrecht. 2013. Drought as a driver of tropical tree species regeneration dynamics and distribution patterns. Pages 261–308 *in* Coomes, David A and Burslem, David FRP and Simonson, William D, editor. Forests and Global Change. Cambridge University Press, Cambridge, UK.

Condit, R. 1998. Ecological implications of changes in drought patterns: shifts in forest composition in Panama. Climatic Change 39:413–427.

Condit, R., B. M. Engelbrecht, D. Pino, R. Pérez, and B. L. Turner. 2013. Species distributions in response to individual soil nutrients and seasonal drought across a community of tropical trees. Proceedings of the National Academy of Sciences of the United States of America 110:5064–5068.

Condit, R., S. P. Hubbell, and R. B. Foster. 1996. Changes in tree species abundance in a neotropical forest: impact of climate change. Journal of Tropical Ecology 12:231–256.

Condit, R., N. Pitman, E. G. Leigh, J. Chave, J. Terborgh, R. B. Foster, P. Nunez, S. Aguilar, R. Valencia, G. Villa, H. C. Muller-Landau, E. Losos, and S. P. Hubbell. 2002. Beta-diversity in tropical forest trees. Science 295:666–669.

Condit, R., W. D. Robinson, R. Ibáñez, S. Aguilar, A. Sanjur, R. Martínez, R. F. Stallard, T. García, G. R. Angehr, L. Petit, S. J. Wright, T. R. Robinson, and S. Heckadon. 2001. The Status of the Panama Canal Watershed and Its Biodiversity at the Beginning of the 21st Century. BioScience 51:389–398.

Condit, R., K. Watts, S. A. Bohlman, R. Pérez, R. B. Foster, and S. P. Hubbell. 2000. Quantifying the deciduousness of tropical forest canopies under varying climates. Journal of Vegetation Science 11:649–658.

Connahs, H., A. Aiello, S. Van Bael, and G. Rodriguez-Castaneda. 2011. Caterpillar abundance and parasitism in a seasonally dry versus wet tropical forest of Panama. Journal of Tropical Ecology 27:51–58.

Cook, K. H., and E. K. Vizy. 2008. Effects of twenty-first-century climate change on the Amazon rain forest. Journal of Climate 21:542–560.

Coomes, D. A., and P. J. Grubb. 2000. Impacts of root competition in forests and woodlands: a theoretical framework and review of experiments. Ecological Monographs 70:171–207.

Currie, D. J., G. G. Mittelbach, H. V. Cornell, R. Field, J.-F. Guégan, B. A. Hawkins, D. M. Kaufman, J. T. Kerr, T. Oberdorff, E. O'Brien, and J. Turner. 2004. Predictions and tests of climate-based hypotheses of broad-scale variation in taxonomic richness. Ecology Letters 7:1121–1134.

Davidson, R., Y. Mauffette, and D. Gagnon. 2002. Light requirements of seedlings: a method for selecting tropical trees for plantation forestry. Basic and Applied Ecology 3:209–220.

Daws, M. I., T. R. Pearson, D. F. P. Burslem, C. E. Mullins, and J. W. Dalling. 2005. Effects of topographic position, leaf litter and seed size on seedling demography in a semi-deciduous tropical forest in Panama. Plant Ecology 179:93–105.

Delire, C., A. Ngomanda, and D. Jolly. 2008. Possible impacts of 21st century climate on vegetation in Central and West Africa. Global and Planetary Change 64:3–15.

Dirzo, R. 1984. Herbivory: a phytocentric overview. Pages 141–165 *in* Rodolfo Dirzo, José Sarukhán, editor. Perspectives on Plant Population Ecology. Sinauer Associates Inc., Sunderland, Massachusetts.

Eichhorn, M. P., R. Nilus, S. G. Compton, S. E. Hartley, and D. F. Burslem. 2010. Herbivory of tropical rain forest tree seedlings correlates with future mortality. Ecology 91:1092–1101.

Engelbrecht, B. M. J. 1998. Ökologie und Ökophysiologie von koexistierenden Piper-Arten im Unterwuchs tropischer Regenwälder: "...wo der Pfeffer wächst..." Technische Universität Darmstadt, Darmstadt, Germany.

Engelbrecht, B. M. J., L. S. Comita, R. Condit, T. A. Kursar, M. T. Tyree, B. L. Turner, and S. P. Hubbell. 2007. Drought sensitivity shapes species distribution patterns in tropical forests. Nature 447:80–83.

Engelbrecht, B. M., T. A. Kursar, and M. T. Tyree. 2005. Drought effects on seedling survival in a tropical moist forest. Trees 19:312–321.

Fernandez, R. J., and J. F. Reynolds. 2000. Potential growth and drought tolerance of eight desert grasses: lack of a trade-off? Oecologia 123:90–98.

Fine, P. V., I. Mesones, and P. D. Coley. 2004. Herbivores promote habitat specialization by trees in Amazonian forests. Science 305:663–665.

Gentry, A. H. 1988. Changes in plant community diversity and floristic composition on environmental and geographical gradients. Annals of the Missouri Botanical Garden 75:1–34.

Gerencia de Hidrometeorología ETESA. 2015. Datos Climáticos Históricos. Available at: http://www.hidromet.com.pa/clima_historicos.php

Gilbert, G. 2005. Dimensions of plant disease in tropical forests. Pages 141–164 *in* Burslem, DFRP and Pinard, MA and Hartley, SE, editor. Biotic Interactions in the Tropics: Their Role in the Maintenance of Species Diversity. Cambridge University Press, Cambridge, UK.

Gillett, J. 1962. Pest pressure, an underestimated factor in evolution. Systematics Association Publication 4:37–46.

Givnish, T. J. 1988. Adaptation to sun and shade: a whole-plant perspective. Functional Plant Biology 15:63–92.

Givnish, T. J. 1999. On the causes of gradients in tropical tree diversity. Journal of Ecology 87:193–210.

de Gouvenain, R. C., R. K. Kobe, and J. A. Silander. 2007. Partitioning of understorey light and dry-season soil moisture gradients among seedlings of four rain-forest tree species in Madagascar. Journal of Tropical Ecology 23:569–579.

Graham, E. A., S. S. Mulkey, K. Kitajima, N. G. Phillips, and S. J. Wright. 2003. Cloud cover limits net CO2 uptake and growth of a rainforest tree during tropical rainy seasons. Proceedings of the National Academy of Sciences of the United States of America 100:572–576.

Grime, J. 1977. Evidence for the existence of three primary strategies in plants and its relevance to ecological and evolutionary theory. The American Naturalist 111:1169–1194.

Guijarro, J. A. 2013. climatol: Some Tools for Climatology: series homogenization, plus windrose and Walter&Lieth diagrams. R package version 2.2.

Han, W. X., J. Y. Fang, P. B. Reich, F. Ian Woodward, and Z. H. Wang. 2011. Biogeography and variability of eleven mineral elements in plant leaves across gradients of climate, soil and plant functional type in China. Ecology Letters 14:788–796.

Harms, K. E., J. S. Powers, and R. A. Montgomery. 2004. Variation in small sapling density, understory cover, and resource availability in four Neotropical forests. Biotropica 36:40–51.

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

Harpole, W. 2010. Neutral theory of species diversity. Nature Education Knowledge 3:60.

Heger, T., and J. M. Jeschke. 2014. The enemy release hypothesis as a hierarchy of hypotheses. Oikos 123:741–750.

Herre, E. A., D. Kyllo, S. Mangan, R. Husband, L. C. Mejia, and A.-H. Eom. 2005. An overview of arbuscular mycorrhizal fungal composition, distribution and host effects from a tropical moist forest. Pages 204–225 *in* Burslem, DFRP and Pinard, MA and Hartley, SE, editor. Biotic Interactions in the Tropics: Their Role in the Maintenance of Species Diversity. Cambridge University Press, Cambridge, UK.

Hidalgo, H. G., J. A. Amador, E. J. Alfaro, and B. Quesada. 2013. Hydrological climate change projections for Central America. Journal of Hydrology 495:94–112.

Holdridge, L. R. 1947. Determination of World Plant Formations From Simple Climatic Data. Science 105:367–368.

Hubbell, S. P. 2001. The Unified Neutral Theory of Biodiversity and Biogeography. Princeton University Press, New Jersey, USA.

Hulme, M., and D. Viner. 1998. A climate change scenario for the tropics. Climatic Change 39:145–176.

Huston, M. A. 1994. Biological Diversity: The Coexistence of Species. Cambridge University Press, Cambridge, UK.

Janos, D. P. 1980. Vesicular-arbuscular mycorrhizae affect lowland tropical rain forest plant growth. Ecology 61:151–162.

John, R., J. W. Dalling, K. E. Harms, J. B. Yavitt, R. F. Stallard, M. Mirabello, S. P. Hubbell, R. Valencia, H. Navarrete, M. Vallejo, and R. B. Foster. 2007. Soil nutrients influence spatial distributions of tropical tree species. Proceedings of the National Academy of Sciences of the United States of America 104:864–869.

Johnson, N. C. 2010. Resource stoichiometry elucidates the structure and function of arbuscular mycorrhizas across scales. New Phytologist 185:631–647.

Jones, M. M., S. Ferrier, R. Condit, G. Manion, S. Aguilar, and R. Pérez. 2013. Strong congruence in tree and fern community turnover in response to soils and climate in central Panama. Journal of Ecology 101:506–516.

Kawecki, T. J., and D. Ebert. 2004. Conceptual issues in local adaptation. Ecology Letters 7:1225–1241.

Kitajima, K., and L. Poorter. 2008. Functional basis for resource niche partitioning by tropical trees. Pages 160–181 *in* Carson, W. P. and Schnitzer, S. A., editor. Tropical Forest Community Ecology. Blackwell, Oxford, UK.

Kobe, R. K. 1999. Light gradient partitioning among tropical tree species through differential seedling mortality and growth. Ecology 80:187–201.

Kubisch, A., R. D. Holt, H.-J. Poethke, and E. A. Fronhofer. 2014. Where am I and why? Synthesizing range biology and the eco-evolutionary dynamics of dispersal. Oikos 123:5–22.

Kursar, T. A., B. M. Engelbrecht, A. Burke, M. T. Tyree, B. EI Omari, and J. P. Giraldo. 2009. Tolerance to low leaf water status of tropical tree seedlings is related to drought performance and distribution. Functional Ecology 23:93–102.

Leigh, E. G., P. Davidar, C. W. Dick, J. Terborgh, J.-P. Puyravaud, H. Steege, and S. J. Wright. 2004. Why do some tropical forests have so many species of trees? Biotropica 36:447–473.

Lewis, S. L. 2006. Tropical forests and the changing earth system. Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences 361:195–210.

Lieberman, M., D. Lieberman, R. Peralta, and G. S. Hartshorn. 1995. Canopy closure and the distribution of tropical forest tree species at La Selva, Costa Rica. Journal of Tropical Ecology 11:161–177.

Lusk, C., D. S. Falster, C. Jara-Vergara, M. Jimenez-Castillo, and A. Saldaña-Mendoza. 2008. Ontogenetic variation in light requirements of juvenile rainforest evergreens. Functional Ecology 22:454–459.

Malhi, Y., L. E. O. C. Aragão, D. Galbraith, C. Huntingford, R. Fisher, P. Zelazowski, S. Sitch, C. McSweeney, and P. Meir. 2009. Exploring the likelihood and mechanism of a climate-change-induced dieback of the Amazon rainforest. Proceedings of the National Academy of Sciences of the United States of America 106:20610–20615.

Malhi, Y., and J. Wright. 2004. Spatial patterns and recent trends in the climate of tropical rainforest regions. Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences 359:311–329.

Markesteijn, L. 2010. Drought Tolerance of Tropical Tree Species: Functional Traits, Trade-offs and Species Distribution. Wageningen, NL.

Marquis, R. J. 1984. Leaf herbivores decrease fitness of a tropical plant. Science 226:537–539.

Mason, C. M., and L. A. Donovan. 2015. Does investment in leaf defenses drive changes in leaf economic strategy? A focus on whole-plant ontogeny. Oecologia 177:1053–1066.

Montgomery, R., and R. Chazdon. 2002. Light gradient partitioning by tropical tree seedlings in the absence of canopy gaps. Oecologia 131:165–174.

Murphy, P. G., and A. E. Lugo. 1986. Ecology of tropical dry forest. Annual Review of Ecology and Systematics 17:67–88.

O'Brien, M. J., D. F. R. P. Burslem, A. Caduff, J. Tay, and A. Hector. 2015. Contrasting nonstructural carbohydrate dynamics of tropical tree seedlings under water deficit and variability. New Phytologist 205:1083–1094.

Ordoñez, J. C., P. M. Van Bodegom, J.-P. M. Witte, I. J. Wright, P. B. Reich, and R. Aerts. 2009. A global study of relationships between leaf traits, climate and soil measures of nutrient fertility. Global Ecology and Biogeography 18:137–149.

Orians, G. H., and O. T. Solbrig. 1977. A cost-income model of leaves and roots with special reference to arid and semiarid areas. The American Naturalist 111:677–690.

Paine, C. T., K. E. Harms, S. A. Schnitzer, and W. P. Carson. 2008. Weak competition among tropical tree seedlings: implications for species coexistence. Biotropica 40:432–440.

Pan, Y., R. A. Birdsey, J. Fang, R. Houghton, P. E. Kauppi, W. A. Kurz, O. L. Phillips, A. Shvidenko, S. L. Lewis, J. G. Canadell, P. Ciais, R. B. Jackson, S. W. Pacala, A. D. McGuire, S. Piao, A. Rautiainen, S. Sitch, and D. Hayes. 2011. A large and persistent carbon sink in the world's forests. Science 333:988–993.

Parker, G., C. Tinoco-Ojanguren, A. Martínez-Yrízar, and M. Maass. 2005. Seasonal balance and vertical pattern of photosynthetically active radiation within canopies of a tropical dry deciduous forest ecosystem in Mexico. Journal of Tropical Ecology 21:283–295.

Paz, H. 2003. Root/Shoot Allocation and Root Architecture in Seedlings: Variation among Forest Sites, Microhabitats, and Ecological Groups. Biotropica 35:318–332.

Peña-Claros, M., L. Poorter, A. Alarcón, G. Blate, U. Choque, T. S. Fredericksen, M. J. Justiniano, C. Leaño, J. C. Licona, W. Pariona, and others. 2012. Soil effects on forest structure and diversity in a moist and a dry tropical forest. Biotropica 44:276–283.

Polley, H. W., C. R. Tischler, H. B. Johnson, and J. D. Derner. 2002. Growth rate and survivorship of drought: CO2 effects on the presumed tradeoff in seedlings of five woody legumes. Tree Physiology 22:383–391.

Poorter, H., K. J. Niklas, P. B. Reich, J. Oleksyn, P. Poot, and L. Mommer. 2012. Biomass allocation to leaves, stems and roots: meta-analyses of interspecific variation and environmental control. New Phytologist 193:30–50.

Poorter, L. 1999. Growth responses of 15 rain-forest tree species to a light gradient: the relative importance of morphological and physiological traits. Functional Ecology 13:396–410.

Poorter, L., and Y. Hayashida-Oliver. 2000. Effects of seasonal drought on gap and understorey seedlings in a Bolivian moist forest. Journal of Tropical Ecology 16:481–498.

Poorter, L., I. McDonald, A. Alarcón, E. Fichtler, J.-C. Licona, M. Peña-Claros, F. Sterck, Z. Villegas, and U. Sass-Klaassen. 2010. The importance of wood traits and hydraulic conductance for the performance and life history strategies of 42 rainforest tree species. New Phytologist 185:481–492.

Pyke, C. R., R. Condit, S. Aguilar, and S. Lao. 2001. Floristic composition across a climatic gradient in a neotropical lowland forest. Journal of Vegetation Science 12:553–566.

Radulovich, R., and P. Sollins. 1991. Nitrogen and phosphorus leaching in zero-tension drainage from a humid tropical soil. Biotropica 23:84–87.

Read, D. J. 1991. Mycorrhizas in ecosystems. Experientia 47:376–391.

Rosindell, J., S. P. Hubbell, and R. S. Etienne. 2011. The unified neutral theory of biodiversity and biogeography at age ten. Trends in Ecology & Evolution 26:340–348.

Rüger, N., U. Berger, S. P. Hubbell, G. Vieilledent, and R. Condit. 2011. Growth strategies of tropical tree species: disentangling light and size effects. PloS One 6:e25330.

Rüger, N., A. Huth, S. P. Hubbell, and R. Condit. 2009. Response of recruitment to light availability across a tropical lowland rain forest community. Journal of Ecology 97:1360–1368.

Ruiz-Lozano, J., and R. Azcón. 1995. Hyphal contribution to water uptake in mycorrhizal plants as affected by the fungal species and water status. Physiologia Plantarum 95:472–478.

Sala, O. E., F. S. Chapin III, J. J. Armesto, E. Berlow, J. Bloomfield, R. Dirzo, E. Huber-Sanwald, L. F. Huenneke, R. B. Jackson, A. Kinzig, and others. 2000. Global biodiversity scenarios for the year 2100. Science 287:1770–1774.

Santiago, L. S., K. Kitajima, S. J. Wright, and S. S. Mulkey. 2004. Coordinated changes in photosynthesis, water relations and leaf nutritional traits of canopy trees along a precipitation gradient in lowland tropical forest. Oecologia 139:495–502.

Santiago, L. S., and S. S. Mulkey. 2005. Leaf productivity along a precipitation gradient in lowland Panama: patterns from leaf to ecosystem. Trees 19:349–356.

Santiago, L. S., E. A. Schuur, and K. Silvera. 2005. Nutrient cycling and plant-soil feedbacks along a precipitation gradient in lowland Panama. Journal of Tropical Ecology 21:461–470.

Santiago, L. S., S. J. Wright, K. E. Harms, J. B. Yavitt, C. Korine, M. N. Garcia, and B. L. Turner. 2012. Tropical tree seedling growth responses to nitrogen, phosphorus and potassium addition. Journal of Ecology 100:309–316.

Schleuning, M., J. Fründ, A.-M. Klein, S. Abrahamczyk, R. Alarcón, M. Albrecht, G. K. Andersson, S. Bazarian, K. Böhning-Gaese, R. Bommarco, and others. 2012. Specialization of mutualistic interaction networks decreases toward tropical latitudes. Current Biology 22:1925–1931.

Schuur, E. A., and P. A. Matson. 2001. Net primary productivity and nutrient cycling across a mesic to wet precipitation gradient in Hawaiian montane forest. Oecologia 128:431–442.

Smith, T., and M. Huston. 1989. A theory of the spatial and temporal dynamics of plant communities. Vegetatio 83:49–69.

Soepadmo, E. 1993. Tropical rain forests as carbon sinks. Chemosphere 27:1025–1039.

Sommer, J. H., H. Kreft, G. Kier, W. Jetz, J. Mutke, and W. Barthlott. 2010. Projected impacts of climate change on regional capacities for global plant species richness. Proceedings of the Royal Society B: Biological Sciences 277:2271–2280.

Spear, E. R., P. D. Coley, and T. A. Kursar. 2015. Do pathogens limit the distributions of tropical trees across a rainfall gradient? Journal of Ecology 103:165–174.

ter Steege, H., N. Pitman, D. Sabatier, H. Castellanos, P. Van der Hout, D. C. Daly, M. Silveira, O. Phillips, R. Vasquez, T. Van Andel, J. Duivenvoorden, A. A. De Oliveira, R. Ek, R. Lilwah, R. Thomas, J. Van Essen, C. Baider, P. Maas, S. Mori, J. Terborgh, P. N. Vargas, H. Mogollon, and W. Morawetz. 2003. A spatial model of tree alpha-diversity and tree density for the Amazon. Biodiversity and Conservation 12:2255–2277.

Sterck, F. J., L. Markesteijn, M. Toledo, F. Schieving, and L. Poorter. 2014. Sapling performance along resource gradients drives tree species distributions within and across tropical forests. Ecology 95:2514–2525.

Stewart, R. H., J. L. Stewart, and W. P. Woodring. 1980. Geologic map of the Panama Canal and vicinity, Republic of Panama. Miscellaneous Investigation Series:Map I–1232, 1 sheet, scale 1:100.000.

Suryatmojo, H., M. Fujimoto, Y. Yamakawa, K. Kosugi, and T. Mizuyama. 2013. Water balance changes in the tropical rainforest with intensive forest management system. International Journal of Sustainable Future for Human Security 1:56–62.

Swaine, M. D. 1996. Rainfall and soil fertility as factors limiting forest species distributions in Ghana. Journal of Ecology 84:419–428.

Swaine, M. D., and P. Becker. 1999. Woody life-form composition and association on rainfall and soil fertility gradients in Ghana. Plant Ecology 145:167–173.

Toledo, M., M. Peña-Claros, F. Bongers, A. Alarcon, J. Balcazar, J. Chuvina, C. Leaño, J. C. Licona, and L. Poorter. 2012. Distribution patterns of tropical woody species in response to climatic and edaphic gradients. Journal of Ecology 100:253–263.

Toledo, M., L. Poorter, M. Peña-Claros, A. Alarcón, J. Balcázar, C. Leaño, J. C. Licona, and F. Bongers. 2011. Climate and soil drive forest structure in Bolivian lowland forests. Journal of Tropical Ecology 27:333–345.

Treseder, K. K. 2004. A meta-analysis of mycorrhizal responses to nitrogen, phosphorus, and atmospheric CO2 in field studies. New Phytologist 164:347–355.

Turner, B. L., and B. M. Engelbrecht. 2011. Soil organic phosphorus in lowland tropical rain forests. Biogeochemistry 103:297–315.

Vitousek, P. M. 1984. Litterfall, nutrient cycling, and nutrient limitation in tropical forests. Ecology 65:285–298.

Vitousek, P., and R. Sanford. 1986. Nutrient cycling in moist tropical forest. Annual Review of Ecology and Systematics 17:137–167.

Wikberg, J., and E. Ögren. 2004. Interrelationships between water use and growth traits in biomass-producing willows. Trees 18:70–76.

Williamson, M. 1997. Biological Invasions (Population and Community Biology Series), 1996th edition. Springer.

Wright, S. J. 1992. Seasonal drought, soil fertility and the species density of tropical forest plant communities. Trends in Ecology & Evolution 7:260–263.

Wright, S. J. 2002. Plant diversity in tropical forests: a review of mechanisms of species coexistence. Oecologia 130:1–14.

Wright, S. J., H. C. Muller-Landau, and J. Schipper. 2009. The future of tropical species on a warmer planet. Conservation Biology 23:1418–1426.

Wright, S. J., and M. J. Samaniego. 2008. Historical, demographic, and economic correlates of land-use change in the Republic of Panama. Ecology and Society 13:17.

Wright, S. J., and C. P. Van Schaik. 1994. Light and the phenology of tropical trees. The American Naturalist 143:192–199.

Wright, S. J., J. B. Yavitt, N. Wurzburger, B. L. Turner, E. V. Tanner, E. J. Sayer, L. S. Santiago, M. Kaspari, L. O. Hedin, K. E. Harms, and others. 2011. Potassium, phosphorus, or nitrogen limit root allocation, tree growth, or litter production in a lowland tropical forest. Ecology 92:1616–1625.

Yavitt, J. B., and S. J. Wright. 2008. Seedling growth responses to water and nutrient augmentation in the understorey of a lowland moist forest, Panama. Journal of Tropical Ecology 24:19–26.

Zelazowski, P., Y. Malhi, C. Huntingford, S. Sitch, and J. B. Fisher. 2011. Changes in the potential distribution of humid tropical forests on a warmer planet. Philosophical Transactions. Series A, Mathematical, Physical, and Engineering Sciences 369:137–160.

Manuscript 1: Effects of drought, pest pressure and light availability on seedling establishment and growth: Their role for distribution of tree species across a tropical rainfall gradient

Effects of drought, pest pressure and light availability on seedling establishment and growth: Their role for distribution of tree species across a tropical rainfall gradient

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Abstract

Tree species distributions associated with rainfall are among the most prominent patterns in tropical forests. Understanding the mechanisms shaping these patterns is important to project impacts of global climate change on tree distributions and diversity in the tropics. Beside direct effects of water availability, additional factors co-varying with rainfall have been hypothesized to play an important role, including pest pressure and light availability. While low water availability is expected to exclude drought-intolerant wet forest species from drier forests (physiological tolerance hypothesis), high pest pressure or low light availability are hypothesized to exclude dry forest species from wetter forests (pest pressure gradient and light availability hypothesis, respectively). To test these hypotheses at the seed-to-seedling transition, the potentially most critical stage for species discrimination, we conducted a reciprocal transplant experiment combined with a pest exclosure treatment at a wet and a dry forest site in Panama with seeds of 26 species with contrasting origin. Establishment success after one year did not reflect species distribution patterns. However, in the wet forest, wet origin species had a home advantage over dry forest species through higher growth rates. At the same time, drought limited survival of wet origin species in the dry forest, supporting the physiological tolerance hypothesis. Together these processes sort species over longer time frames, and exclude species outside their respective home range. Although we found pronounced effects of pests and some effects of light availability on the seedlings, they did not corroborate the pest pressure nor light availability hypotheses at the seed-to-seedling transition. Our results underline that changes in water availability due to climate change will have direct consequences on tree regeneration and distributions along tropical rainfall gradients, while indirect effects of light and pests are less important.

Keywords

Plant-animal interaction, plant-pathogen interaction, community composition, rainfall gradients, tropical forests, tree distribution, water availability, specialized natural enemies

Introduction

Tropical forests are among the most diverse communities worldwide. Changes of rainfall and soil moisture with global climate change will potentially have dire consequences for tropical forests; however, the uncertainty of projections remains high (Malhi et al. 2009). One of the most prominent patterns in tropical forests is an increase of tree species richness with rainfall and a decrease with dry season intensity (e.g. ter Steege et al. 2003). At the same time, tree distribution and forest composition are strongly related to rainfall, and species turn-over is high across tropical rainfall gradients (Condit et al. 2002, 2013, Amissah et al. 2014, Fayolle et al. 2014). Understanding the mechanisms underlying tree distribution patterns, community composition and diversity across rainfall gradients is necessary to improve projections of the effects of global change on tropical forests and to optimize management, conservation and restoration strategies.

Several factors have been hypothesized to shape tree distribution patterns across rainfall gradients, including direct effects of water availability. According to the physiological tolerance hypothesis (Currie et al. 2004), drought-intolerant species are excluded from dry forests, thus leading to differences in species composition and species numbers among dry and wet forests. The direct role of drought tolerance, i.e. the ability to withstand periods of low water availability, in limiting wet forest species from occurring in forests with a pronounced dry season is supported by experimental studies (Engelbrecht et al. 2007, Baltzer et al. 2008, Sterck et al. 2014). However, at the same time many dry forest species do not occur in wet sites (Bongers et al. 1999, Engelbrecht et al. 2007, Toledo et al. 2012). The physiological tolerance hypothesis thus fails to explain a large part of variation of tree distribution (Engelbrecht et al. 2007) and the high species turnover observed across tropical rainfall gradients (Condit et al. 2002). Other environmental factors that co-vary with rainfall have been hypothesized to indirectly influence tree species distributions. These include increases of insect herbivore and pathogen pressure (summarized as pest pressure) and decreases of light availability with rainfall (Huston 1994, Coley and Barone 1996, Givnish 1999, ter Steege et al. 2003, Brenes-Arguedas et al. 2009, 2011, Baltzer and Davies 2012, Sterck et al. 2014).

Herbivores and pathogens have long been hypothesized to influence species distributions and diversity along tropical rainfall gradients (Coley and Barone 1996, Givnish 1999). According to the pest pressure gradient hypothesis (Baltzer and Davies 2012), species originating from dry forests with low herbivore pressure are less defended and therefore excluded from wet forests with high herbivore pressure. Despite its potential importance for explaining community compositions in tropical forest, empirical support for this hypothesis remains scarce. Evidence for changes of pest pressure with rainfall or moisture remains contradictory, and no differences of herbivore nor pathogen damage between species origins have been found in reciprocal transplant experiments at the seedlings stage, indicating that defenses did not differ between species of dry, seasonal and wet, aseasonal forests (Brenes-Arguedas et al. 2009, Baltzer and Davies 2012, Spear et al. 2015). Thus, the relevance of the pest pressure hypothesis for explaining species distributions remains to be shown.

Light availability has been hypothesized to influence species distributions along rainfall gradients, by excluding light-demanding dry origin species from wet forests with low understory light levels (Huston 1994, Brenes-Arguedas et al. 2011). Higher light requirements of dry forest species have been hypothesized as a consequence of a trade-off between shade and drought tolerance (Smith and Huston 1989, Huston 1994), based mainly on a trade-off between biomass allocation to roots, which would confer drought tolerance, and allocation to leaves, which confers shade tolerance. However, there is no conclusive support for a trade-off between drought and shade tolerance in tropical forest plants (Engelbrecht et al. 2007, Markesteijn and Poorter 2009, Markesteijn et al. 2011, Amissah et al. 2015), as traits conferring drought or shade tolerance are complex, not necessarily related and can be uncoupled. Higher light requirements of dry forest species have also been hypothesized due to their evolution in higher light environments in dry forests (Smith and Huston 1989, Brenes-Arguedas et al. 2011). Although lower light conditions in wetter forests have long been assumed (Smith and Huston 1989, Coomes and Grubb 2000), few studies have directly compared light availability along rainfall gradients (Harms et al. 2004, Brenes-Arguedas et al. 2011). The results do not support that there is a general pattern (Harms et al. 2004). Instead, nutrients and species composition additionally strongly influence forest structure and understory light availability (Toledo et al. 2011). Thus, the role of light in shaping species distributions across rainfall gradients also remains unclear.

Apart from environmental factors, intrinsic trade-offs between stress tolerance and growth rate (Grime 1977, Smith and Huston 1989), may also lead to exclusion of drought-tolerant dry origin species from wet forests. Adaptations to stressful, resource-limited environments have been hypothesized to be coupled with intrinsically low growth rates, based on biomass investment into either roots, which confer higher drought (stress) tolerance or into leaves, which allows for higher growth rates. Other traits that confer drought tolerance, like high wood density, small vessel diameter or high non-structural carbohydrate concentrations, are also associated with low growth rates (Poorter et al. 2010, O'Brien et al. 2015). Thus, drought-tolerant dry origin species should have intrinsically lower growth rates, which put them at a disadvantage when water is not limiting as in wet forests. Under such conditions, they may thus be outcompeted by drought-intolerant, fast-growing wet origin species. However, at the level of whole-plant performance, evidence for a drought tolerance-growth trade-off and its role for species distributions across rainfall gradients remains scarce and contradictory (e.g. Fernandez and Reynolds 2000, Polley et al. 2002, Wikberg and Ögren 2004).

Plants responses to drought, pest pressure and light availability differ among life stages. Early life stages, especially seedling emergence, are considered vulnerable to abiotic and biotic stressors (Harper 1977, Daws et al. 2005, Spear et al. 2015), and may thus be critical in shaping species distributions. Plant defenses often increase with ontogeny (Barton and Koricheva 2010), and the same absolute amount of leaf damage should have larger impact on small seedlings compared to bigger, older plants, thus rendering initial life stages especially vulnerable to pests. Experimental studies on factors shaping tree distributions across rainfall gradients have so far mainly focused on established seedlings (Engelbrecht et al. 2007, Baltzer et al. 2008, Brenes-Arguedas et al. 2008, 2009, 2011, Baltzer and Davies 2012, but see Spear et al. 2015). In our study we therefore specifically focused on the role of seed-to-seedling transition and first-year establishment for distribution patterns.

The aim of this study was to test how the combined effects of drought, pests and light availability affect early seedling performance of tree species with contrasting origins (dry vs. wet), and how these differences in seedling performance influence species distribution patterns. We hypothesized that species have a performance advantage within their respective home (native) range compared to foreign (alien) species, resulting in exclusion of the foreign species. We expected that drought limits performance of wet forest species in drier sites (physiological tolerance hypothesis), and that pests and/or light availability limits the performance of dry forest species in wetter sites (pest pressure and light availability hypothesis, respectively). To test these hypotheses, we conducted a reciprocal transplant experiment along a rainfall gradient in Panama, with species with contrasting origins. Pests were excluded for half of the seeds, and light and soil moisture conditions were monitored during one year, including a dry and a wet season. Specific expectations for plant performance in the experiment are depicted in Figure 1.

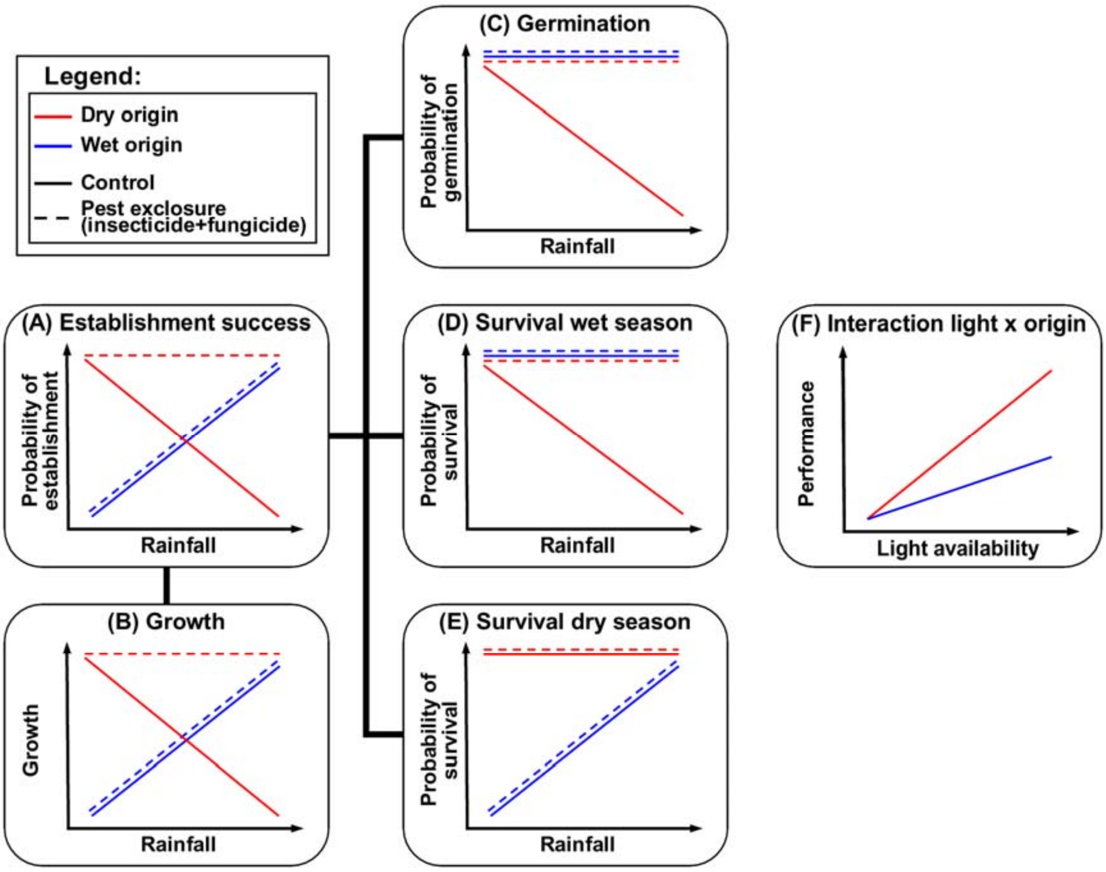

Figure 1: Schematic representation of the specific hypotheses for the effects of drought, light and pests on seedling performance. For overall probability of establishment success (A) and growth (B), we hypothesized that species perform better in their respective home range outcompeting foreign species, with drought limiting wet origin species in the dry site, and pest damage limiting dry origin species in the wet site. Consequently, we expected that the pest exclosure enhances performance only for (poorly defended) dry origin species in the wet site (with high pest pressure) (i.e. three-way interaction between treatment (exclosure/control), origin (dry/wet) and site (dry/wet)). For germination (C) and survival in the wet season (D), when water availability and pest pressure are assumed to be high, we expected that wet origin species have higher survival than dry origin species in the wet site, under control conditions (site x origin interaction). We expected that (poorly defended) dry origin species are limited by pest pressure in the wet site, indicated by higher performance when pest pressure is alleviated through pest exclosure (three-way site x origin x treatment interaction). In contrast, (well defended) wet origin species exhibit no differences in germination/survival between sites, independent of the pest exclosure. We expected dry season survival (E) of (drought-sensitive) wet origin species to be lower than survival of dry origin species in dry sites (significant site x origin interaction). Because pest pressure is assumed to be lower in the dry season, we expected no increase in survival with pesticide treatment for any combination of site and origin (no three-way interaction). With increasing light availability (F) we expected a stronger increase in all performance parameters for dry origin species (light x origin interaction), reflecting their higher light requirements.

Materials and methods

Study sites

The study was conducted at the Isthmus of Panama, which exhibits a pronounced rainfall gradient from 1600 mm/year at the Pacific Coast to over 3000 mm/year at the Atlantic coast across a distance of only 65 km (Engelbrecht et al. 2007, Condit et al. 2013). The length of the dry season, which typically starts in January and ends in May, correlates negatively with annual rainfall (Engelbrecht et al. 2007). Mean annual temperature is 25°C with little variation across the gradient or throughout the year.

The experiment was conducted in two forests about 50 km apart: a drier semi-deciduous forest located in the national park Camino de Cruces (9° 2'N, 79°35'W, 2000 mm annual rainfall; modeled based on BIOCLIM data, Hijmans et al. 2005), and a wetter evergreen lowland forest in the national park San Lorenzo (9°16'N, 79°58'W, 3200 mm annual rainfall). Both sites are located in the Tropical Moist Forest Life Zone (Holdridge and Budowski 1959, Pyke et al. 2001). However, rainfall and moisture regime, as well as species composition vary greatly. Dry season length is approximately 150 and 120 days, and rainfall in the driest quarter of the year 530 mm and 800 mm (Pyke et al. 2001, Engelbrecht et al. 2007), respectively. Soil water potentials in the upper soil layer of the dry site reach values well below -2 MPa in the dry season, but remain high throughout the year in the wet site (Engelbrecht, unpublished data). Both forest sites were mature secondary forest located on sedimentary bedrock. Only about 10% of the species in the areas overlap (Pyke et al. 2001, Condit et al. 2002). In the following we refer to these sites as “dry” and “wet”, respectively. Permits for working in the national parks were granted by the “Autoridad Nacional del Ambiente (ANAM).

Experimental design

At each forest site, 60 paired plots (90 cm x 90 cm) were established, with each pair including a pesticide treatment (fungal pathogen and insect herbivore exclosure, see below) and a control plot (2 sites x 2 treatments x 30 plots). Seeds of 15 species with "wet origin" and 11 species with "dry origin" (Table S1, for definitions see below) were sown into each plot, with one seed of each species in each plot. Germination, seedling survival in the dry and the wet season, and growth were followed over one year.

Seedling plots

The plots were set-up in the forest understory avoiding any gaps, with pairs separated at least 70 m from each other, spanning an overall area of about 300 ha in San Lorenzo (wet site) and 150 ha in Cruces (dry site). The control and exclosure plots were separated by at least 2 m. Where relevant, the control plots were set-up uphill from the exclosure plots to prevent runoff from the treatment to the control (three times in San Lorenzo and two times in Cruces). To allow access of insect herbivores but prevent seed or seedling removal by rodents or other mammals, which were not the focus of this study, all plots were caged with 2.5 x 2.5 cm wire mesh.

Study species and plant material

Study species were selected to include common species with strong association to the dry or the wet side of the isthmus. We focused on shade-tolerant species, since they represent about 80% of the species in these forests (Welden et al. 1991, Bongers et al. 2005). Species with small seeds (< 0.5 cm length) were excluded to facilitate their manipulation and localization in the field. Potential study species were selected based on their occurrence in 50 1 ha plots spanning the rainfall gradient (CTFS 2015) and/or their abundance in a wet and a dry forest plot (Sherman, 6 ha and Cocoli, 4 ha, respectively, see CTFS 2015). Species with predominantly wet Caribbean side occurrence, that did not occur on the dry Pacific side of the Isthmus, or that had at least double the abundance in the wet than the dry side plots were classified as "wet origin species", whereas species occurring predominantly on the dry Pacific side, that did not occur on the wet Caribbean side of the Isthmus, or that had at least double the abundance in the dry than the wet side plots were classified as "dry origin species" (Table S1).

Seeds were collected in mature secondary forests across the Isthmus within their respective natural home range in the national parks San Lorenzo, Soberania, Chagres and Camino de Cruces during the dry season and beginning of the wet season 2012 (March to mid-May). Ripe seeds were collected from a minimum of three mother trees per species by directly harvesting from the tree, or from freshly fallen fruits. Damaged seeds were removed after visual inspection for damage by predators or pathogens. Final selection of the study species was based on the availability of enough undamaged seeds, resulting in 15 "wet" and 11 "dry" origin species.

Pest exclosure treatment

To exclude fungal pathogens and insect herbivores (summarized as pests) a combination of a fungicide and an insecticide (summarized as pesticides) was applied monthly to the treatment plots. Actara (active ingredient: Thiamethoxam), a systemic broad-spectrum insecticide, and Diligent (active ingredients: Methalaxyl and Chlorothalonil), a systemic broad-spectrum fungicide with protectant properties effective against true fungi as well as oomycetes were used. According to the specification of the manufacturer, Actara was used in a solution of 0.5 g/l water, and Diligent in a solution of 5 g/l. Each exclosure plot was sprayed with a mixture of 40 ml of the insecticide and 40 ml of the fungicide solution. The control plots were sprayed with the same amount of rainwater, to ensure that results were not biased by additional water availability in the treatment. Studies using similar pesticide treatments have discarded negative influences on non-target organisms, including the plants themselves (Bagchi et al. 2014, Gripenberg et al. 2014). Seeds in the exclosure plots were additionally pre-treated with the broad-spectrum insecticide Brigadier (active ingredient: Bifenthrin), and the fungicide Diligent (see above) to avoid seed predation. The insecticide was used undiluted with 50 ml/kg seeds. The seeds were briefly soaked in both solutions. Seeds of the control plots were soaked in rainwater.

Seed sowing

120 seeds per species (2 sites x 2 treatments x 30 plots, with one seed per species per plot, totaling 3600 seeds) were sown at the end of the dry season/beginning of the wet season (starting in March 2012). To ensure high germination rates in the typically

recalcitrant seeds and to mimic natural seeding periods, seeds were sown as soon as possible after collection (maximum two days later), and distributed evenly between exclosure and control plot and wet and dry site (i.e. not all seeds of one species were planted at the same time). Seeds were planted on the mineral soil under the leaf litter, in a 15 x 15 cm grid, with species assigned randomly to the positions. Leaf litter was disturbed as little as possible to ensure natural microhabitat conditions in the plots. To prevent washing away and to facilitate relocation, seeds were fixed to the ground with wooden toothpicks and positions marked.

Seed germination, survival and growth

Seed germination and seedling survival were monitored between March 2012 and April 2013, i.e. during the transition between initial dry and wet season, a wet season and a second dry season. Rainfall during the study period did not differ substantially from the long-term average, except for almost the double amount of rainfall in November and December 2012 (ESP 2015). During the first 3.5 months, the time of highest germination, censuses were conducted biweekly to ensure that all germinating seeds were recorded; radicle emergence was counted as germination. Thereafter, censuses were conducted at monthly intervals for seedling survival, based on aboveground living biomass, and for occasional further germination. Overall growth was assessed at the last census based on seedling height, measured from the ground to the highest meristem.

From the census data we quantified six performance parameters: (1) *overall establishment success* (proportion of remaining seedlings at the end of the experiment relative to the original number of seeds sown; covers the period from March 2012 to April 2013); (2) *overall growth* (height of the seedlings at the end of the experiment in April 2013); (3) *germination* (proportion of seeds that germinated until the end of the experiment, relative to the original number of seeds sown); (4) *survival during the wet season* (proportion of seedlings that survived until December 2012, relative to the number of germinated seeds); and (5) *survival during the dry season* (proportion of seedlings that survived until April 2013, relative to the number of seedlings present at the start of the dry season).

Soil moisture and light

We recorded gravimetric soil moisture at each census, and averaged over the dry and wet seasons, respectively. For light availability, canopy openness was assessed once during the dry and wet season, respectively (Figure 2).

For soil moisture, three random soil cores 15 cm deep were taken within 1 m from each plot pair; fresh and dry weight (after drying to constant weight at 105°C) were determined and percent gravimetric water content was calculated based on dry weight. We assessed canopy openness (percent open sky) from hemispherical photographs taken 1 m over each plot during the dry (April 2012) and during the wet season (October 2012), using a Nikon Coolpix P5000 camera with a Fisheye Converter FC-E8. Photographs were analyzed with the program Gap Light Analyzer v2.

Gravimetric soil moisture and canopy openness varied significantly across sites and between seasons: gravimetric moisture was lower in the dry than in the wet forest and lower in the dry than in the wet season (Figure 2 a). Conversely, canopy openness was higher in the dry than in the wet forest, and higher in the dry than in the wet season (Figure 2 b).

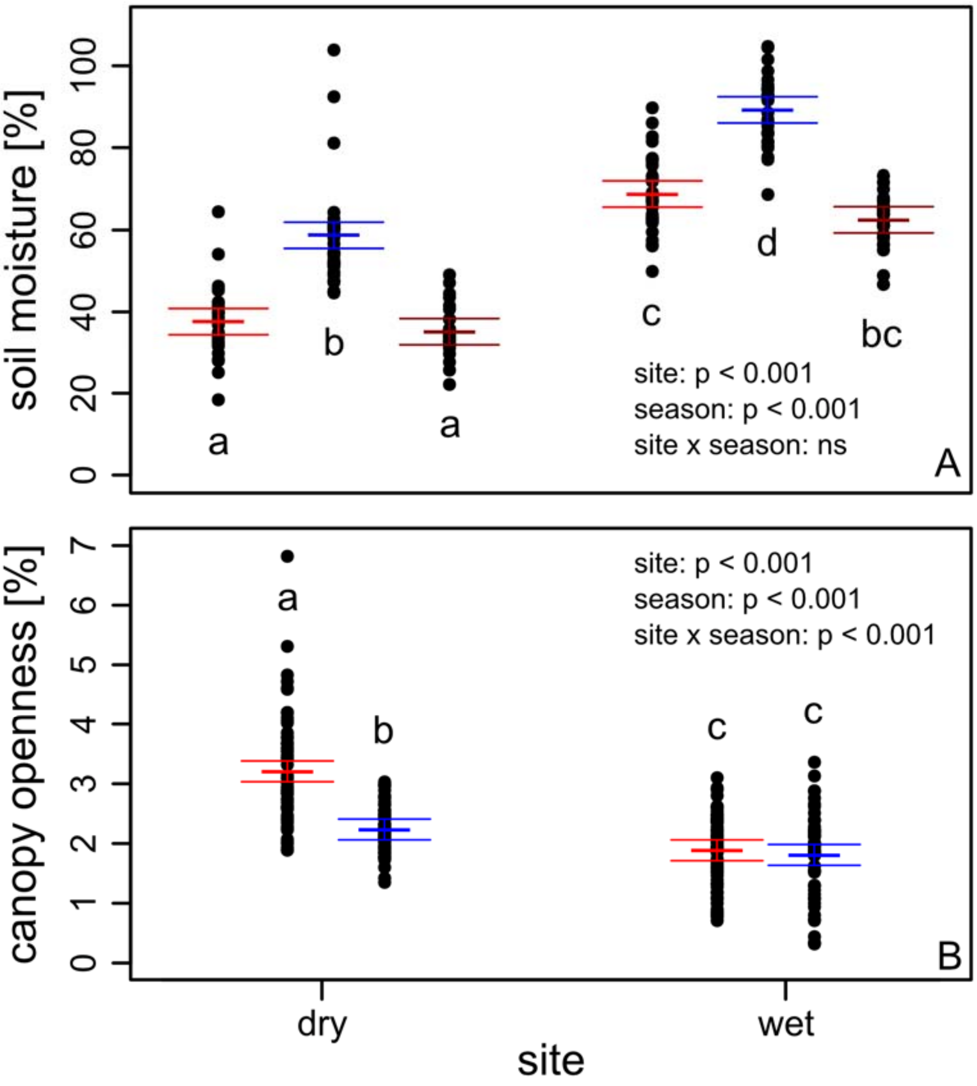

Figure 2: Gravimetric soil moisture (A) and canopy openness (B) in the dry and wet site across seasons. Colors indicate the end of the dry season 2012 (red), wet season 2012 (blue) and dry season 2013 (dark red). Included are results of an ANOVA for effects of site, season and site x season interactions. Different letters represent significant differences at the 0.05 level in a Tukey post-hoc test. Presented are means (thick horizontal lines), 95% CI (thin lines), and raw data (points).

Statistical analyses

Our main aim was to assess the effect of site (as a categorization of moisture, see below), species origin, pest exclosure and light on probabilities of germination and survival, as well as on growth.

We initially tested for correlations between the explanatory variables (Table S2). Soil moisture correlated strongly between seasons as well as with the factor site ($r \geq 0.8$, see Table S2). Therefore, soil moisture and site could not be maintained together in the same model. Models treating soil moisture as continuous variable and models with the factor site (wet/dry, see below) yielded qualitatively the same results. We present results of the models with the factor site, because we were interested in responses to the large-scale rainfall gradient rather than small-scale responses within sites, and because using the factor site better reflected the experimental setup with a separate "dry" and "wet" site.

The performance parameters we analyzed were: (1) *establishment success*, (2) *growth*, (3) *germination*, (4) *survival during the wet season* and (5) *survival during the dry season* (Table 1). To account for heteroscedasticity and non-normality of the residuals, growth data was log-transformed using the natural logarithm.

We initially assessed species effects on performance by fitting a separate model for each performance parameter (Generalized linear mixed effects with binomial distribution (GLMM), or linear mixed effects (LMM) for growth, see Figure S1). Species was used as fixed effect factor in each model. Random intercepts were plot-pairs and plots, with plot nested in plot-pairs.

To assess the effects of site, origin, treatment and light availability on performance, one model per performance parameter was fitted (GLMM or LMM, respectively). For every model, fixed effect factors were site (dry/wet), origin of the species (dry/wet), treatment (pest exclosure/control) and the average light availability (canopy openness in %) for the period analyzed (dry season, wet season, annual mean of dry and wet season, respectively). We also included the triple interaction term site x origin x treatment (which includes the pairwise interactions site x origin, site x treatment and origin x treatment), and the interaction term origin x light availability (Table 1). Random intercepts were species, plot-pair and plot. We nested species in plots and plots in plot-pairs. Single term deletion of non-significant terms was used for model selection. We removed sequentially first all interaction terms and then all explanatory variables that led to a model with a lower Akaike Information Criteria (AIC).

Table 1: Effects of site, species origin, pest exclosure and light on performance parameters of tropical tree seedlings.

	Establishment success	**Growth**	**Germination**	**Survival wet season**	**Survival dry season**
Single term effects					
Site	ns	**<0.001** (-)	<0.1 (+)	ns	ns
Origin	ns	**<0.05** (+)	ns	ns	**<0.05** (-)
Treatment	**<0.001** (+)	ns	**<0.001** (+)	**<0.05** (+)	**<0.001** (+)
Canopy openness	ns	ns	ns	**<0.05** (+)	ns
Interactions					
Site x origin	ns	**<0.01**	ns	ns	**<0.05**
Site x treatment	**<0.01**	ns	**<0.01**	ns	ns
Origin x treatment	ns	ns	ns	ns	ns
Origin x canopy openness	<0.1	ns	ns	ns	ns
Site x origin x treatment	**<0.01**	ns	<0.1	<0.1	ns

Summary of the results of the Generalized Linear Mixed Effects Models (GLMM) and Linear Mixed Effects Model (LMM, for growth) for the six performance parameters. Significant relations are in bold. Detailed results are given in Table S3.
(+) / (-): positive or negative effect of pest exclosure, wet site, wet origin or high light on performance parameters. These are only given for single-term results, not for the interactions.

To test our specific expectations (Figure 1), we conducted eight planned comparisons using least squares means (Lenth 2014) with Tukey correction as post-hoc tests (Table 2). To assess if seed germination and seedling survival varied across sites or with species origin under the *natural condition* of the habitat, we assessed under control conditions the effect of origin of the species within the dry and the wet site, respectively, and the effect of site on wet origin and dry origin species, respectively (i.e. four contrasts, Table 2 a and b). To assess to what extent germination and survival were affected by pests, we assessed the pest exclosure effect in each site (wet/dry) and in species with different origin (wet/dry, i.e. four contrasts, Table 2 c and d). Tukey post-hoc comparisons, means and standard errors in tables and figures are from the least squares table (Lenth 2014).

Table 2: Planned comparisons of (A) effects of origins and sites under natural (control) conditions, and (B) effects of pest exclosure on performance parameters.

	A. Effects of site and species origin under natural conditions				**B. Effects of pest exclosure within sites and origins**			
	a. Effect of origin within sites		**b. Effect of site within origin**		**c. Exclosure effect within dry site**		**d. Exclosure effect within wet site**	
Performance parameter	**Dry site**	**Wet site**	**Dry origin**	**Wet origin**	**Dry origin**	**Wet origin**	**Dry origin**	**Wet origin**
Establishment success	ns	ns	ns	ns	**<0.001**	**<0.001**	ns	**<0.001**
Growth	**0.02**	**<0.001**	**<0.001**	ns	ns	ns	ns	ns
Germination	ns	ns	0.06	ns	**<0.001**	ns	ns	ns
Survival wet season	ns	ns	ns	0.05	**0.02**	**0.001**	ns	**<0.001**
Survival dry season	**0.02**	ns	ns	**0.04**	0.09	**0.03**	ns	**<0.001**

(a) Effects of species origin within the dry and the wet site, and (b) effects of site on species with dry and wet origin under control conditions. Effects of exclosure (c) within the dry site on dry origin and wet origin species, and (d) within the wet site, on dry and wet origin species. Post-hoc analyses are based on least squares means contrasts (Lenth 2014) with Tukey correction. Significant contrasts are in bold.

All statistical tests were done using R 3.0.2 (R Core Team 2013) with the packages lme4 1.0.5 (Bates et al. 2013), lsmeans 2.00-5 (Lenth 2014) and LMERConvenienceFunctions 2.5 (Tremblay et al. 2013).

Results

Species differed significantly in overall establishment success, as well as in germination, dry and wet season survival, and growth (all $p < 0.001$, Figure S1). Pest exclosure (i.e. pesticide treatment) had an overall significant positive effect on all performance parameters except growth (Table 1), underlining the importance of herbivores and pathogens in limiting seed germination and seedling survival in tropical forests. Light availability (i.e. canopy openness) only had a positive effect on wet season survival (Table 1). Several performance parameters were affected by site and origin or by interactions between site, origin, treatment and light, but these effects differed among performance parameters (Table 1).

Below we first present the results for overall seedling establishment and growth during the study (Figure 3). The establishment success after one year is the cumulative result of germination and survival patterns, which are presented separately (Figure 4), and integrates processes in the wet and the dry season over the course of the experiment (see also Figure 1). To test our main hypotheses, we first focus on species performance under natural pest pressure (i.e. controls) to compare the performance of home vs. foreign species (i.e. origins) within sites and across sites (planned comparisons in Table 2 a and b). Then we focus on the effects of pest exclusion within sites and across origins (planned comparisons in Table 2 c and d). Finally, we depict the effects of light. Full results of the three-way interactions, as well as of pairwise interactions and individual factors are summarized in Table 1, and details are given in Table S3.

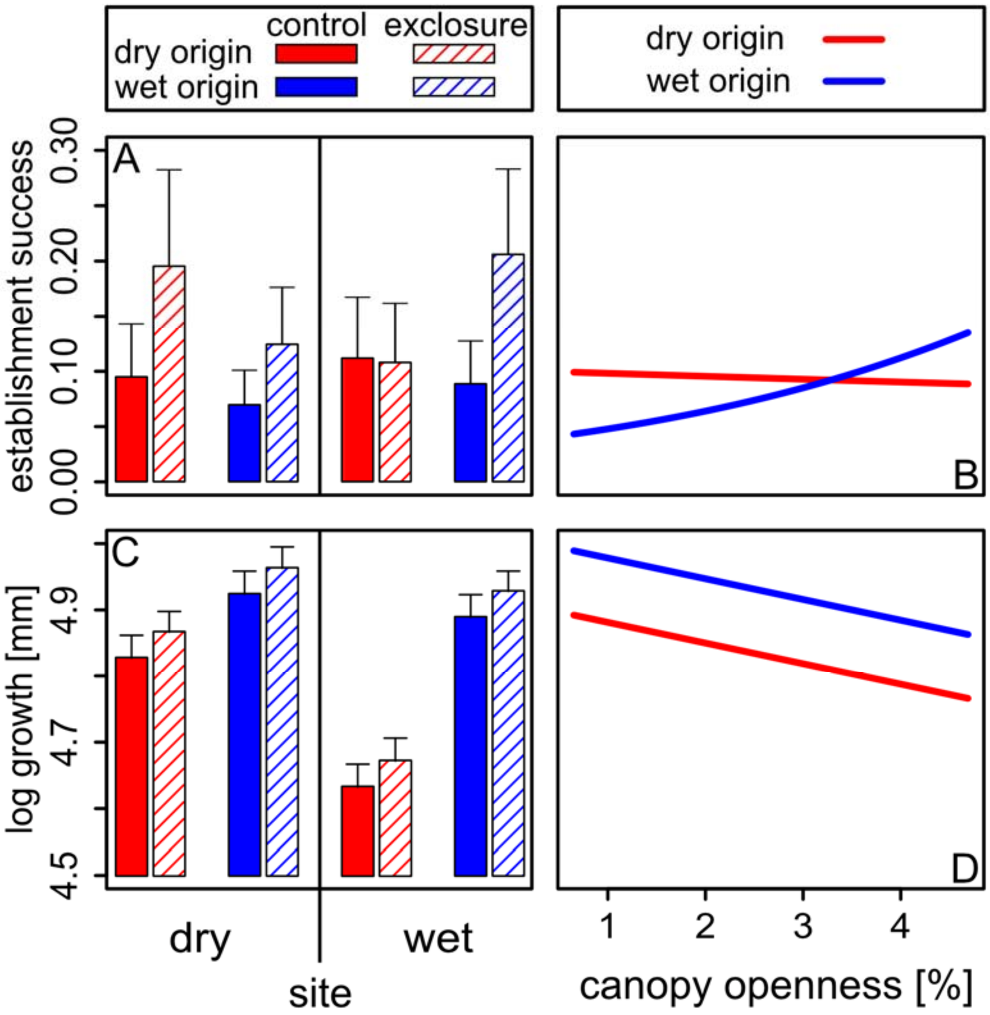

Figure 3: Overall probability of establishment success (A and B) and growth (C and D) at the end of the experiment after one year, as affected by moisture (dry vs. wet site), origin (dry vs. wet), pest exposure (control vs. exclosure) and light availability (canopy openness). Panels A and B show means and standard errors from the least squares means table (Lenth 2014). For canopy openness (B and D), results of exclosure and control seeds and seedlings were pooled, since we did not expect light availability to influence the effect of the exclosure treatment. For overall analyses see Table 1, for planned contrasts (post-hoc-tests) see Table 2.

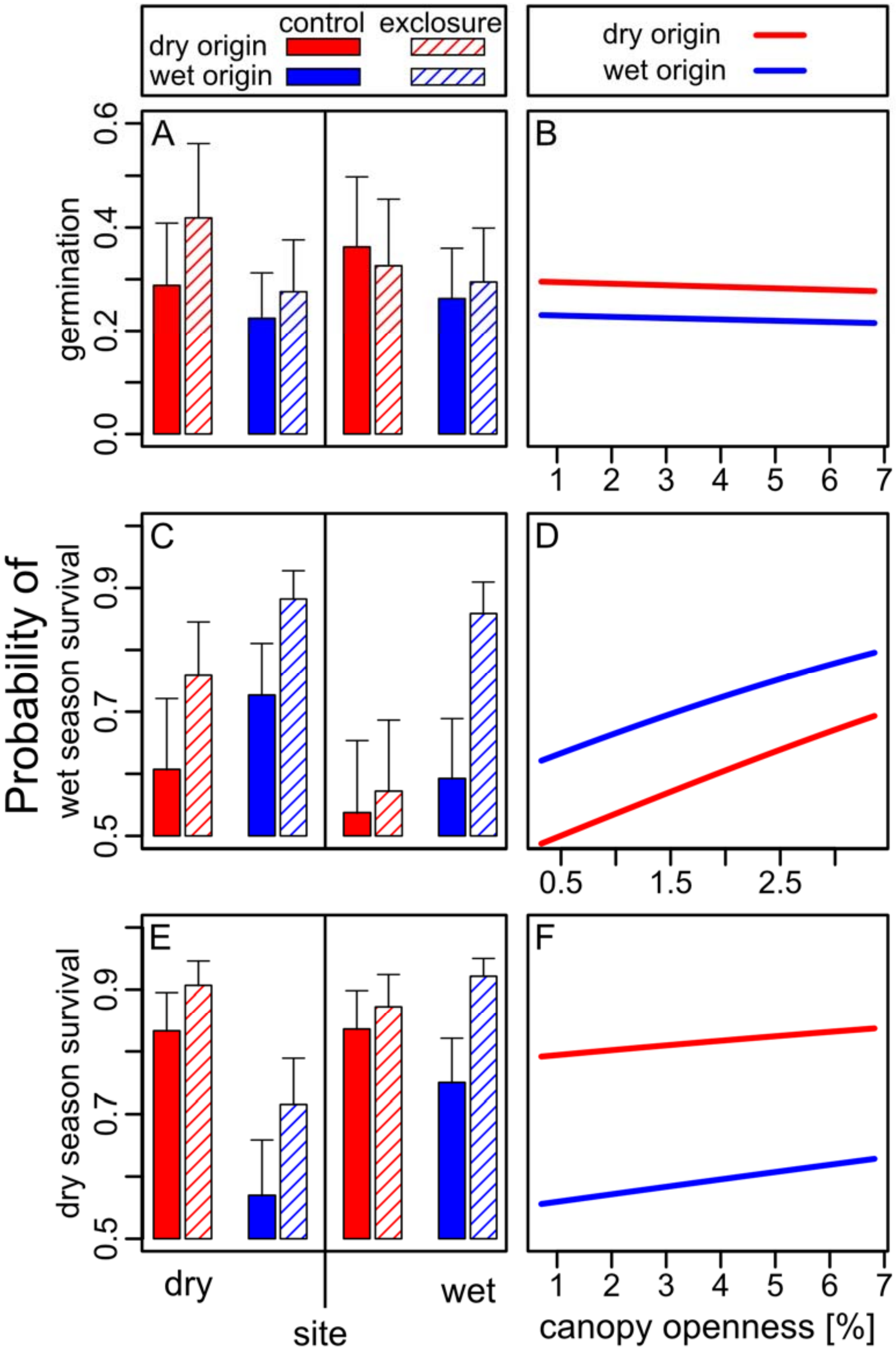

Figure 4: Probability of seed germination (A and B), wet season seedling survival (C and D) and dry season survival (E and F) for species with dry or wet origin as affected by moisture (dry vs. wet site), herbivore exposure (control vs. exclosure) and light availability (canopy openness). Panels A, C and E show means and standard errors from the least squares means table (Lenth 2014). For canopy openness, results of exclosure and control seeds and seedlings were pooled (see also Figure 3). For overall analyses see Table 1, for planned contrasts (post-hoc-tests) see Table 2.

Seedling establishment

At the end of the experiment after one year, 22.2% of the seedlings out of all seeds that were sown survived (799 out of 3600). We expected species to have a disadvantage outside their home range exhibiting lower overall establishment success, with dry origin species being limited by pest pressure and/or low light availability in the wet site, and wet origin species being limited by drought in the dry site. Contrary to our expectation, for control seedlings there were no significant differences in the establishment success of the two origins within sites (Table 2 a, Figure 3 a), nor across sites (Table 2 b, Figure 3 a). Thus, under natural conditions, dry origin and wet origin species did not differ in their probability to establish in both sites. Pest exclosure significantly enhanced seedling establishment, and the strength of the effect depended on site and origin (significant treatment x site x origin interaction, Table 1, Figure 3 a): the pest exclosure enhanced establishment success of wet origin species on both sites (Table 2 c and d), but of dry origin species only in the dry site (Table 2 c). Thus, contrary to our expectations, pest exclosure had no positive effect on dry origin species in the wet site (Table 2 d). Light had no overall significant effect on the establishment success over the whole experimental period. There was a marginally significant origin x light interaction; however, opposite to our expectations, wet origin species profited more from higher light availability than dry origin species (Table 1, Figure 3 b). None of the other performance parameters exhibited a significant light x origin interaction (Table 1).

Growth

On average, after one year species had grown to a height of 14.1 cm (3.5 – 62.5 cm). We found a significant site x origin interaction (Table 1): Dry origin species grew significantly less than wet origin species in the wet site (Figure 3 c). Even in the dry site dry origin species grew less than wet origin species, although the difference was less pronounced than in the wet site (Table 2 a, Figure 3 c). Dry origin species grew less in the wet site than in their dry home range, while wet origin species showed no difference in growth performance across sites (Table 2 b, Figure 3 c). Independent of species origin, overall growth was lower in the wet site compared to the dry site (Table 1). Dry origin species had an overall lower growth than wet origin species independent of site (Table 1). Pest exclosure did not affect growth, neither alone nor in interaction with origin nor site, indicating that pests did not limit growth in our experiment (Table 1, Table 2 c and d). Growth was also not affected by the variation of light availability encountered in this study (Table 1, Figure 3 d).

Germination

Out of the seeds sown in the experiment, 38% germinated (1384 of 3600). Germination ranged from 0% to 94% among species (Figure S1). Within sites, no differences were found between origins under natural conditions (Table 2 a, Figure 4 a). Contrary to our expectations, dry origin species showed a marginally significant trend ($p = 0.06$) to germinate better in the wet than in the dry site under natural control conditions, while wet origin species did not show any differences between sites (Table 2 b). As expected, pest exclosure benefited dry origin species more than wet origin species, but opposite to our expectations, only in their home range (Table 2 c and d, Figure 4 a). Wet origin species showed no higher germination rates when pests were excluded, neither in the dry nor in the wet site (Table 2 c and d). Light availability did not affect germination, neither for dry nor for wet origin species (Table 1, Figure 4 b).

Seedling survival during the wet season

In the wet season, 72% of the germinated seeds survived (994 of 1384). Although the interaction site x origin x treatment was marginally significant (Table 1), trends did not conform to our expectations (Figure 4 c). Wet origin species did not show higher survival rates than the dry origin species in any of the sites (Table 2 a, Figure 4 c), although we had expected them to show lower mortality during the wet season than dry origin species. Also opposite to our expectations, there was a trend for wet origin species to perform better in the dry site, while dry origin species showed no difference between sites (Table 2 b). Pest exclosure benefited wet origin species both in the dry and the wet site, and dry origin species only in the dry site (Table 2 c and d). Wet season survival of the seedlings increased with increasing light availability, but there were no differences between the two origins (Table 1, Figure 4 d).

Seedling survival during the dry season

The subsequent dry season was survived by 80% of the seedlings (799 of 994). There was a significant site x origin interaction in the dry season, with seedling survival in both sites dependent on their origin (Table 1, Figure 4 e). Under natural control conditions in the dry site, wet origin species had a significantly lower survival than dry origin species (Table 2 a). Wet origin species also had a lower survival in the dry than in the wet site (Table 2 b). Survival in the exclosure followed the same pattern (Figure 4 e), indicating that drought rather than pests led to the lower survival of wet origin species in the dry site. In contrast, dry origin species under control conditions showed no differences in survival between sites (Table 2 b). Pest exclosure increased survival of wet origin species both in the dry and the wet site (Table 2 c and d). For dry origin species, the exclosure effect was only marginally significant in the dry site (Table 2 c). During the dry season, the triple interaction site x origin x treatment was not significant (Table 1). Light availability had no significant effects in the dry season (Table 1, Figure 4 f), although the difference in light availability between the sites was highest during the dry season (Figure 2 b), probably due to leaf shedding.

Discussion

In contrast to our expectations, the overall establishment success (i.e. germination and one-year survival) did not reflect the distribution patterns of the species (Figure 3 a). Under natural habitat conditions (i.e. exposed to pest pressure) seedlings had no home advantage in their respective home site, nor did their establishment success vary across sites (Figure 3 a and Table 2 a and b). Consistent with lower drought tolerance of wet origin species, dry season seedling survival in the dry site was significantly lower for wet than for dry origin species (Figure 4 e, Table 2 a). Although this did not result in an overall home advantage of dry forest species within the time-frame of our study (Figure 3), it may lead to the exclusion of wet forest species from dry forests in more intense dry seasons and over longer time frames.

Neither the pest pressure hypothesis, nor the light availability hypothesis were supported to be important in early life stages for excluding dry origin species from wet forests (Figure 3). However, growth patterns were consistent with a home advantage of wet origin species in the wet site (Figure 3 c): Wet origin species grew significantly faster than dry origin species (Table 2 a), and this effect was much more pronounced in the wet than in the dry site (significant site x origin interaction, Table 1). This home advantage of wet forest species was already visible one year after germination. It may accumulate over time, and lead to the eventual exclusion of dry origin species from wet sites. Below we discuss our results and their implications for factors and life stages shaping tree distributions across rainfall gradients in more detail.

Exclusion of wet forest species from dry sites

Our results indicate that our focal wet forest species were less drought-tolerant than the dry forest species, and that drought limited their survival in the dry site during the dry season, as we had expected: Their dry season survival in the dry forest was much lower compared to dry forest species (Figure 4 e, Table 2 a), and compared to the wet forest site (Figure 4 e, Table 2 b). We can rule out that these effects were due to pest pressure, because seedlings in controls and exclosures followed the same pattern (Table 2 c and d, Figure 4 e), or that they were due to light, because light availability had no effect on seedling survival (Table 1, Figure 4 f). Therefore, drought was directly responsible for reducing survival in the dry site. These results confirm previous studies in tropical forests worldwide which show that seedlings of wet forest species (or occurring in wet sites) are less drought-tolerant, i.e. more susceptible to drought, than seedlings of species occurring in dry forests exposed to a strong dry season (Engelbrecht et al. 2007, Baltzer et al. 2008, Comita and Engelbrecht 2013, Amissah et al. 2014, Sterck et al. 2014).

Despite their lower drought tolerance, after one year wet forest species in the dry site did not perform poorer than dry forest species neither in terms of overall establishment nor growth (Figure 3 a and c, Table 2 a and b). Slightly, but non-significantly higher wet season survival in wet compared to dry origin species in the dry site (Figure 4 c, Table 2 a), may have counterbalanced their lower survival during the dry season, resulting in no overall difference in establishment success during our study period (Table 2 a).

The strength of the dry season (i.e. the duration and the water deficit reached) varies considerably across years, and consequently, dry season seedling performance also varies (Comita and Engelbrecht 2013). Pronounced seedling mortality, especially of drought-sensitive species, occurs predominantly in particularly dry years (Comita and Engelbrecht 2013), while the dry season in our study period was well within the long-term average (ESP 2015). Rather than contradicting the physiological tolerance hypothesis, our results, together with the previous studies, thus underline the importance of strong and repeated dry seasons, such as those occurring during El Niño Southern Oscillation (ENSO) events, for exclusion of wet origin species from dry forests.

Exclusion of dry forest species from wet sites

While the mechanisms underlying distribution limits of wet forest species to dry sites are quite well understood, the mechanisms excluding dry origin species from wet forests are not yet resolved. Contrary to our expectations, we found no indication of either high pest damage or low shade tolerance limiting the performance of dry origin species in the wet site.

Pest pressure hypothesis

Consistent with our hypotheses, pest exclosure through insecticide and fungicide treatment had a significant positive overall effect on establishment success, germination and survival (Table 1), indicating that these processes were limited by pests. Growth was not affected by pests in congruence with results from Eichhorn et al. (Eichhorn et al. 2010), who argued that levels of herbivory were exceedingly high in the few studies which found negative effects of herbivore damage on growth.

However, contrary to our expectations, the performance of early life stages of dry origin species in the wet site was not limited by herbivores or pathogens, as shown by the lack of a positive effect of alleviating potential damage through pest exclosure (Table 2 d). Furthermore, we did not find any indication of overall higher pest pressure in the wet site, which would have manifested itself in a significant site x treatment interaction with a higher treatment effect in the wet site (see Table 1). On the contrary, the effect of pest pressure was higher in the dry site for establishment success and germination, (see Table 1 and below). Thus, our results did not support the pest pressure hypothesis.

To our knowledge, so far only three studies have explicitly tested the pest pressure hypothesis. All three used transplant experiments with species of contrasting origins across tropical rainfall gradients (Brenes-Arguedas et al. 2009, Baltzer and Davies 2012, Spear et al. 2015). Two studies in Panama (Brenes-Arguedas et al. 2009, Spear et al. 2015) found higher overall damage and higher pathogen damage in a wet aseasonal than in a dry seasonal forest, consistent with higher pest pressure but contrary to our results, while a study at the Malay-Thai peninsula found no evidence for higher pest pressure in an aseasonal compared to a seasonal forest (Baltzer and Davies 2012). None of these studies found significantly higher damage in dry than wet origin species indicative of lower defenses in the dry forest species, as required for the pest pressure hypothesis. Instead, the results of Spear et al. (Spear et al. 2015) suggest that the susceptibility of species, i.e. their likelihood to die after pathogen or herbivore damage, varies, with wet origin species being less susceptible. They proposed that higher susceptibility rather than lower defenses may limit the distribution of dry origin species in wet sites. However, if these processes are important for species distribution, an overall performance outcome consistent with the pest pressure hypothesis (i.e. stronger negative effects of pest on dry species performance) would still be expected, regardless if it is driven by defenses or susceptibility. Our results do not support the importance of differences in defenses nor susceptibility for germination or early seedling performance.

We expected higher pest limitation of dry forest species in wet forests, due to the combined effects of higher pest pressure in wetter forests and lower defenses of dry origin species (Figure 1). Instead, the positive effect of pest exclosure for dry origin species was consistently higher in the dry site compared to the wet site for all performance parameters (Figure 3, Figure 4, Table 2 c and d), and for wet origin species the effect of pest exclosure was equally high in both sites (Figure 3, Figure 4, Table 2 c and d). These results might hint towards a higher degree of specialization of the herbivore community in the wet than in the dry forest: If transplanting dry origin species to the wet forest introduced them to a specialized herbivore community with which they did not co-evolve, lower pest limitation compared to native wet forest species, as we observed, would be expected (compare enemy release hypothesis). On the other hand, if the herbivore community in the dry forest is more generalistic, wet origin species would be expected not to show higher release from pest pressure outside their home range, again consistent with our observation. If specialization of pests indeed increases across rainfall gradients, it would put the pest pressure gradient hypothesis into question, since dry forest species may escape their enemies and have an advantage in wet forests. While overall, specialization of insect herbivores and fungal pathogens is not as strong as originally thought (Novotny and Basset 2005, Gilbert and Webb 2007), we are not aware of any study comparing the degree of specialization of herbivore communities across rainfall gradients. Targeted studies analyzing specialization across rainfall gradients will be needed to evaluate this possibility.

In summary, our results do not support any of the patterns expected from the pest pressure hypotheses for early life stages, and - taken together with previous studies - decisive support for the pest pressure gradient hypothesis remains elusive.

Light availability hypothesis

Light responses in our experiment did not significantly differ between wet origin and dry origin species (no significant origin x light interaction, Table 1). We found no indication that dry origin species were more light-demanding than wet origin species, as expected from the light availability hypothesis. On the contrary, wet origin species even showed a trend to higher light requirements, indicated by the marginally significant trend to higher establishment success with increasing light than dry origin species (origin x light interaction, Table 1, Figure 3 b). Previous studies similarly did not find support for higher light requirements in dry than wet forest species (Engelbrecht et al. 2007, Markesteijn and Poorter 2009, Brenes-Arguedas et al. 2011). Additionally, although light availability was significantly higher in the dry than in the wet site (Figure 2), differences were small (see also Brenes-Arguedas et al. 2011). Overall canopy openness showed only little variation with values between 1 and 7%. These values are typical within the understory of tropical forests (Harms et al. 2004, Brenes-Arguedas et al. 2011). The small variation may contribute to the overall small effect of light on species performance observed in this study. Our results, together with previous studies, suggest that light does not play a significant role in shaping species distributions across tropical rainfall gradients.

Growth and the role of a drought tolerance-growth trade-off

Wet forest species had a home advantage in terms of growth: in the wet forest growth rates of wet origin species were higher than of dry origin species (Table 2 a). Through this growth difference, wet origin species may over time outperform and exclude dry origin species from wet forests. Previous studies in the area have also found lower growth rates in dry compared to wet forest species in independent species sets (only three species overlapping, Brenes-Arguedas et al. 2008, 2009). Similar patterns were also found in studies in the Malay-Thai peninsula, where widespread, dry distribution species had lower growth rates than aseasonal, wet distribution species (Baltzer et al. 2007, Baltzer and Davies 2012). This suggests that lower seedling growth rates in dry than wet forest species are a general and widespread pattern.

We have discarded above that the lower growth rates of dry forest species were due to pest damage or light requirements. An alternative factor that may lead to this pattern is low nutrient availability in wet forests, and indeed high nutrient requirement of dry forest species have been suggested to exclude them from nutrient poor wet forests (Huston 1994, ter Steege et al. 2003, Brenes-Arguedas et al. 2008). If dry origin species have higher nutrient requirements and wetter forests have lower nutrients, this could explain the reduced growth of dry origin species with increasing rainfall found in our study (Table 2 b). However, dry and wet origin species do not differ in nutrient requirements in Central Panama (Brenes-Arguedas et al. 2008), and relations between rainfall and nutrient availability are weak (Condit et al. 2013). Differential nutrient requirements can therefore be ruled out as a cause for overall lower growth rates of dry forest species and for playing a major role in excluding dry origin species from wet forests, although they do influence distribution across nutrient gradients (Condit et al. 2013).

Instead, lower growth rates in dry forest species are consistent with a stress tolerance-growth trade-off, which has been hypothesized based on costs associated with adaptations to low resource availability which should lead to inherently lower growth rates, even under optimum conditions, in stress-tolerant species (Grime 1977, Smith and Huston 1989). There is ample evidence for a stress tolerance-growth trade-off based on shade (e.g. Wright et al. 2010). Also, several traits promoting tolerance to drought are traded-off against growth rates (Poorter et al. 2010, O'Brien et al. 2015). Nevertheless, although often implied, direct empirical evidence for a whole-plant drought tolerance-growth trade-off remains surprisingly scarce. Support for a trade-off between drought survival and maximum growth rates or shoot growth rate across species was found e.g. by O'Brien et al. (O'Brien et al. 2015), Polley et al. (Polley et al. 2002) and Wikberg et al. (Wikberg and Ögren 2004), in tropical tree seedlings, tropical and subtropical woody legumes, and in willows, respectively. Consistently, in our study there was a marginally significant negative relation between dry season survival on the dry site and maximum growth rates (assessed as the upper 95 percentile of growth on the wet site, GLMER: $p = 0.07$, based on data for the 16 species with more than 3 survivors). However, the only rigorous experimental study that explicitly tested for this trade-off, which was conducted in eight desert grasses, did not support it (Fernandez and Reynolds 2000).

Species with dry distribution have been experimentally shown to be more tolerant to drought stress than species with wet distribution (Baltzer et al. 2007, 2008, Engelbrecht et al. 2007), and higher drought-tolerance in dry origin species is consistent with the data from our study (see above). Inherently lower growth rates of dry compared to wet origin species found in this (Table 2 a) and other studies (Baltzer et al. 2007, Brenes-Arguedas et al. 2008, 2009) thus provide additional indirect support for a stress tolerance-growth trade-off with respect to drought. This trade-off may underlie the exclusion of dry forest species from wet sites and be fundamental in shaping species distributions along rainfall gradients.

The role of early life stages for species distributions

In this study, we focused on the initial life stages of germination and early seedling establishment, since these stages are considered the most vulnerable in the face of biotic and abiotic stressors (Harper 1977, Daws et al. 2005) and may thus be critical in shaping species distribution patterns across tropical rainfall gradients. However, germination (i.e. radicle emergence) patterns did not reflect the occurrence patterns of the species (Figure 4 a, Table 2 a and b), indicating that species partitioning along the rainfall gradient did not occur at this stage. We found support that differential dry season survival (in dry sites) and differential growth (in wet sites) during early life stages contribute to shaping tree distribution patterns across tropical rainfall gradients (see above). However, effects were weak and not sufficient to lead within the initial year to a clear home advantage of the species on the dry or wet side, respectively. Across a topographic moisture gradient, processes within one year after emergence were also insufficient to explain habitat preferences of adult plants (Daws et al. 2005). This strongly suggests that longer time spans reaching into later life stages, and repeated and pronounced dry seasons are important for filtering tree distribution patterns.

The importance of later life stages and longer time periods for shaping distribution patterns is supported by local scale studies: If habitat associations of adults are shaped by failure to germinate or to establish, older juveniles and adults should exhibit the same habitat associations. However, most species have different associations at seedling and late life stages (Comita et al. 2007).

Conclusions

We found two processes that may lead to the differential distribution patterns of dry and wet origin species after longer time periods and at later life stages. We showed that drought limits the survival of wet origin species in dry forests, which supports the physiological tolerance hypothesis. Dry origin species had lower growth rates than wet origin species, especially in the wet forest site, consistent with a drought tolerance-growth trade-off. Our results support that repeated and intense dry season drought limits performance and consequent distribution of wet origin species in dry forests, and suggest that dry origin species are outperformed in wet forests due to inherently lower growth rates, based on a drought tolerance-growth trade-off.

Although pest pressure had a strong overall influence on species establishment success, we found no support for the hypothesis that high pest pressure excludes dry origin species from wet forests (pest pressure gradient hypothesis). We also found no evidence for the hypothesis that dry origin species have higher light requirements than wet origin species, and are thus excluded from wetter forests with darker understory (light gradient hypothesis).

Our results underline that changes in water availability due to climate change will have direct consequences on species regeneration and distributions along rainfall gradients, while indirect effects of pest pressure and light availability play a subordinate role.

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References

Amissah, L., G. M. Mohren, F. Bongers, W. D. Hawthorne, and L. Poorter. 2014. Rainfall and temperature affect tree species distribution in Ghana. Journal of Tropical Ecology 30:435–446.

Amissah, L., G. M. J. Mohren, B. Kyereh, and L. Poorter. 2015. The effects of drought and shade on the performance, morphology and physiology of Ghanaian tree species. PloS One 10:e0121004.

Bagchi, R., R. E. Gallery, S. Gripenberg, S. J. Gurr, L. Narayan, C. E. Addis, R. P. Freckleton, and O. T. Lewis. 2014. Pathogens and insect herbivores drive rainforest plant diversity and composition. Nature 506:85–88.

Baltzer, J., S. Davies, S. Bunyavejchewin, and N. Noor. 2008. The role of desiccation tolerance in determining tree species distributions along the Malay-Thai Peninsula. Functional Ecology 22:221–231.

Baltzer, J. L., and S. J. Davies. 2012. Rainfall seasonality and pest pressure as determinants of tropical tree species' distributions. Ecology and Evolution 2:2682–2694.

Baltzer, J. L., S. J. Davies, N. S. M. Noor, A. R. Kassim, and J. V. LaFrankie. 2007. Geographical distributions in tropical trees: can geographical range predict performance and habitat association in co-occurring tree species? Journal of Biogeography 34:1916–1926.

Barton, K. E., and J. Koricheva. 2010. The ontogeny of plant defense and herbivory: characterizing general patterns using meta-analysis. The American Naturalist 175:481–493.

Bates, D., M. Maechler, B. Bolker, and S. Walker. 2013. lme4: Linear mixed-effects models using Eigen and S4. R package version 1.0-5.

Bongers, F., M. P. E. Parren, and D. Traore. 2005. Forest Climbing Plants of West Africa - Diversity, Ecology and Management. CABI, London.

Bongers, F., L. Poorter, R. Rompaey, and M. Parren. 1999. Distribution of twelve moist forest canopy tree species in Liberia and Cote d'Ivoire: response curves to a climatic gradient. Journal of Vegetation Science 10:371–382.

Brenes-Arguedas, T., P. D. Coley, and T. A. Kursar. 2009. Pests vs. drought as determinants of plant distribution along a tropical rainfall gradient. Ecology 90:1751–1761.

Brenes-Arguedas, T., M. Rios, G. Rivas-Torres, C. Blundo, P. D. Coley, and T. A. Kursar. 2008. The effect of soil on the growth performance of tropical species with contrasting distributions. Oikos 117:1453–1460.

Brenes-Arguedas, T., A. B. Roddy, P. D. Coley, and T. A. Kursar. 2011. Do differences in understory light contribute to species distributions along a tropical rainfall gradient? Oecologia 166:443–456.

Coley, P. D., and J. A. Barone. 1996. Herbivory and plant defenses in tropical forests. Annual Review of Ecology and Systematics 27:305–335.

Comita, L. S., R. Condit, and S. P. Hubbell. 2007. Developmental changes in habitat associations of tropical trees. Journal of Ecology 95:482–492.

Comita, L. S., and B. M. Engelbrecht. 2013. Drought as a driver of tropical tree species regeneration dynamics and distribution patterns. Pages 261–308 *in* Coomes, David A and Burslem, David FRP and Simonson, William D, editor. Forests and Global Change. Cambridge University Press, Cambridge, UK.

Condit, R., B. M. Engelbrecht, D. Pino, R. Pérez, and B. L. Turner. 2013. Species distributions in response to individual soil nutrients and seasonal drought across a community of tropical trees. Proceedings of the National Academy of Sciences of the United States of America 110:5064–5068.

Condit, R., N. Pitman, E. G. Leigh, J. Chave, J. Terborgh, R. B. Foster, P. Nunez, S. Aguilar, R. Valencia, G. Villa, H. C. Muller-Landau, E. Losos, and S. P. Hubbell. 2002. Beta-diversity in tropical forest trees. Science 295:666–669.

Coomes, D. A., and P. J. Grubb. 2000. Impacts of root competition in forests and woodlands: a theoretical framework and review of experiments. Ecological Monographs 70:171–207.

CTFS. 2015. Center for Tropical Forest Science, CTFS. Available at: http://ctfs.arnarb.harvard.edu/webatlas/

Currie, D. J., G. G. Mittelbach, H. V. Cornell, R. Field, J.-F. Guégan, B. A. Hawkins, D. M. Kaufman, J. T. Kerr, T. Oberdorff, E. O'Brien, and J. Turner. 2004. Predictions and tests of climate-based hypotheses of broad-scale variation in taxonomic richness. Ecology Letters 7:1121–1134.

Daws, M. I., T. R. Pearson, D. F. P. Burslem, C. E. Mullins, and J. W. Dalling. 2005. Effects of topographic position, leaf litter and seed size on seedling demography in a semi-deciduous tropical forest in Panama. Plant Ecology 179:93–105.

Eichhorn, M. P., R. Nilus, S. G. Compton, S. E. Hartley, and D. F. Burslem. 2010. Herbivory of tropical rain forest tree seedlings correlates with future mortality. Ecology 91:1092–1101.

Engelbrecht, B. M. J., L. S. Comita, R. Condit, T. A. Kursar, M. T. Tyree, B. L. Turner, and S. P. Hubbell. 2007. Drought sensitivity shapes species distribution patterns in tropical forests. Nature 447:80–83.

ESP. 2015. Data sets provided by the Terrestrial-Environmental Sciences Program of the Smithsonian Tropical Research Institute. Available at: http://stri.si.edu/sites/esp/

Fayolle, A., M. D. Swaine, J.-F. Bastin, N. Bourland, J. A. Comiskey, G. Dauby, J.-L. Doucet, J.-F. Gillet, S. Gourlet-Fleury, O. J. Hardy, B. Kirunda, F. N. Kouamé, and A. J. Plumptre. 2014. Patterns of tree species composition across tropical African forests. Journal of Biogeography 41:2320–2331.

Fernandez, R. J., and J. F. Reynolds. 2000. Potential growth and drought tolerance of eight desert grasses: lack of a trade-off? Oecologia 123:90–98.

Gilbert, G. S., and C. O. Webb. 2007. Phylogenetic signal in plant pathogen-host range. Proceedings of the National Academy of Sciences of the United States of America 104:4979–4983.

Givnish, T. J. 1999. On the causes of gradients in tropical tree diversity. Journal of Ecology 87:193–210.

Grime, J. 1977. Evidence for the existence of three primary strategies in plants and its relevance to ecological and evolutionary theory. The American Naturalist 111:1169–1194.

Gripenberg, S., R. Bagchi, R. E. Gallery, R. P. Freckleton, L. Narayan, and O. T. Lewis. 2014. Testing for enemy-mediated density-dependence in the mortality of seedlings: field experiments with five Neotropical tree species. Oikos 123:185–193.

Harms, K. E., J. S. Powers, and R. A. Montgomery. 2004. Variation in small sapling density, understory cover, and resource availability in four Neotropical forests. Biotropica 36:40–51.

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

Hijmans, R. J., S. E. Cameron, J. L. Parra, P. G. Jones, and A. Jarvis. 2005. Very high resolution interpolated climate surfaces for global land areas. International Journal of Climatology 25:1965–1978.

Holdridge, L. R., and G. Budowski. 1959. Mapa ecológico de Panamá (1:1,000,000). Interamerican Institute of Agricultural Science, Turrialba.

Huston, M. A. 1994. Biological Diversity: The Coexistence of Species. Cambridge University Press, Cambridge, UK.

Lenth, R. V. 2014. lsmeans: Least-Squares Means. R package version 2.00-5.

Malhi, Y., L. E. O. C. Aragão, D. Galbraith, C. Huntingford, R. Fisher, P. Zelazowski, S. Sitch, C. McSweeney, and P. Meir. 2009. Exploring the likelihood and mechanism of a climate-change-induced dieback of the Amazon rainforest.

Proceedings of the National Academy of Sciences of the United States of America 106:20610–20615.

Markesteijn, L., and L. Poorter. 2009. Seedling root morphology and biomass allocation of 62 tropical tree species in relation to drought-and shade-tolerance. Journal of Ecology 97:311–325.

Markesteijn, L., L. Poorter, F. Bongers, H. Paz, and L. Sack. 2011. Hydraulics and life history of tropical dry forest tree species: coordination of species' drought and shade tolerance. New Phytologist 191:480–495.

Novotny, V., and Y. Basset. 2005. Host specificity of insect herbivores in tropical forests. Proceedings of the Royal Society B: Biological Sciences 272:1083–1090.

O'Brien, M. J., D. F. R. P. Burslem, A. Caduff, J. Tay, and A. Hector. 2015. Contrasting nonstructural carbohydrate dynamics of tropical tree seedlings under water deficit and variability. New Phytologist 205:1083–1094.

Polley, H. W., C. R. Tischler, H. B. Johnson, and J. D. Derner. 2002. Growth rate and survivorship of drought: CO2 effects on the presumed tradeoff in seedlings of five woody legumes. Tree Physiology 22:383–391.

Poorter, L., I. McDonald, A. Alarcón, E. Fichtler, J.-C. Licona, M. Peña-Claros, F. Sterck, Z. Villegas, and U. Sass-Klaassen. 2010. The importance of wood traits and hydraulic conductance for the performance and life history strategies of 42 rainforest tree species. New Phytologist 185:481–492.

Pyke, C. R., R. Condit, S. Aguilar, and S. Lao. 2001. Floristic composition across a climatic gradient in a neotropical lowland forest. Journal of Vegetation Science 12:553–566.

R Core Team. 2013. R: a language and environment for statistical computing.

Smith, T., and M. Huston. 1989. A theory of the spatial and temporal dynamics of plant communities. Vegetatio 83:49–69.

Spear, E. R., P. D. Coley, and T. A. Kursar. 2015. Do pathogens limit the distributions of tropical trees across a rainfall gradient? Journal of Ecology 103:165–174.

ter Steege, H., N. Pitman, D. Sabatier, H. Castellanos, P. Van der Hout, D. C. Daly, M. Silveira, O. Phillips, R. Vasquez, T. Van Andel, J. Duivenvoorden, A. A. De Oliveira, R. Ek, R. Lilwah, R. Thomas, J. Van Essen, C. Baider, P. Maas, S. Mori, J. Terborgh, P. N. Vargas, H. Mogollon, and W. Morawetz. 2003. A spatial model of tree alpha-diversity and tree density for the Amazon. Biodiversity and Conservation 12:2255–2277.

Sterck, F. J., L. Markesteijn, M. Toledo, F. Schieving, and L. Poorter. 2014. Sapling performance along resource gradients drives tree species distributions within and across tropical forests. Ecology 95:2514–2525.

Toledo, M., M. Peña-Claros, F. Bongers, A. Alarcon, J. Balcazar, J. Chuvina, C. Leaño, J. C. Licona, and L. Poorter. 2012. Distribution patterns of tropical woody species in response to climatic and edaphic gradients. Journal of Ecology 100:253–263.

Toledo, M., L. Poorter, M. Peña-Claros, A. Alarcón, J. Balcázar, C. Leaño, J. C. Licona, O. Llanque, V. Vroomans, P. Zuidema, and others. 2011. Climate is a stronger driver of tree and forest growth rates than soil and disturbance. Journal of Ecology 99:254–264.

Tremblay, A., D. University, J. Ransijn, and U. of Copenhagen. 2013. LMERConvenienceFunctions: A suite of functions to back-fit fixed effects and forward-fit random effects, as well as other miscellaneous functions. R package version 2.5.

Welden, C. W., S. W. Hewett, S. P. Hubbell, and R. B. Foster. 1991. Sapling survival, growth, and recruitment: relationship to canopy height in a neotropical forest. Ecology 72:35–50.

Wikberg, J., and E. Ögren. 2004. Interrelationships between water use and growth traits in biomass-producing willows. Trees 18:70–76.

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

Supporting information

Effects of drought, pest pressure and light availability on seedling establishment and growth: Their role for distribution of tree species across a tropical rainfall gradient

Julian Gaviria and Bettina M. J. Engelbrecht

Figure S1: Performance parameters of the 26 focal species analyzed.

Table S1: Focal species, including their classification into dry or wet origin.

Table S2: Correlations of site and abiotic factors.

Table S3: Detailed summary table of the models (GLMM and LMM).

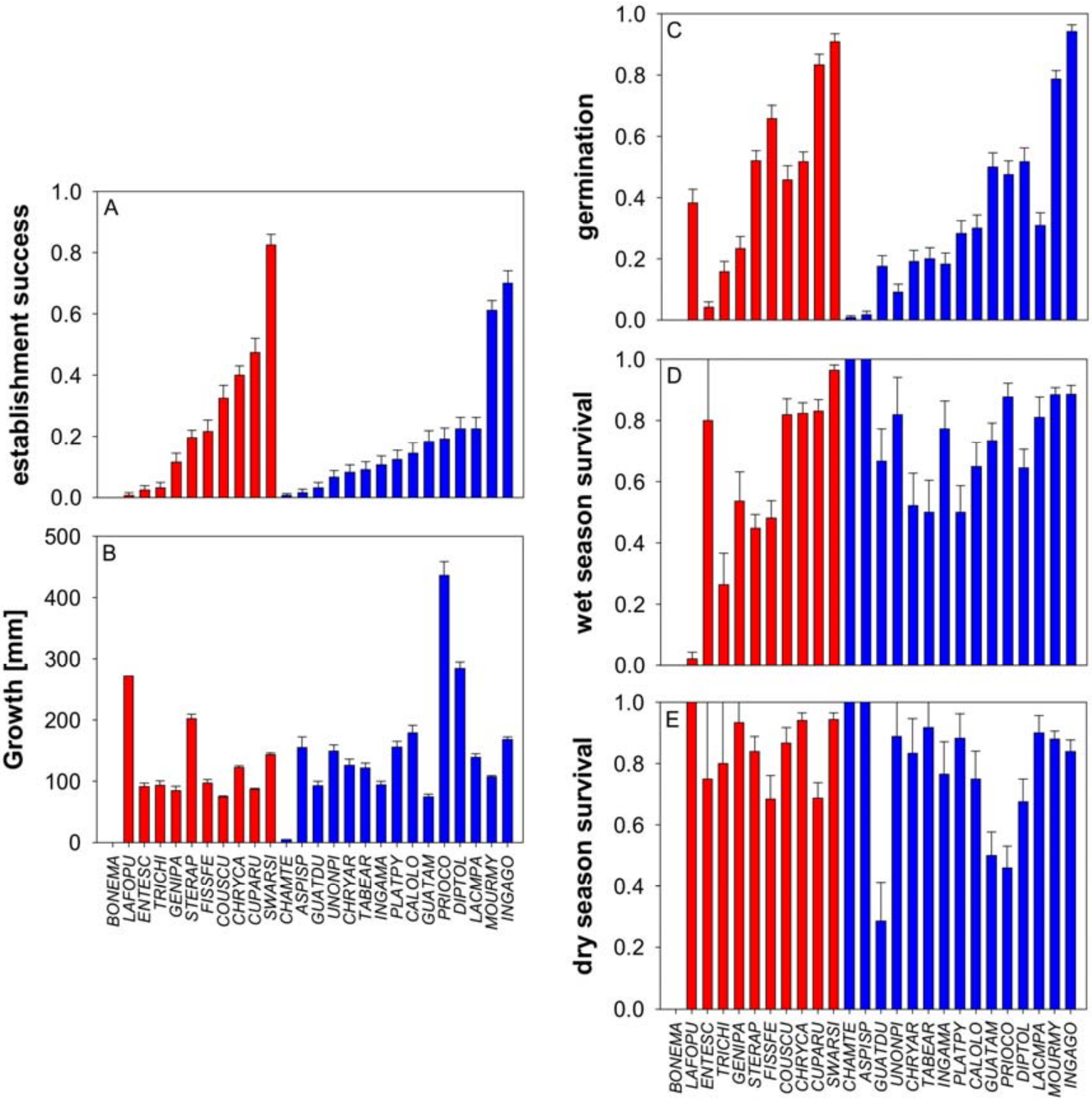

Figure S1: Performance parameters of the 26 focal species analyzed. Probability of establishment success (A), growth (B), probability of germination (C), probability of wet season survival (D) and probability of dry season survival (E), sorted by species' origin (dry: red, wet: blue) and average establishment success. Data are averages and standard errors. Species effects on all performance parameters were highly significant (GLMM for probability of establishment, germination, total survival, survival wet and dry season and LMM for growth: $p < 0.001$). For full species names see Table S1.

Table S1: Focal species, including their classification into dry or wet origin.

Species	Abbreviation	Family (-ceae)	Origin	Criteria for classification
Aspidosperma spruceanum Benth. ex Müll. Arg.	ASPISP	Apocyna	wet	1
Bonellia macrocarpa (Cav.) B. Ståhl & Källersjö	BONEMA	Primula	dry	3
Calophyllum longifolium Willd.	CALOLO	Calophylla	wet	1
Chamaedorea tepejilote Liebm.	CHAMTE	Areca	wet	1
Chrysophyllum argenteum Jacq.	CHRYAR	Sapota	wet	1,2
Chrysophyllum cainito L.	CHRYCA	Sapota	dry	1,2
Coussarea curvigemmia Dwyer	COUSCU	Rubia	dry	1
Cupania rufescens Triana & Planch.	CUPARU	Sapinda	dry	1
Dipteryx oleifera Benth.	DIPTOL	Faba	wet	1
Enterolobium schomburgkii (Benth.) Benth.	ENTESC	Faba	dry	1
Fissicalyx fendleri Benth.	FISSFE	Faba	dry	1
Genipa americana L.	GENIAM	Rubia	dry	1,2
Guatteria amplifolia Triana & Planch.	GUATAM	Annona	wet	1
Guatteria dumetorum R.E. Fr.	GUATDU	Annona	wet	1
Inga goldmanii Pittier	INGAGO	Faba	wet	1,2
Inga marginata Willd.	INGAMA	Faba	wet	1
Lacmellea panamensis (Woodson) Markgr.	LACMPA	Apocyna	wet	1
Lafoensia punicifolia DC.	LAFOPU	Lythra	dry	1
Mouriri myrtilloides (Sw.) Poir.	MOURMY	Melastomata	wet	1
Platymiscium pinnatum (Jacq.) Dugand	PLATPI	Faba	wet	1
Prioria copaifera Griseb.	PRIOCO	Faba	wet	1
Sterculia apetala (Jacq.) H. Karst.	STERAP	Malva	dry	1
Swartzia simplex (Sw.) Spreng.	SWARSI	Faba	dry	1
Tabernaemontana arborea Rose	TABEAR	Apocyna	wet	1
Trichilia hirta L.	TRICHI	Melia	dry	1
Unonopsis pittieri Saff.	UNONPI	Annona	wet	3

Criteria used:
1: occurrence maps according to CTFS, available at http://ctfs.arnarb.harvard.edu/webatlas/maintreeatlas.php
2: abundance data according to CTFS, available at http://ctfs.arnarb.harvard.edu/webatlas/datasets/
3: description of the species according to CTFS

Table S2: Correlations of site and abiotic factors.

	Site	Light dry	Light wet	Light mean	Moisture dry 1	Moisture wet	Moisture dry 2
Light dry	-0.64						
Light wet	-0.36	0.47					
Light mean	-0.63	0.92	0.74				
Moisture dry 1	0.87	-0.66	-0.25	-0.60			
Moisture wet	0.82	-0.59	-0.14	-0.50	0.89		
Moisture dry 2	0.92	-0.65	-0.30	-0.58	0.79	0.78	
Moisture mean	0.85	-0.61	-0.16	-0.52	0.90	0.99	0.82

Light dry, wet and mean: % canopy openness during the dry season, the wet season, and the mean, respectively. Moisture dry 1, wet, dry 2 and mean: % gravimetric soil moisture during the transition between first dry season and wet season, during the wet season, during the second dry season, and mean soil moisture. Values are Spearman rank correlation factors (r).

Table S3: Summary table of the Models (GLMM and LMM).

		Establishment success	Growth	Germination	Survival wet season	Survival dry season
Intercept	Estimate	-2.187	4.851	-0.904	-0.141	1.457
	z (t) value	-3.224	168.4	-1.545	-0.241	3.687
	p value	**0.001**	**<0.001**	0.122	0.81	**<0.001**
Site	Estimate	0.183	-0.199	0.340	-0.286	0.053
	z (t) value	0.715	-4.716	1.890	-0.949	0.175
	p value	0.474	**<0.001**	*0.059*	0.343	0.861
Origin	Estimate	-1.110	0.096	-0.334	0.544	-1.112
	z (t) value	-1.283	2.31	-0.435	0.863	-2.291
	p value	0.199	**0.021**	0.664	0.388	**0.022**
Treatment	Estimate	0.841	0.039	0.575	0.711	0.751
	z (t) value	4.264	1.306	3.348	2.431	4.167
	p value	**<0.001**	0.192	**<0.001**	**0.015**	**<0.001**
Canopy openness	Estimate	-0.031	-0.031	-0.015	0.284	0.049
	z (t) value	-0.212	-1.213	-0.236	1.963	0.401
	p value	0.832	0.226	0.813	**0.049**	0.689
Site x origin	Estimate	0.078	0.166	-0.133	-0.319	0.863
	z (t) value	0.242	2.816	-0.561	-0.824	2.312
	p value	0.809	**0.005**	0.574	0.41	**0.021**
Site x treatment	Estimate	-0.880	-0.024	-0.737	-0.57	0.278
	z (t) value	-3.135	-0.399	-3.042	-1.451	0.764
	p value	**0.002**	0.69	**0.002**	0.147	0.445
Origin x treatment	Estimate	-0.199	0.063	-0.302	0.322	0.445
	z (t) value	-0.721	1.052	-1.271	0.75	1.208
	p value	0.471	0.293	0.204	0.453	0.227
Origin x canopy openness	Estimate	0.338	-0.018	0.158	0.278	0.324
	z (t) value	1.931	-0.343	1.477	1.137	1.491
	p value	*0.054*	0.732	0.140	0.256	0.136
Site x origin x treatment	Estimate	1.221	-0.052	0.623	0.968	1.095
	z (t) value	3.145	-0.436	1.863	1.689	1.483
	p value	**0.002**	0.663	*0.062*	*0.091*	0.138

Estimate (regression coefficient), z- value and p-value of the explanatory variables site, origin, treatment and canopy openness are given for the response variables establishment success, germination, overall survival, wet and dry season survival. For growth, t- value is given instead of z- value.

Bold p-values show significant effects at the 0.05 level, italic p-values close-significant trends at the 0.1 level.

Manuscript 2: Herbivore defenses of tree species increase across a tropical rainfall gradient

Herbivore defenses of tree species increase across a tropical rainfall gradient

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Abstract

One of the most prominent ecological patterns in tropical forests is the distinct distribution of tree species along rainfall gradients. Because pronounced changes of rainfall are projected with climate change, understanding the underlying processes is imperative. Herbivores have long been hypothesized to play a crucial role in shaping the patterns through excluding poorly defended dry forest species from wet forests with high pest pressure. However, empirical support for the hypothesis is still lacking. Here we show that herbivore defenses increase with tree species association with wetter forests, and independently also increase with shade tolerance across a pronounced rainfall gradient in Panama. These results support that high herbivore pressure acts as a filter excluding dry forest species from wet forests and provide the first empirical evidence that herbivores are important in shaping tree species distributions across lowland tropical rainfall gradients.

Keywords

Plant distribution, biotic interactions, herbivory, herbivore preference, moisture, dual-choice test

Introduction

Tropical forests are among the most diverse systems on earth. The most pervasive patterns in these forests are relations of tree species distributions (Engelbrecht et al. 2007), changes of forest community composition (Pyke et al. 2001), and increases of species richness (Givnish 1999, ter Steege et al. 2003) with rainfall. However, the mechanisms driving these patterns remain poorly understood. Pronounced changes of precipitation patterns in the tropics are projected with global change with pervasive consequences for tropical forests. Understanding the processes underlying the observed gradients in forest composition is crucial to improve projections of consequences of climate change for tropical forests, and to inform mitigation and adaptation strategies.

Various hypotheses have been brought forward about processes that underlie the observed changes of forest composition and diversity with rainfall, including direct effects of water availability on plant-water relations, as well as indirect effects of additional factors important for plant performance, such as insect herbivores and pathogens, nutrients and light (Givnish 1999).

Differential tree drought tolerance directly shapes species distribution patterns and community composition across tropical rainfall gradients (Engelbrecht et al. 2007, Baltzer et al. 2008). Species occurring in wetter forests are more drought-sensitive and thus are excluded from drier forests, contributing to the decrease of species richness with decreasing rainfall (Engelbrecht et al. 2007). However, while such direct drought effects play an important role, a considerable part of the variation of species distribution across rainfall gradients remains unexplained (Engelbrecht et al. 2007).

Herbivores have additionally long been hypothesized to play a significant role for the observed patterns. Herbivory is important in tropical forests with pervasive effects on plant performance, regeneration and fitness (Eichhorn et al. 2010). Herbivory also has been shown to influence tropical tree performance and distributions along edaphic gradients (Fine et al. 2004).

Across rainfall gradients, herbivore pressure has been hypothesized to influence species distribution and community composition through excluding species with low defenses from wet sites with high herbivore pressure (i.e. pest pressure gradient hypothesis, Baltzer and Davies 2012). Additionally, herbivore pressure can affect community diversity through density dependent mortality, which allows for species coexistence (Janzen-Connell Hypothesis, e.g. Bagchi et al. 2014). Higher pest pressure in wetter forests has thus been hypothesized to underlie the increase of plant diversity across tropical rainfall gradients (Givnish 1999).

Although these hypotheses for the role of herbivores in shaping tropical forest communities along moisture gradients are compelling, empirical support is still lacking. The pest pressure gradient hypothesis is based on two underlying assumptions: firstly, that herbivore pressure increases with moisture, and secondly, that herbivore defenses increase with moisture, as a consequence of selection pressure and/or environmental filtering. However, the opposite patterns of these assumptions have also been hypothesized, and the evidence remains contradictory, as is depicted below.

That herbivore pressure increases with moisture in tropical forests has been hypothesized by Givnish (1999), who argued that herbivorous insects should benefit from moist conditions, leading to high abundance in wet forests. In contrast, herbivore abundance has also been hypothesized to decrease with moisture: According to the slow-growth-high-mortality hypothesis, higher defenses in wet forest plants, evolved in the past, should delay the development time of herbivores, increasing their exposure to predators and parasitoids, and consequently their mortality, leading to lower current abundance in wet forests (Connahs et al. 2011).

Laboratory and field studies show contrasting evidence for moisture effects on insects. Increasing survival, longevity and reproduction with moisture has been shown under controlled conditions for various insect taxa (Broufas et al. 2009, Lu and Wu 2011). Temporally, insect abundance and diversity as well as herbivore damage often increase in the wet season in tropical forests (Wolda 1978, Connahs et al. 2011), although several studies show no clear pattern between rainfall and insect seasonality (Wolda 1988). Spatially, studies on insect abundance and diversity along tropical moisture gradients also show no consistent patterns. Higher insect abundance and diversity in more humid plots across a small scale transect were found in a rain forest in Costa Rica (Janzen and Schoener 1968). In contrast, lower abundance of beetles and lower number of leaf chewing beetle species in a wet than in a dry forest was found along a regional rainfall gradient in Panama (Charles and Basset 2005, Ødegaard 2006), and caterpillar abundance did not vary (Connahs et al. 2011). In summary, the evidence for changes of herbivore pressure with moisture remains contradictory.

For variation of plant defenses against herbivores across rainfall gradients, the hypotheses are also conflicting. Plant defenses have been hypothesized to increase with rainfall, due to adaptation to higher herbivore pressure in wet forests (Coley and Aide 1991, Coley and Barone 1996, Brenes-Arguedas et al. 2009, Baltzer and Davies 2012). An increase of herbivore defenses with rainfall has additionally been expected to result indirectly from lower light conditions in wetter forests favoring shade tolerant species (Givnish 1999, Brenes-Arguedas et al. 2011): Shade-tolerant plants are well-known to be better defended against herbivores than light requiring species (Coley and Barone 1996), because the costs of replacing leaf tissue are higher under limiting light (resource allocation theory (Barbour et al. 1980) based on the resource light). In contrast, herbivore defenses have also been hypothesized to show the opposite pattern and increase towards drier forests, arguing that in dry forests water is a strong limiting factor which selects for species with tough leaves and high tannins and phenol contents, which are at the same time effective herbivore defenses (Givnish 1999). Limiting water availability could also lead to higher costs of tissue loss towards drier forests, and thus result in higher defenses in dry forest species (Givnish (1999); resource allocation theory based on the resource water).

Empirical evidence for gradients in herbivore defenses with rainfall remains scarce. Results from transplant experiments showed no differences in herbivory rates between species restricted to aseasonal forests vs. widespread species or wet distribution vs. dry distribution species (Brenes-Arguedas et al. 2009, Baltzer and Davies 2012), suggesting no systematic differences in defenses. For individual leaf traits involved in herbivore defenses, comparisons between wet and dry sites support higher defenses in wetter forests for some traits (e.g. leaf toughness, low nutritional quality), but not for others (condensed tannins or crude fiber content), and for yet others (phenolics) the evidence is mixed (Coley and Aide 1991, Coley and Barone 1996, Santiago and Mulkey 2005, Dirzo and Boege 2008).

The actual herbivore damage experienced by plants is the combined outcome of herbivore pressure and the effectiveness of herbivore defenses. Again, no consistent spatial patterns emerge across moisture gradients. A review by Coley and Barone (1996) concluded that dry forest species suffer higher herbivory than wet forest species. In contrast, in a transplant experiment in Panama higher levels of herbivory were found in a wet forest than in a dry forest (Brenes-Arguedas et al. 2009), and plants in wet floodplains in Mexico had higher levels of herbivory than plants in dry hills (Boege and Dirzo 2004). No overall differences in the levels of herbivory between tree species in dry vs. wet forests were found in a further review (Dirzo and Boege 2008), and consistently, no difference of herbivory rates were detected in a transplant experiment between a seasonal and an aseasonal forest along the Malaysia/Thailand peninsula (Baltzer and Davies 2012).

All in all, although herbivores have for a long time been assumed to shape community composition in tropical forests along moisture gradients, the evidence about changes of herbivore pressure and plant defenses across tropical rainfall gradients remains contradictory.

In this study, we experimentally tested three alternative hypotheses concerning changes of herbivore defenses across tropical rainfall gradients:
[1] Plant species associated with wet forests exhibit *higher* defenses against herbivores than species associated with dry forests due to increasing herbivore pressure (pest pressure gradient hypothesis). Higher defenses in wetter forests are the footprint of higher herbivore pressure leading to environmental filtering from herbivores and/or of adaptation to higher selection pressure from herbivores;
[2] Species associated with wet forests exhibit *higher* defenses than species associated with dry forests, because under lower light levels in wetter forests, species are more shade tolerant and therefore better defended to avoid costly tissue replacement (resource allocation theory with respect to the resource light);
[3] Species associated with wet forests exhibit *lower* defenses than species associated with dry forests, because water limitation in drier forests increases the costs of tissue

replacement (resource allocation theory with respect to the resource water). Additionally, leaf traits associated with drought resistance may also mediate low leaf palatability.

We provide evidence for the hypotheses that plant defenses increase with association to wetter forests and independently also increase with shade tolerance. Our results thus support the idea that high herbivore pressure acts as a filter excluding dry forest species with low defenses from wet forests, and provide the first empirical evidence that herbivores are important in shaping tree species distribution patterns across tropical rainfall gradients.

Materials and methods

We evaluated differences in integrated herbivore defenses across a wide range of 50 focal shrub and tree species (Table S1) with known distribution along a pronounced rainfall gradient in Panama (Condit et al. 2013). We assessed feeding rejection behavior of a generalist herbivore in dual choice tests and related rejection rates to species' distribution across the rainfall gradient, as well as to their shade tolerance, association to soil nutrients, leaf longevity, several mechanical defense traits, and phylogenetic position (Figure S1).

Study area

The study was conducted in lowland tropical forests along a pronounced rainfall gradient across the Isthmus of Panama, ranging from 1600 mm/year to over 3000 mm/year along a distance of only 65 km (Engelbrecht et al. 2007, Condit et al. 2013). Dry season length (i.e. days with rainfall lower than evapotranspiration) correlates negatively with annual rainfall, and ranges from 147 days on the Pacific coast to 117 days on the Caribbean coast (Engelbrecht et al. 2007).

Study species, their regional distribution and shade tolerance

50 focal study species were selected out of 550 tree species with known distribution patterns with respect to rainfall (moisture, M) and nutrient conditions (nitrogen (N), phosphorus (P) and potassium (K)) in the study area (Condit et al. 2013). High association values indicate association to wet forests and nutrient rich soils, respectively, and low values indicate association to dry forests or nutrient poor soils. Species shade tolerance (ST) was based on the effect of light on tree growth in a large long-term forest dynamics plot in the center of the Isthmus of Panama (Rüger et al. 2011). The light dependence of growth of each species (b) was averaged between two census intervals and converted to a shade tolerance index (ST), so that 0 represents the most light-demanding and 1 the most shade tolerant species (ST = -b + 1). Values of the associations to rainfall, nutrients and shade tolerance of the focal species covered almost the entire available range (Figure S2).

Leaf material

We focused on juvenile plants (10 cm – 1 m height), the life stage at which local habitat filtering occurs in tropical forests (Baldeck et al. 2013). We collected leaves of individuals growing naturally in the understory of mature secondary forests across the Isthmus in the national parks San Lorenzo, Soberania, Chagres, Camino de Cruces, and in a forest near the Albrook community of Panama City. One fully expanded, young, healthy leaf was collected from each of at least 10 individuals per species, with each species collected in at least three different sites and individuals at least 30 m apart, avoiding any gaps or other high-light environments. We focused on young, fully expanded leaves to minimize effects of leaf age, and because herbivory on mature leaves has been shown to have the largest impact on survival (Eichhorn et al. 2010), although the majority of leaf area removal occurs on expanding leaves (Coley and Barone 1996, Eichhorn et al. 2010). All collections were done at the height of the rainy season in 2012, avoiding any confounding effects of acute drought stress. Leaves were collected in plastic bags and stored cool until processing in the laboratory after a maximum of six hours.

Standard species

Ixora coccinea L. (Rubiaceae) was used as a standard in all dual choice tests. It was selected based on preliminary experiments with a range of species common in gardens in the study area to allow for high availability throughout the experiments. The model herbivore (see below) exhibited intermediate preference for this non-native shrub, which was a prerequisite for dual choice tests with a wide range of study species. The youngest, fully expanded leaves were used in all tests.

Generalist herbivore

Dual choice tests were conducted with a generalist herbivore, the caterpillars of the moth *Spodoptera frugiperda* (J.E. Smith). The use of a generalist herbivore that does not occur in the forest (i.e. the habitat of the focal plant species) allowed to compare feeding behavior across a wide range of plant species and to avoid potential confounding effects of co-evolution with some of the focal plants. The relevance of the feeding behavior of generalist herbivores is supported by increasing evidence that tropical insect communities are more generalistic than originally assumed (Novotny and Basset 2005). The caterpillars were reared in the laboratory under standardized conditions (Perkins 1979) on artificial diet based on bean meal and agar. Each individual was used only for one trial to prevent any learning effects.

Dual choice feeding trials

For each dual choice feeding trial, one caterpillar (4th instar) was put in a Petri dish (90 mm diameter) with one leaf disk of the focal study species and one leaf disk of *Ixora coccinea* as a standard. Leaf disks (2 cm² area) were punched out with a cork borer avoiding the main leaf rib. Leaf disks were placed on moist sponges to prevent desiccation during the trials. Prior to the trials, caterpillars were starved for 6 h. Each feeding trial took 6 h. In cases where a caterpillar had not eaten from any leaf disk at the end of a feeding trial, the trial was repeated with new leaves and a new caterpillar. In general, 10 replicate trials [6-19] were conducted for each focal tree species.

A rejection index (RI) was calculated based on the remaining leaf area, as

$RI = -\log((LA_{Fs} + 0.0001) / (LA_S + 0.0001))$, where LA_{Fs} is the remaining leaf area of the focal species, and LA_S the remaining leaf area of the standard. Positive RI values indicate a rejection of the focal species, while negative values indicate a preference of the focal species relative to the standard. A RI of 0 indicates no rejection for any of the species. For further analyses, the median was used for each species, which is robust against outliers and skewed distributions.

Leaf defense traits and phylogeny

A number of mechanical leaf traits known to affect herbivory (Coley and Barone 1996, Moles et al. 2013) were assessed for each species. The occurrence of hairs or latex (present/absent), and deciduousness (deciduous/evergreen) were determined from various sources (Croat 1978, Woodson and Schery 1981) and from observations of the collected leaves. Leaf toughness was measured with a pressure gauge (Kursar and Coley 2003), and leaf mass per area (LMA) assessed from leaf area and leaf dry mass of the samples.

A phylogenetic tree of the study species was created based on the APG3 tree (Bremer et al. 2009), with branch lengths computed using the bladj function of Phylocom (Webb et al. 2011) and node age data according to Wikström et al. (2001).

Analysis

We analyzed the relation between RI and distribution indices and trait values as explanatory variables (Figure S1).

Differences of RI among species were assessed with a Kruskal-Wallis Test. Correlation among the explanatory variables was initially assessed (Table S2). In general, correlations among the examined species habitat associations and among physical defense traits were weak (Dormann et al. 2013). The only exception was a strong positive correlation between leaf mass per area (LMA) and leaf toughness, and leaf toughness was therefore removed from the analysis (Dormann et al. 2013).

A phylogenetic generalized least squares model (GLS) was used to assess the influence of environmental associations (M, N, P, K, and ST), leaf traits (LMA, latex, hairs and deciduousness) and phylogenetic relationships among species on the RI. All variables passed tests for normality of the residuals and homogeneity of variances. An

Ornstein-Uhlenbeck Motion model (Martins and Hansen 1997) was used as correlation structure based on the phylogenetic tree (see above).

Model simplification was done using likelihood ratio tests (Zuur 2009). The phylogenetic correlation structure as well as the explanatory variables N, P, K, LMA, latex, hairs and deciduousness were removed from the model, resulting in a minimal adequate model including M and ST as explanatory variables.

All statistics were performed with R, version 3.0.0 (R Core Team 2013) with the packages: nlme version 3.1-109 (Pinheiro et al. 2013) and ape version 3.0-8 (Paradis et al. 2004), and functions of the package AED version 1.0 (Zuur 2009).

Results

Rejection behavior varied significantly among plant species (Kruskal-Wallis test: $H_{49} = 261.7$, $p < 0.001$), covering almost the entire range from complete rejection to complete consumption of the focal species relative to the standard (Figure 1).

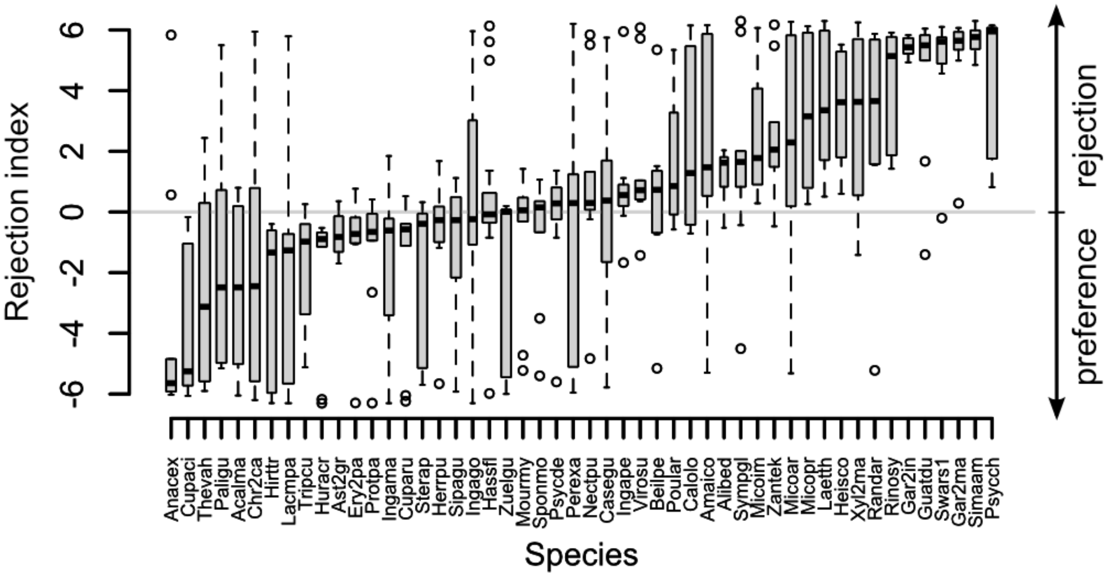

Figure 1: Herbivore leaf rejection differed significantly among 50 woody species from the Isthmus of Panama. It ranged from high preference of the focal species relative to a standard (negative rejection index values) to high rejection (positive rejection index values). Rejection indices were assessed in dual-choice feeding trials. The bar charts show the median (black line), interquartile range (grey box), minimum and maximum values within 1.5 times the interquartile range (dotted line) and outliers (points). For full species names see Table S1.

Rejection increased significantly with association of the focal species with wetter forests, as well as with shade tolerance, i.e. tree species associated with wetter forests were rejected stronger than species associated with drier forests, and shade tolerant species were rejected stronger than light requiring species (Figure 2).

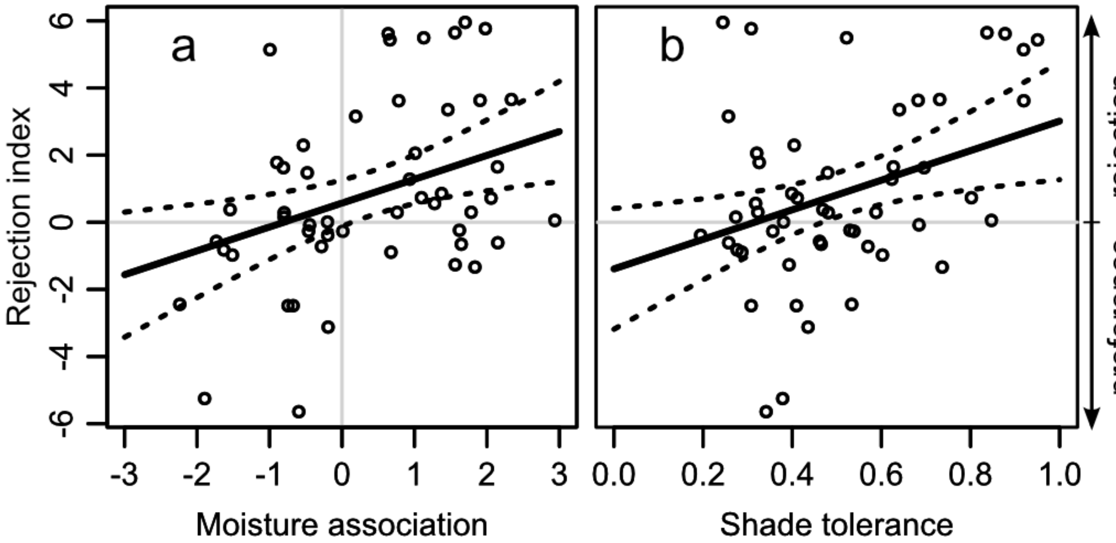

Figure 2: Leaf rejection increased with species' association with wet forests (a) and with species' shade tolerance (b). Both factors independently affected rejection behavior, since wet forest association and shade tolerance were not correlated with each other (Table S2). A high rejection index indicates a high rejection of the focal species compared to the standard, i.e. high herbivore defenses. Each point represents one tree species ($n = 50$). Dotted lines indicate ±95% confidence intervals. Association with moisture (a): $y = -0.71x + 1.67$, $p = 0.009$; shade tolerance (b): $y = -4.40x + 1.67$, $p = 0.01$.

Shade tolerance and moisture association were not correlated (Table S2), indicating that they affected rejection behavior independently. Association with moisture and shade tolerance together explained 24% of the variance of rejection ($R^2 = 0.24$, $F_{2,47} = 8.76$, $p < 0.001$).

The association of the tree species with soil nutrient availability, which reflects leaf nutrient contents (Ordoñez et al. 2009, Han et al. 2011), did not affect the rejection behavior. This lack of a correlation of rejection with nutrient associations shows that variation of defenses was directly driving the rejection behavior rather than preference for high leaf nutrient contents.

Of the physical and life history traits considered (Figure S1) none had a significant effect on leaf rejection. This is in line with previous findings that chemical defenses, rather than mechanical defenses, are more important in tropical systems (Eichhorn et al. 2007). The phylogenetic relatedness of the species also did not affect the rejection behavior.

Discussion

The observed relation of rejection behavior with moisture association of the species shows that leaf defenses increased towards wetter forests. Such a gradient in leaf defenses is consistent with environmental filtering mediated by herbivores, excluding poorly defended species from wetter forests with higher herbivore pressure, and/or with the evolution of more effective defenses in wetter forests. It thus provides strong community-level support for the pest pressure hypothesis (Baltzer and Davies 2012) and shows that herbivores are important drivers of plant distributions across tropical rainfall gradients. Our results imply that herbivore pressure is increasing towards wetter forests. The study therefore also lends indirect support for stronger negative density dependent mortality, allowing for higher levels of species coexistence and thus contributing to increasing tropical forest diversity with increasing rainfall (Givnish 1999).

Our results are also consistent with the resource allocation theory (Barbour et al. 1980) with respect to light, which states that herbivore defenses are higher in shade tolerant than in light requiring species. This trend is well supported in tropical woody plants (Coley and Barone 1996), and also clearly emerged in this study. However, species shade tolerance was not related to association with moisture (Table S2), i.e. species associated with wet sides were not more shade tolerant than species associated with dry sites (Brenes-Arguedas et al. 2011). The increase of defenses with association with moist sites was therefore not an indirect outcome of more shade tolerant species – with higher defenses and/or higher C/N ratios – but emerged independently. These results thus underline the direct importance of moisture for the distribution of tree defenses.

Our results are clearly not consistent with the resource allocation theory with respect to moisture, which predicts decreasing defenses towards wetter forests (Givnish 1999). In our system, water availability limits plant growth only in the dry season (Comita and Engelbrecht 2009). Thus resource limitation, which leads to high costs for replacing tissue loss, occurs only during times when herbivore pressure is low, weakening the need for strong defenses hypothesized by the resource allocation theory. In the wet season, when herbivore pressure is high, the resource water is abundant, reducing the cost for tissue replacement. Furthermore, increasing understory light levels towards drier forests and in the dry season when deciduous species shed their leaves facilitate tissue replacement, and reduce the importance of defenses in dry forests.

For pathogens, support for the pest pressure hypothesis was recently found in a reciprocal transplant study by showing that wet origin species had a higher tolerance against pathogen damage compared to dry origin species (Spear et al. 2015). For insect herbivores, to our knowledge only two studies explicitly tested the pest pressure hypothesis across tropical rainfall gradients (Brenes-Arguedas et al. 2009, Baltzer and Davies 2012). Both used a reciprocal transplant experiment with species of contrasting origins, but neither study found significantly higher herbivory rates in seedlings of species from drier than from wetter origin (Brenes-Arguedas et al. 2009, Baltzer and Davies 2012), reflecting differences in defenses, as required for the pest pressure hypothesis. Nevertheless, while Brenes-Arguedas et al. (2009) did not find significant differences between plant origins, there was a pronounced trend for higher leaf damage, especially through herbivores, in species with dry origin. Plant defenses frequently become more pronounced as plants mature (Boege and Marquis 2005, Barton and Koricheva 2010), and differences in herbivore damage and their consequences for plant performance may accumulate with longer exposure to herbivore pressure in the habitat. We therefore suggest that the differences in herbivory rates and plant performance between dry and wet origin species expected from the pest pressure hypothesis may take longer periods to manifest themselves than the relatively short timeframe of the existing studies.

Conclusions

By showing that herbivore defenses increase towards wetter forests this study provides the first community-level support that insect herbivores act as an important filter shaping tree species distributions across tropical rainfall gradients. Additionally, drought directly affects species distribution along rainfall gradients by excluding drought-sensitive species associated with wet forests from dry forests (Engelbrecht et al. 2007, Baltzer and Davies 2012). Changes in rainfall regimes in the tropics due to climate change will thus affect tree distributions and community composition both through direct drought effects on plants as well as through indirect effects on plant-herbivore interactions.

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References

Bagchi, R., R. E. Gallery, S. Gripenberg, S. J. Gurr, L. Narayan, C. E. Addis, R. P. Freckleton, and O. T. Lewis. 2014. Pathogens and insect herbivores drive rainforest plant diversity and composition. Nature 506:85–88.

Baldeck, C. A., K. E. Harms, J. B. Yavitt, R. John, B. L. Turner, R. Valencia, H. Navarrete, S. Bunyavejchewin, S. Kiratiprayoon, A. Yaacob, M. N. N. Supardi, S. J. Davies, S. P. Hubbell, G. B. Chuyong, D. Kenfack, D. W. Thomas, and J. W. Dalling. 2013. Habitat filtering across tree life stages in tropical forest communities. Proceedings of the Royal Society B: Biological Sciences 280:20130548.

Baltzer, J., S. Davies, S. Bunyavejchewin, and N. Noor. 2008. The role of desiccation tolerance in determining tree species distributions along the Malay-Thai Peninsula. Functional Ecology 22:221–231.

Baltzer, J. L., and S. J. Davies. 2012. Rainfall seasonality and pest pressure as determinants of tropical tree species' distributions. Ecology and Evolution 2:2682–2694.

Barbour, M. G., J. H. Burk, W. D. Pitts, F. S. Gilliam, and M. W. Schwartz. 1980. Terrestrial Plant Ecology. The Benjamin/Cummings Publishing Company, Inc., California, USA.

Barton, K. E., and J. Koricheva. 2010. The ontogeny of plant defense and herbivory: characterizing general patterns using meta-analysis. The American Naturalist 175:481–493.

Boege, K., and R. Dirzo. 2004. Intraspecific variation in growth, defense and herbivory in *Dialium guianense* (Caesalpiniaceae) mediated by edaphic heterogeneity. Plant Ecology 175:59–69.

Boege, K., and R. J. Marquis. 2005. Facing herbivory as you grow up: the ontogeny of resistance in plants. Trends in Ecology & Evolution 20:441–448.

Bremer, B., K. Bremer, M. Chase, M. Fay, J. Reveal, D. Soltis, P. Soltis, and P. Stevens. 2009. An update of the Angiosperm Phylogeny Group classification for the orders and families of flowering plants: APG III. Botanical Journal of the Linnean Society 161:105–121.

Brenes-Arguedas, T., P. D. Coley, and T. A. Kursar. 2009. Pests vs. drought as determinants of plant distribution along a tropical rainfall gradient. Ecology 90:1751–1761.

Brenes-Arguedas, T., A. B. Roddy, P. D. Coley, and T. A. Kursar. 2011. Do differences in understory light contribute to species distributions along a tropical rainfall gradient? Oecologia 166:443–456.

Broufas, G., M. Pappas, and D. Koveos. 2009. Effect of relative humidity on longevity, ovarian maturation, and egg production in the olive fruit fly (Diptera: Tephritidae). Annals of the Entomological Society of America 102:70–75.

Charles, E., and Y. Basset. 2005. Vertical stratification of leaf-beetle assemblages (Coleoptera: Chrysomelidae) in two forest types in Panama. Journal of Tropical Ecology 21:329–336.

Coley, P. D., and T. M. Aide. 1991. Comparison of herbivory and plant defenses in temperate and tropical broad-leaved forests. Pages 25–49 *in* Price, P. W. and Lewinsohn, T. M. and Fernandes, G. W. and Benson, W. W., editor. Plant–Animal Interactions: Evolutionary Ecology in Tropical and Temperate Regions. John Wiley & Sons, New York, USA.

Coley, P. D., and J. A. Barone. 1996. Herbivory and plant defenses in tropical forests. Annual Review of Ecology and Systematics 27:305–335.

Comita, L. S., and B. M. Engelbrecht. 2009. Seasonal and spatial variation in water availability drive habitat associations in a tropical forest. Ecology 90:2755–2765.

Condit, R., B. M. Engelbrecht, D. Pino, R. Pérez, and B. L. Turner. 2013. Species distributions in response to individual soil nutrients and seasonal drought across a community of tropical trees. Proceedings of the National Academy of Sciences of the United States of America 110:5064–5068.

Connahs, H., A. Aiello, S. Van Bael, and G. Rodriguez-Castaneda. 2011. Caterpillar abundance and parasitism in a seasonally dry versus wet tropical forest of Panama. Journal of Tropical Ecology 27:51–58.

Croat, T. B. 1978. Flora of Barro Colorado Island. Stanford University Press, Stanford, California, USA.

Dirzo, R., and K. Boege. 2008. Patterns of herbivory and defense in tropical dry and rain forests. Pages 63–78 *in* Carson, Walter and Schnitzer, Stefan, editor. Tropical Forest Community Ecology. Blackwell Science, West Sussex, UK.

Dormann, C. F., J. Elith, S. Bacher, C. Buchmann, G. Carl, G. Carré, J. R. G. Marquéz, B. Gruber, B. Lafourcade, P. J. Leitão, and others. 2013. Collinearity: a review of methods to deal with it and a simulation study evaluating their performance. Ecography 36:027–046.

Eichhorn, M. P., K. Fagan, S. G. Compton, D. H. Dent, and S. E. Hartley. 2007. Explaining leaf herbivory rates on tree seedlings in a Malaysian rain forest. Biotropica 39:416–421.

Eichhorn, M. P., R. Nilus, S. G. Compton, S. E. Hartley, and D. F. Burslem. 2010. Herbivory of tropical rain forest tree seedlings correlates with future mortality. Ecology 91:1092–1101.

Engelbrecht, B. M. J., L. S. Comita, R. Condit, T. A. Kursar, M. T. Tyree, B. L. Turner, and S. P. Hubbell. 2007. Drought sensitivity shapes species distribution patterns in tropical forests. Nature 447:80–83.

Fine, P. V., I. Mesones, and P. D. Coley. 2004. Herbivores promote habitat specialization by trees in Amazonian forests. Science 305:663–665.

Givnish, T. J. 1999. On the causes of gradients in tropical tree diversity. Journal of Ecology 87:193–210.

Han, W. X., J. Y. Fang, P. B. Reich, F. Ian Woodward, and Z. H. Wang. 2011. Biogeography and variability of eleven mineral elements in plant leaves across gradients of climate, soil and plant functional type in China. Ecology Letters 14:788–796.

Janzen, D. H., and T. W. Schoener. 1968. Differences in insect abundance and diversity between wetter and drier sites during a tropical dry season. Ecology 49:96–110.

Kursar, T. A., and P. D. Coley. 2003. Convergence in defense syndromes of young leaves in tropical rainforests. Biochemical Systematics and Ecology 31:929–949.

Lu, Y., and K. Wu. 2011. Effect of relative humidity on population growth of *Apolygus lucorum* (Heteroptera: Miridae). Applied Entomology and Zoology 46:421–427.

Martins, E. P., and T. F. Hansen. 1997. Phylogenies and the comparative method: a general approach to incorporating phylogenetic information into the analysis of interspecific data. The American Naturalist 149:646–667.

Moles, A. T., B. Peco, I. R. Wallis, W. J. Foley, A. G. Poore, E. W. Seabloom, P. A. Vesk, A. J. Bisigato, L. Cella-Pizarro, C. J. Clark, and others. 2013. Correlations between physical and chemical defences in plants: tradeoffs, syndromes, or just many different ways to skin a herbivorous cat? New Phytologist 198:252–263.

Novotny, V., and Y. Basset. 2005. Host specificity of insect herbivores in tropical forests. Proceedings of the Royal Society B: Biological Sciences 272:1083–1090.

Ødegaard, F. 2006. Host specificity, alpha-and beta-diversity of phytophagous beetles in two tropical forests in Panama. Biodiversity and Conservation 15:83–105.

Ordoñez, J. C., P. M. Van Bodegom, J.-P. M. Witte, I. J. Wright, P. B. Reich, and R. Aerts. 2009. A global study of relationships between leaf traits, climate and soil measures of nutrient fertility. Global Ecology and Biogeography 18:137–149.

Paradis, E., J. Claude, and K. Strimmer. 2004. APE: analyses of phylogenetics and evolution in R language. Bioinformatics 20:289–290.

Perkins, W. D. 1979. Laboratory rearing of the fall armyworm. Florida Entomologist 62:87–91.

Pinheiro, J., D. Bates, S. DebRoy, D. Sarkar, and R. C. Team. 2013. nlme: Linear and Nonlinear Mixed Effects Models. R package version 3.1-109.

Pyke, C. R., R. Condit, S. Aguilar, and S. Lao. 2001. Floristic composition across a climatic gradient in a neotropical lowland forest. Journal of Vegetation Science 12:553–566.

R Core Team. 2013. R: a language and environment for statistical computing.

Rüger, N., U. Berger, S. P. Hubbell, G. Vieilledent, and R. Condit. 2011. Growth strategies of tropical tree species: disentangling light and size effects. PloS One 6:e25330.

Santiago, L. S., and S. S. Mulkey. 2005. Leaf productivity along a precipitation gradient in lowland Panama: patterns from leaf to ecosystem. Trees 19:349–356.

Spear, E. R., P. D. Coley, and T. A. Kursar. 2015. Do pathogens limit the distributions of tropical trees across a rainfall gradient? Journal of Ecology 103:165–174.

ter Steege, H., N. Pitman, D. Sabatier, H. Castellanos, P. Van der Hout, D. C. Daly, M. Silveira, O. Phillips, R. Vasquez, T. Van Andel, J. Duivenvoorden, A. A. De Oliveira, R. Ek, R. Lilwah, R. Thomas, J. Van Essen, C. Baider, P. Maas, S. Mori, J. Terborgh, P. N. Vargas, H. Mogollon, and W. Morawetz. 2003. A spatial model of tree alpha-diversity and tree density for the Amazon. Biodiversity and Conservation 12:2255–2277.

Webb, C., D. Ackerly, and S. Kembel. 2011. Phylocom: software for the analysis of phylogenetic community structure and character evolution (with Phylomatic and Ecovolve), version 4.2, user's manual.

Wikström, N., V. Savolainen, and M. W. Chase. 2001. Evolution of the angiosperms: calibrating the family tree. Proceedings of the Royal Society B: Biological Sciences 268:2211–2220.

Wolda, H. 1978. Seasonal fluctuations in rainfall, food and abundance of tropical insects. Journal of Animal Ecology 47:369–381.

Wolda, H. 1988. Insect seasonality: why? Annual Review of Ecology and Systematics 19:1–18.

Woodson, R. E., and R. W. Schery. 1981. Flora of Panama. Parts 2-9. Annals of the Missouri Botanical Garden. Allen Press, Lawrence, Kansas, USA.

Zuur, A. F. 2009. Mixed Effects Models and Extensions in Ecology with R. Springer, New York, USA.

Supporting information

Herbivore defenses of tree species increase across a tropical rainfall gradient

Julian Gaviria, Björn Reineking, Ingolf Steffan-Dewenter and Bettina M. J. Engelbrecht

Figure S1: Overview of hypotheses and results for factors influencing herbivore leaf rejection

Figure S2: Comparison of associations with environmental variables between focal species and the entire available range

Table S1: Species used in this study and their families and abbreviations

Table S2: Spearman rank correlation coefficients (r) among the environmental responses and leaf defensive traits of the focal species

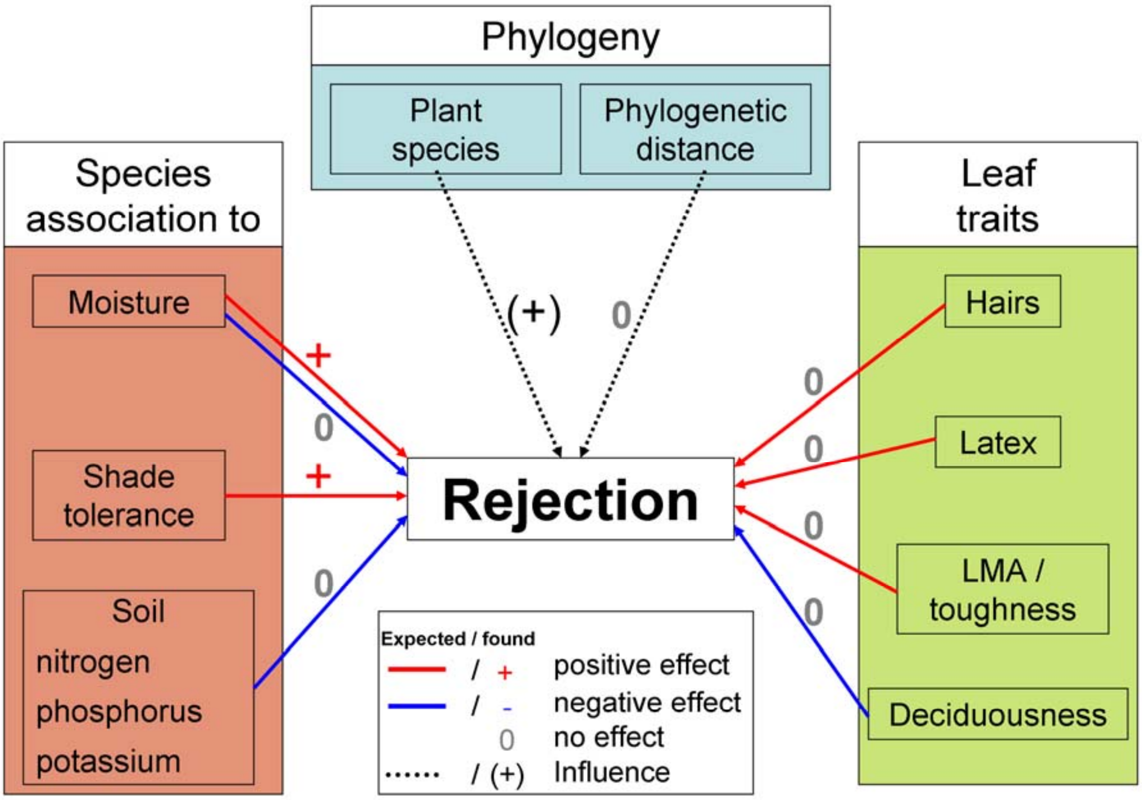

Figure S1: Overview of hypotheses and results for factors influencing herbivore leaf rejection. The arrows show the hypotheses, the signs the results. For moisture, two alternative hypotheses were considered (hypotheses 1 and 3, see text): an increase vs. a decrease of rejection with the association of the species with wet forests (pest pressure hypothesis and resources allocation hypothesis with respect to moisture, respectively). Shade tolerance was expected to increase rejection (resource allocation hypothesis with respect to light). Associations with high soil nutrients, indicative of high leaf nutrient contents, were expected to reduce rejection. Occurrence of hairs and latex as well as high leaf toughness or leaf mass per area (LMA) were expected to increase rejection, while rejection of deciduous species was expected to be lower than of evergreen species. The caterpillars were expected to show differential rejection behavior against plant species and to show more similar rejection values for closely related species.

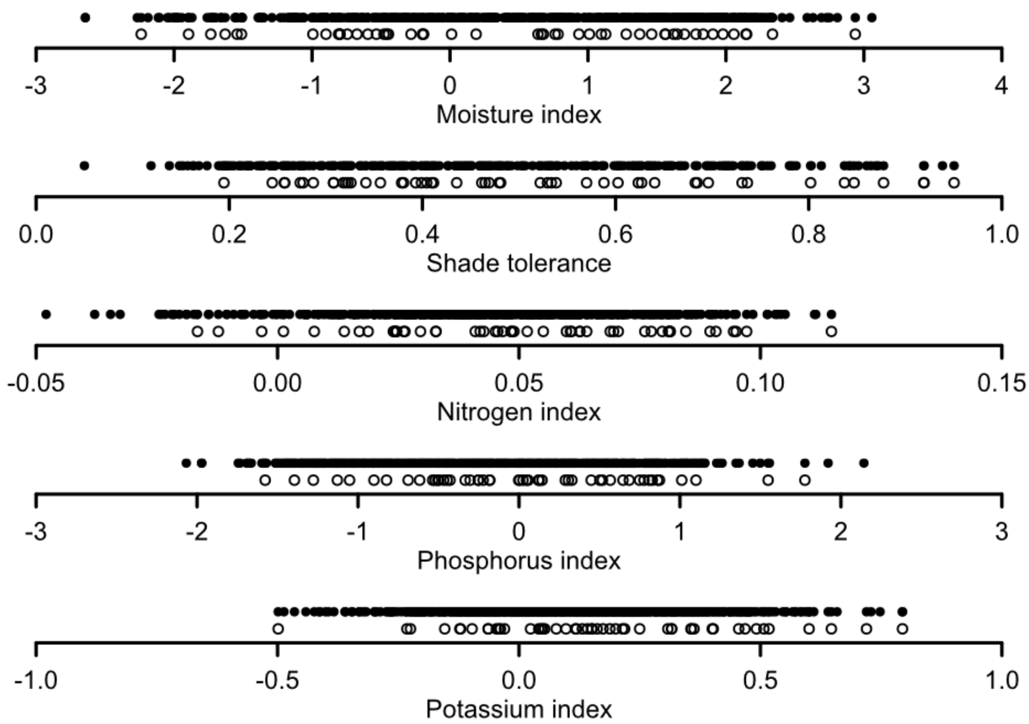

Figure S2: Comparison of associations with environmental variables between focal species and the entire available range The 50 focal species (open symbols) covered almost the entire range (filled symbols) of association with moisture (Condit et al. 2013), shade tolerance (Rüger et al. 2011) and association with major nutrients (nitrogen, phosphorus and potassium, Condit et al. 2013) encountered in the forest communities of the study area.

Table S1: Species used in this study and their families and abbreviations. The families follow the APG III system (Bremer et al. 2009).

species	ID	family
Acalypha macrostachya Jacq.	Acalma	Euphorbiaceae
Alibertia edulis (Rich.) A.Rich. ex DC.	Alibed	Rubiaceae
Amaioua corymbosa Kunth	Amaico	Rubiaceae
Anacardium excelsum Skeels	Anacex	Anacardiaceae
Astronium graveolens Jacq.	Ast2gr	Anacardiaceae
Beilschmiedia pendula (Sw.) Hemsl.	Beilpe	Lauraceae
Calophyllum longifolium Willd.	Calolo	Calophyllaceae
Casearia guianensis (Aubl.) Urb.	Casegu	Salicaceae
Chrysophyllum cainito L.	Chr2ca	Sapotaceae
Cupania cinerea Poepp. & Endl.	Cupaci	Sapindaceae
Cupania rufescens Triana & Planch.	Cuparu	Sapindaceae
Erythroxylum panamense Turcz.	Ery2pa	Erythroxylaceae
Garcinia intermedia (Pittier) Hammel	Gar2in	Clusiaceae
Garcinia madruno (Kunth) Hammel	Gar2ma	Clusiaceae
Guatteria dumetorum R. E. Fr.	Guatdu	Annonaceae
Hasseltia floribunda Kunth	Hassfl	Salicaceae
Heisteria concinna Standl.	Heisco	Olacaceae
Herrania purpurea (Pittier) R.E.Schult.	Herrpu	Malvaceae
Hirtella triandra Sw.	Hirttr	Chrysobalanaceae
Hura crepitans L.	Huracr	Euphorbiaceae
Inga goldmanii Pittier	Ingago	Fabaceae:Mimos.
Inga marginata Willd.	Ingama	Fabaceae:Mimos.
Inga pezizifera Benth.	Ingape	Fabaceae:Mimos.
Lacmellea panamensis (Woodson) Markgr.	Lacmpa	Apocynaceae
Laetia thamnia L.	Laetth	Salicaceae
Miconia argentea (Sw.) DC.	Micoar	Melastomataceae
Miconia impetiolaris (Sw.) D.Don ex DC.	Micoim	Melastomataceae
Miconia prasina (Sw.) DC.	Micopr	Melastomataceae
Mouriri myrtilloides (Sw.) Poir	Mourmy	Melastomataceae
Nectandra purpurea Mez	Nectpu	Lauraceae
Palicourea guianensis Aubl.	Paligu	Rubiaceae
Perebea xanthochyma H.Karst.	Perexa	Moraceae
Poulsenia armata (Miq.) Standl.	Poular	Moraceae
Protium panamense I.M.Johnst.	Protpa	Burseraceae
Psychotria chagrensis Standl.	Psycch	Rubiaceae
Psychotria deflexa DC.	Psycde	Rubiaceae
Randia armata (Sw.) DC.	Randar	Rubiaceae
Rinorea sylvatica (Seem.) Kuntze	Rinosy	Violaceae
Simarouba amara Aubl.	Simaam	Simaroubaceae
Siparuna guianensis Aubl.	Sipagu	Siparunaceae
Spondias mombin L.	Sponmo	Anacardiaceae
Sterculia apetala (Jacq.) H. Karst.	Sterap	Malvaceae
Swartzia simplex Spreng.	Swars1	Fabaceae:Papil.
Symphonia globulifera L. f.	Sympgl	Clusiaceae
Thevetia ahouai (L.) A.DC.	Thevah	Apocynaceae
Triplaris cumingiana Fisch. & C.A. Mey. ex C.A. Mey.	Tripcu	Polygonaceae
Virola surinamensis (Rol. ex Rottb.) Warb.	Virosu	Myristicaceae
Xylopia macrantha Triana & Planch.	Xyl2ma	Annonaceae
Zanthoxylum ekmanii (Urb.) Alain	Zantek	Rutaceae
Zuelania guidonia (Sw.) Britton & Millsp.	Zuelgu	Salicaceae

Table S2: Spearman rank correlation coefficients (r) among the environmental responses and leaf defensive traits of the focal species. Values of |r|<0.7 are typically low enough to prevent collinearity from affecting the model estimation (Dormann et al. 2013). M: association with moisture; N, P and K: association with soil nitrogen, phosphorus and potassium, respectively; tough: leaf toughness; LMA: leaf mass per area; ST: shade tolerance; hairs, latex and deciduous: occurrence of hairs, latex and deciduousness. Statistically significant correlations are in boldface.

	M	N	P	K	Tough	LMA	ST	Hairs	Latex
N	-0.135								
P	**-0.292**	0.026							
K	**-0.394**	-0.101	**0.536**						
Tough	0.099	0.053	-0.183	0.102					
LMA	0.054	-0.055	-0.151	0.137	**0.781**				
ST	0.122	-0.148	-0.007	0.128	**0.47**	**0.52**			
Hairs	-0.115	-0.017	0.006	-0.061	-0.075	-0.167	-0.085		
Latex	0.195	-0.117	-0.185	0.058	**0.308**	0.25	0.211	-0.129	
deciduous	-0.197	-0.046	**0.352**	0.248	**-0.378**	-0.153	-0.267	0.03	-0.12

Manuscript 3: Effects of drought, nutrient and light availability on tropical tree distribution: Results from a regional multisite, multispecies seedling transplant experiment

Effects of drought, nutrient and light availability on tropical tree distribution: Results from a regional multisite, multispecies seedling transplant experiment

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Abstract

Extensive changes of rainfall patterns are projected for tropical regions. An understanding of how rainfall regimes and soil moisture affect performance and distribution patterns of tropical tree species is urgently needed to improve models of consequences of such changes for tropical forest systems. Apart from direct effects of water availability, other factors such as nutrient and light availability have been hypothesized to limit performance and shape distribution patterns. However, the role of these different factors remains unclear.

We conducted a regional-scale multisite, multispecies seedling transplant experiment along a pronounced rainfall gradient across the Isthmus of Panama, and examined the effects of soil moisture, phosphorus and light availability, as well as drought resistance of the species on seedling performance. We directly linked the results to known species distribution patterns.

Our results indicate that soil water availability across space and time was the dominant factor limiting seedling regeneration across moist tropical forests, while nutrient and light availability played a minor role. Seedling performance across the rainfall gradient increased with species drought resistance. In congruence with previous studies, exclusion of wet origin species from dry forests is likely to occur through differential drought tolerance. However, especially dry years are needed. In contrast, low light and nutrient availability did not seem to exclude dry origin species from wet forests at the seedling stage. Dry origin species may be overgrown by wet origin species, which had intrinsic higher growth rates. These results suggest that performance differences in other life stages or over longer time spans are more important for shaping species distribution patterns.

Overall, our results underline that changes in rainfall patterns with global change will directly affect seedling performance of tropical trees, and will have pervasive consequences for species, forest composition and ecosystem function.

Keywords

Rainfall gradient, precipitation, water availability, soil moisture, nutrient availability, light availability, tropical forest, population dynamics, survival, growth, regeneration, distribution, habitat association

Introduction

Tropical forests are among the most diverse plant communities on earth. The controls of species distribution patterns and alpha and beta diversity in such species-rich communities remain a central question in ecology.

The most prominent pattern of diversity in tropical forests is an increase of species numbers with increasing rainfall and decreasing dry season length (Swaine and Becker 1999, ter Steege et al. 2003, Davidar et al. 2005). At the same time the change of forest composition, beta diversity, is extremely high along tropical rainfall gradients (Hall and Swaine 1976, Condit et al. 2002, Davidar et al. 2007). Species distribution patterns also vary with rainfall (Swaine 1996, Pyke et al. 2001, Engelbrecht et al. 2007, Condit et al. 2013).

While the patterns are well documented, the causes underlying these patterns remain poorly understood. Pronounced changes of rainfall patterns are projected for the tropics (Hulme and Viner 1998, Hidalgo et al. 2013), with potentially dramatic changes of forest distribution and occurrence and far reaching consequences for feedback mechanisms to climate (Malhi et al. 2009, IPCC 2013). Yet, these projections remain highly uncertain (IPCC 2013). Improving our understanding of the mechanisms underlying changes in tropical forest composition with rainfall is therefore urgently needed.

Variations in tree drought resistance together with spatial variation in water availability directly affect species distribution, and in turn forest diversity. The physiological tolerance hypothesis (Currie et al. 2004) states that drought periods act as a filter, excluding drought-sensitive species from drier areas and thus leading to lower species numbers. Pronounced correlations between species drought resistance and performance, and their distribution across tropical rainfall gradients offer strong support for drought directly shaping species distribution patterns (reviewed in Comita and Engelbrecht 2013).

However, the physiological tolerance hypothesis alone fails to explain a considerable part of the variation in species distribution along tropical rainfall gradients (Engelbrecht et al. 2007). Namely, it does not explain why dry forest species are excluded from wet forests leading to the observed high species turn-over (Condit et al.

2002). Additional factors that co-vary with rainfall, including nutrient and light availability, have been hypothesized to play an important role in shaping distribution patterns across rainfall gradients.

In lowland tropical forests nutrient availability, especially phosphorus, limits tree growth and survival, species distribution and ecosystem function (Vitousek 1984, Condit et al. 2013). In general, phosphorus availability decreases with increasing rainfall due to leaching (Austin and Vitousek 1998, Schuur and Matson 2001). Therefore, species growing in nutrient rich dry forests should have higher nutrient requirements, and thus be excluded from nutrient-poor wet forests (Swaine and Becker 1999, ter Steege et al. 2003, Brenes-Arguedas et al. 2008). We will refer to this as the nutrient availability distribution hypothesis.

In the understory of tropical forests, light availability is very low (Harms et al. 2004, Brenes-Arguedas et al. 2011). Additionally, higher light conditions in dry than wet forests are expected because of lower stem densities, lower leaf area indices and lower cloud cover as well as a higher proportion of deciduous species (Wright 1992, Markesteijn 2010, Brenes-Arguedas et al. 2011). There are strong differences in species' response to light availability, and some species may therefore cope better with low light availabilities than others (Swaine and Whitmore 1988). Dry forest species have been hypothesized to be more light-demanding than wet forest species, because they are adapted to the higher light regime in those forests (Brenes-Arguedas et al. 2011). Additionally, a trade-off between plant drought and shade tolerance has been hypothesized based on allocation trade-offs to optimize water uptake vs. light capture (Huston 1994). Drought resistant dry forest species should therefore have higher light requirements than wet forest species, and high light requirements may lead to the exclusion of dry origin species from wet, dark forests (light availability distribution hypothesis, Brenes-Arguedas et al. 2011).

These long-standing hypotheses are central to pressing ecological questions, and are frequently cited. However, rigorous studies of the role of the various factors potentially shaping distribution and diversity patterns across tropical rainfall gradients remain scarce (Engelbrecht et al. 2007, Brenes-Arguedas et al. 2008, 2009, 2011, Baltzer and Davies 2012, Spear et al. 2015). Correlations between moisture, light and nutrients make it challenging to separate the environmental factors and to test the specific hypotheses (Swaine 1996, Condit et al. 2013). To our knowledge, only one study has specifically tested the light availability distribution hypothesis (Brenes-Arguedas et al. 2011), and did not find support. To date, there is no convincing evidence that wetter forests are consistently darker (Engelbrecht 1998, Harms et al. 2004), that there is a trade-off between drought and shade tolerance in tropical plants (Engelbrecht et al. 2007, Markesteijn and Poorter 2009), or that dry forest species are more light-demanding (Brenes-Arguedas et al. 2011). Similarly, while partitioning of local and regional nutrient gradients by tropical trees has been shown (John et al. 2007, Condit et al. 2013), and fertilization increased performance in field experiments (Yavitt and Wright 2008, Alvarez-Clare et al. 2013), there is no conclusive support that nutrients are important for shaping species distributions along rainfall gradients (Brenes-Arguedas et al. 2008). Thus, the factors excluding dry forest species from wet forests and leading to the high beta diversity of tropical forests across rainfall gradients remain open.

Plant responses to environmental conditions may change with life stage, with younger stages being generally more vulnerable to stress (Harper 1977, Daws et al. 2005). Drought for example should hit younger seedlings stronger than older ones, since they have a smaller root system and may reach less water resources during dry seasons (Condit et al. 1995). Due to this constrains, the youngest seedling stage is the one when discrimination between origins is expected to take place (Grubb 1977). Here, we considered first-year seedling responses to drought, the stage when they naturally face their first dry season, and the one when they should be most vulnerable to drought.

The goal of this study was to assess the effect of drought, nutrients and light for species performance, and their respective role for species regional distribution patterns across a rainfall gradient. We established a regional seedling transplant experiment with 26 species in six forest sites across a pronounced rainfall gradient at the Isthmus of Panama. We followed seedling performance over one year, and assessed soil moisture and phosphorus content and light conditions. We directly linked the results to independently assessed drought resistance, and to distribution

(Engelbrecht et al. 2007), to test which of the factors influence regional species distribution patterns across the rainfall gradient.

We tested the physiological tolerance, the nutrient distribution and the light distribution hypothesis. Each of these hypotheses can be separated into two assumptions: First that environmental conditions change, with wet forests having higher moisture availability, lower nutrient availability, and casting deeper shade than dry forests, respectively. Second that dry and wet forest species differ in resource requirements, with dry forest species being more nutrient- and light-demanding, and more drought resistant than wet forest species. Each of these combinations of variation of environmental factors and species resource requirements could lead to a species performance advantage in their respective home range, relative to "foreign" species, and thus exclude foreign species and shape distribution patterns across rainfall gradients.

Overall, we expected an increase of species growth and survival with increasing soil moisture, phosphorus and light availability, but the effect should differ between species associated to dry vs. wet forests (origin x environmental factor interaction).
Specifically, we expected moisture to have a stronger effect on wet forest species especially in the dry season, when water is potentially limiting, and that the effect observed in the field correlates with independently assessed drought resistance of the species. We further expected phosphorus and light to have a stronger effect on dry forest species, and this effect to be especially pronounced in the wet season, when nutrient and light availability are lowest, and water is not limiting.

Materials and methods

Study area and forest sites

The study was conducted in lowland tropical forests in Central Panama. In the area, annual rainfall doubles from 1600 mm/year at the Pacific Coast to more than 3000 mm/year at the Caribbean Coast along a gradient of only 65 km; dry season length correlates negatively with annual rainfall, ranging from 147 days in the dry Pacific to 117 days in the wet Caribbean (Engelbrecht et al. 2007, Condit et al. 2013).

Soil nutrient variability in the area is high (Turner and Engelbrecht 2011, Condit et al. 2013), due to its complex geology (Pyke et al. 2001).

Experimental sites were chosen to include the full range of annual rainfall with three sites in the wetter range, and three sites in the drier range (Table S1), and to be accessible during the rainy season. Five of the sites had been previously established (Condit et al. 2002), and one additional 1 ha site (Cardenas) was established on the drier site of the isthmus following the same sampling design. Sites were mature secondary forests or old-growth forests, ranged from semi-deciduous to evergreen forests, and included a variety of geological formations including sedimentary and volcanic rocks and soil types (Pyke et al. 2001, Turner and Engelbrecht 2011). Although all are moist forests, we refer to drier ones as “dry” and the wetter ones as “wet”, for brevity.

Species and plant material

26 focal woody species were examined in this study. Focal species included shrubs, small and large trees, and were from 25 genera in 19 families (Table S2). Species were chosen based on the following criteria (a) a wide range of drought resistance, (b) a wide range of distribution patterns with respect to rainfall, and (c) capability to regenerate in the forest understory. Based on these criteria we opportunistically included those species for which we could collect and germinate sufficient seeds to yield more than 150 healthy seedlings per species for transplanting.

Drought resistance has previously been experimentally quantified as the percent seedling survival in dry relative to irrigated conditions in irrigation experiments in the forest understory (Table S2, Engelbrecht and Kursar 2003, Engelbrecht et al. 2007). Drought resistance of the focal species covered the full range from 0% to 98%. Higher values indicate higher drought resistance, while lower values indicate higher drought sensitivity. Distribution (referred to as origin in the analyses, see below) was quantified based on occurrence (presence/absence) of the species at 122 inventory sites across the rainfall gradient (Table S2) following Engelbrecht et al. (2007), with higher (more positive) values indicating a higher association of the species to dry sites. Distribution indices ranged from -4 to 6, and covered most of the range of common species in the area (-9.9 – 6.3, Engelbrecht et al. 2007). For our focal species, the distribution index from Engelbrecht et al. (2007) covered a broader range from “dry” to “wet” origin species than other distribution indices in the area (e.g. Condit et al. 2013), and it was thus preferred. The indices correlated and analyses gave qualitatively the same results. The study focused on species that are able to regenerate in the forest understory (i.e. excluding strict pioneers sensu Swaine and Whitmore 1988), since shade tolerant species represent the majority of the species in these forests (Bongers et al. 2005).

Seeds were collected mainly in the Barro Colorado Nature Monument in the center of the rainfall gradient, but also in other areas of the Panama Canal Watershed, from May - October 2006. Seeds were germinated in the greenhouse under moderately low light conditions (about 7%) in mixed forest soil. Seedlings were maintained with regular abundant watering, until they were transplanted to the forest plots about 6 - 8 weeks before the onset of the dry season (October / November 2006) to allow for establishment under moist conditions. Species age (1 - 7 months) and size (4 - 25 cm height) at the start of the experiment were thus equivalent to those in the natural forest environment. The first census (see below) was conducted at the start of the dry season, in December 2006. Of initially 3900 seedlings, 3374 (>85%) survived until the first census. All mortality prior to the first census was considered a transplanting effect, and seedlings were excluded from the analyses.

Experimental design

25 seedling plots were systematically established in each of the six 1 ha forest sites, with plots located in the center of a 20 m x 20 m grid. Seedling plots were 1.20 m x 1.20 m, with seedlings planted in a 20 cm x 20 cm grid. No gaps were present when establishing the plots. One seedling of each species was planted to each plot (150 in total), with species assigned randomly to positions, and seedlings assigned randomly to the plots (within species). Bare root seedlings (carefully extracted from the pots on-site) were directly transplanted into the soil. Disturbance of the leaf litter during transplanting was kept to a minimum to ensure natural microhabitat conditions in the plots.

Monitoring of seedling performance

Seedling performance in terms of growth (based on leaf area) and survival were assessed over one year. Seedling total leaf area and survival were monitored every three weeks from December 2006 - June 2007, and again at the end of the experiment in December 2007.
To assess the leaf area of each seedling, the length and width of every living leaf (or leaflet) was measured. The species-specific relation between leaf length x width, and leaf area measured with a leaf area meter (LICOR 3100), determined for at least 15 leaves per species, was used to calculate the potential area of each leaf (all $R^2 > 0.83$, see Table S3). The amount of damage to the leaf area (absent or dead) was estimated in 10% categories for each leaf, and subtracted from the potential leaf area. Survival was assessed based on stem color and elasticity. Dead seedlings were maintained in the census, and their status retrospectively changed if any signs of resprouting were detected.

Relative growth rate based on leaf area (RGR_{LA} in $cm^2\ cm^{-2}\ time^{-1}$) was calculated as $RGR_{LA} = (LA_{end} - LA_{start}) \times LA_{start}^{-1} \times \text{time period}^{-1}$ with LA_{start} and LA_{end} being leaf area at the beginning and the end of the time period analyzed (dry season, wet season, annual), respectively. Survival data (0 / 1) for each time period (dry season, wet season, annual) was directly implemented in the models (see below). For comparative purposes with other studies, monthly growth rates per time period were additionally calculated, using the actual time in months the seasons lasted (compare Figure S1). Similarly, we calculated survival rates per time period as well as mortality rates per month for each time period.

Monitoring of environmental conditions

Gravimetric soil water content, light availability in terms of canopy openness and soil phosphorus concentration were assessed in each seedling plot. Gravimetric soil water content was monitored with every seedling census (see above). Canopy openness was determined once at the height of the dry season (March 2007) and once during the wet season (July 2007) and soil phosphorus concentration once in the wet season (August 2007).

For gravimetric soil water content, a sample of the upper 10 cm of mineral soil was collected from a random location in each plot (off the seedlings). Fresh weight (FW) and dry weight (DW, after drying to constant weight at 105°C) of the samples were determined, and gravimetric soil water content (GW) calculated as
$GW = (FW - DW) \times DW^{-1} \times 100$.

To assess soil phosphorus content, soil cores of the upper 10 cm deep were collected. Plant-available phosphorus was extracted from soils using Mehlich-3 solution (Mehlich 1984) and concentrations of inorganic phosphorus in the Mehlich-3 extract determined by automated molybdate colorimetry (Turner and Engelbrecht 2011). Soil phosphorus concentrations of individual plots varied about a hundred-fold from 0.1 to 11.4 mg/kg (Figure S2).

Canopy openness (in %) was assessed with hemispherical photographs. Canopy openness assessed from hemispherical photographs correlates well with direct measures of light intensity in tropical forest understory (Engelbrecht and Herz 2001). Hemispherical photographs were taken using a Nikon Coolpix P5000 camera with a Fisheye Converter, and analyzed with the program Gap Light Analyzer v2 (Frazer et al. 1999). Overall, canopy openness ranged from 1.8 – 19.4% (Figure S2).

Analyses

The effects of environmental parameters on seedling growth and survival were analyzed both for the entire year, and separately for the dry and the wet season, and related to drought resistance and origin of the species.
We based analyses on seasonal and annual growth and survival, since we were interested in the overall outcome of processes during the dry season and the wet season or the whole year, and their role for species distributions. Monthly survival and growth rates were calculated for descriptive purposes and for comparability with other studies.

We defined the dry season as the period between the first strong and consistent decrease of soil moisture until the first census in which a > 10% increase in soil moisture was observed followed by another interval of soil moisture increase (Figure

S1). The wet season lasted from that date to the end of the experiment. Dry and wet season length thus varied among sites with the dry season lasting 129 - 87 and the wet season lasting 238 - 280 days in drier and wetter sites, respectively. The dry season was shorter than the long-term average (147 – 117 days (Engelbrecht et al. 2007), see above).

We initially assessed correlations among the explanatory variables soil moisture, phosphorus and light availability for each time period, as well as with rainfall (Table S4), and with origin and drought resistance. Correlations were weak enough to keep all variables in one model, since values of $|r|<0.7$ are typically low enough to prevent collinearity from affecting model estimations (Dormann et al. 2013).

One model per performance parameter (survival and RGR_{LA}) and time period (over the dry season, the wet season, and annual) was set up. Probability of survival was analyzed with Generalized Linear Mixed Effect Models (GLMM) using binomial distribution, and growth with Linear Mixed Effect Models (LMM). Random effects were site, plot and species, with plot nested in site and species nested in plot. The explanatory variables (fixed effects) used in every model were soil moisture, soil phosphorus content, canopy openness, species origin, and drought resistance. A main aim of this study was to test for differences in responses to environmental parameters with species origin (see hypotheses); we therefore included the interaction terms origin x soil moisture, origin x soil phosphorus and origin x canopy openness in every model. We additionally included the interaction soil moisture x drought resistance, to test if species drought resistance reduced negative effects of drought. Preliminary analyses showed a strong negative effect of soil phosphorus on species growth and survival. To test if this pattern is influenced by soil moisture availability, we also included the interaction soil moisture x soil phosphorus. Addition of this interaction did not qualitatively change the remaining results.

For dry season soil moisture we used the values observed when the minimum was reached in each site (Figure S1), which should be critical for survival. For the wet season we used the average of the wettest wet season months (December 2006, June 2007 and December 2007, see Figure S1), and for the annual analysis the whole-year average soil moisture.

For canopy openness, we used the respective value for each season, or the average for the annual analysis. Soil moisture as well as canopy openness measures were highly correlated between the different time periods (Table S4).

We checked the normality of the residuals and homogeneity of variances by visual inspection and all tests for over-dispersion passed. Single term deletion was used to sequentially remove non-significant factors from the model, resulting in a minimum adequate model (Zuur 2009). The results presented in Table 1 are the ones for the minimum model; slopes of non-significant variables show the values of their last occurrence in the model.

All the analyses were performed with the freeware R version 3.0.2 (R Core Team 2013) and the package lme4 1.0-5 (Bates et al. 2013). For the LMM, we assessed p-values using the package lmerTest 2.0-6 (Kuznetsova et al. 2014). Graphs were created with the package LMERConvenienceFunctions 2.5 (Tremblay et al. 2013). For clarity of the figures, origin and soil phosphorus content were categorized, rather than presented as continuous variables.

Results

Environmental conditions

With increasing rainfall, soil moisture increased, while soil phosphorus and canopy openness decreased, as expected (Figure S2, Table S4). The increase of soil moisture with rainfall was strong and significant in both seasons, and over the year (r = 0.56, 0.62 and 0.61 for the dry, wet and annual values, respectively, Figure S2, Table S4), and soil moisture was significantly higher in the wet season than the dry season in all plots (Figure S2). The relation with soil phosphorus was much weaker (r = -0.36), mainly because the rainiest site, situated on limestone, had the highest values of available phosphorus (Figure S2). Canopy openness only very weakly decreased with rainfall (r from -0.02 to -0.19), and only in the wet season the relation was significant (Table S4). In four of the sites, canopy openness was significantly higher in the dry than the wet season, while in the other two sites there were no significant seasonal differences (Figure S2).

Overall survival and growth rates

56% of the seedlings survived until the end of the experiment after one year (1879/3374, species range 20 - 82%). Over the dry season 70% of the seedling survived (species range 40 - 88%), and 80% (range 43 - 93%) of the ones surviving the dry season survived during the wet season. Monthly mortality rates were almost 3 times higher during the dry season (average 8% per month, range 3 - 17% per month) compared to the wet season (average 3% per month, species range 1 - 7% per month), with an overall annual mortality rate of 4% per month (species range 2 - 6% per month).

Relative leaf area change (RGR_{LA}) was overall lower in the dry season than in the wet season: On average, RGR_{LA} in the dry season was almost zero (0.0009 cm^2 cm^{-2} $season^{-1}$ or 0.0003 cm^2 cm^{-2} $month^{-1}$) with a wide variation among species from -0.253 to 0.227 cm^2 cm^{-2} $season^{-1}$ (or -0.066 to 0.066 cm^2 cm^{-2} $month^{-1}$). Thus, some species reduced their leaf area through leaf shedding, while others grew and developed new leaves over the dry season. In the wet season, RGR_{LA} was overall positive with an average of 0.243 cm^2 cm^{-2} $season^{-1}$ (0.027 cm^2 cm^{-2} $month^{-1}$). However, the variation among species was even higher ranging from 0.021 to 0.939 cm^2 cm^{-2} $season^{-1}$ (0.001 to 0.117 cm^2 cm^{-2} $season^{-1}$). Over the entire year RGR_{LA} averaged 0.122 (-0.116 to 0.583) cm^2 cm^{-2} $year^{-1}$ (corresponding to 0.028 (-0.001 to 0.089) cm^2 cm^{-2} $month^{-1}$).

Effect of environmental factors on performance and relations to species distribution

Moisture

Soil moisture had an overall positive effect on performance. Survival over the year increased with soil moisture for all species. The increase of survival with moisture was especially pronounced in the dry season (Table 1 a, Figure 1 A) when water availability is potentially limiting, underlining the importance of spatial variation of water availability during the dry season for seedling performance. Even in the wet season, survival was still significantly positively influenced by spatial variation of soil moisture, but the slope was much lower than in the dry season Table 1 b).

Table 1 Influence of soil moisture, soil phosphorus, light (canopy openness), origin and drought resistance of the species, as well as their interactions on seedling survival and growth (RGR_{LA}) for the dry season (a), the wet season (b) and the whole year (c). Given are the slopes of the relations and their significance; bold values are significant at the 0.05 level. The results are based on one model per time period and performance parameter (GLMM for survival, LMM for growth) totaling seven models. For non-significant variables that were removed during the model selection process, the slope for their last occurrence in the model is given.

	a) Dry season		b) Wet season		c) Annual	
Variables	survival	RGR_{LA}	survival	RGR_{LA}	survival	RGR_{LA}
Moisture	**0.040*****	**1.3e-2****	**0.019*****	2.4e-3	**0.035****	5.1e-3
Phosphorus	**-0.433*****	**-1.0e-1****	-0.086	8.8e-3	**-0.449****	**-2.0e-1***
Light	**-0.111****	**-2.3e-2(*)**	0.039	**6.8e-2*****	**-0.128***	1.7e-3
Drought resistance	**0.020*****	**5.3e-3*****	**0.013*****	1.6e-5	**0.032*****	1.4e-3
Origin	**0.083****	**-7.1e-2*****	**-0.064(*)**	**2.2e-2***	0.014	**-9.2e-2***
Interactions						
Origin x moisture	-5.3e-4	**1.5e-3****	0.001	3.3e-4	0.001	**1.9e-3***
Origin x phosphorus	-0.015	7.1e-4	0.010	-3.6e-3	0.009	2.0e-4
Origin x light	0.020	3.0e-3	0.002	6.3e-3	0.024	1.1e-2
Moisture x phosphorus	**0.015****	**4.3e-3****	0.2e-3	6.5e-4	**0.008(*)**	**5.6e-3****
Moisture x drought resistance	-6.6e-5	**-9.7e-5****	-0.1e-3	-2.1e-5	**-0.2e-3***	-8.2e-5

Significance codes: $p < 0.001$ ‘***’, $p < 0.01$ ‘**’, $p < 0.05$ ‘*’, $p < 0.1$ ‘(*)’, $p > 0.1$ ‘ns’

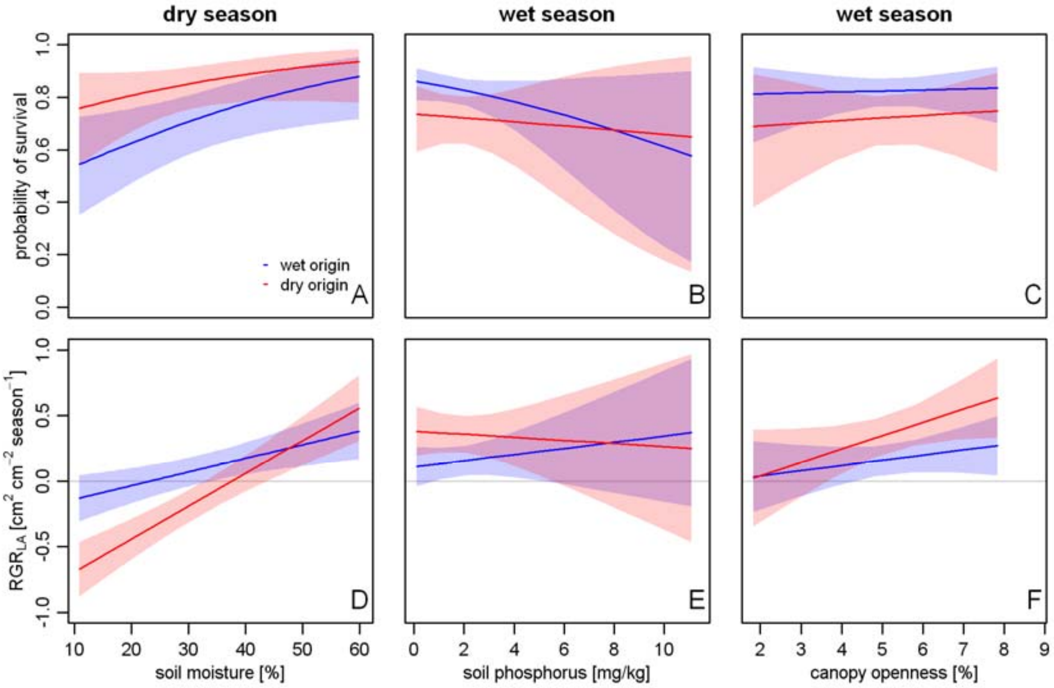

Figure 1: Relation of performance of seedlings of wet and dry origin species to moisture, phosphorus and light, and the respective season where they are expected to be most limiting: moisture in the dry season (A, D), and phosphorus (B, E) and light (canopy openness, C, F) in the wet season. Performance is given with respect to survival (A, B, C) and growth (RGR$_{LA}$, D, E, F). For clarity, origin is represented as a discrete factor (dry/wet) instead of continuous variable, as analyzed (Table 1). Shaded bands show the 95% Confidence Intervals (CI) for dry (red) and wet (blue) origin species, respectively. For significance of the main effects and interactions see Table 1.

The effect of spatial variation of soil moisture on growth was much weaker than on survival (Table 1). In the dry season, soil moisture again had a significant positive effect on RGR$_{LA}$, but in the wet season, and over the whole year, the soil moisture did not significantly influence growth (Table 1 b and c).

We had expected that wet origin species benefit more from higher soil moisture than dry origin species. However, survival increased strongly for all species, regardless of origin in both seasons and over the whole year, i.e. seedling survival responses in the experiment did not vary with origin (Table 1). While we did find a significant interaction between origin and soil moisture for dry season and annual RGR$_{LA}$, opposite to what we had expected, dry origin species benefited more from higher water availability than wet origin species. The interaction between origin and soil moisture was not significant for wet season RGR$_{LA}$.

Phosphorus

Growth and survival were overall negatively influenced by soil phosphorus availability during the dry season and over the whole year (Table 1 a and c), and were not affected by phosphorus during the wet season (Table 1 b), which is in contrast to our expectation that phosphorus should overall increase performance, and that this effect should be strongest in the wet season. The negative effect of high phosphorus on performance occurred in the driest sites (moisture x phosphorus interaction) in the dry season (see Table 1 a, Figure 2 A and D). Thus, high phosphorus conditions exacerbated the negative effects of drought, which is supported by the finding that the interaction was not significant during the wet season (Table 1 b, Figure 2 B and E). The negative effects of phosphorus in dry sites in the dry season are directly reflected in the annual survival and growth (Table 1 c, Figure 2 C and F).

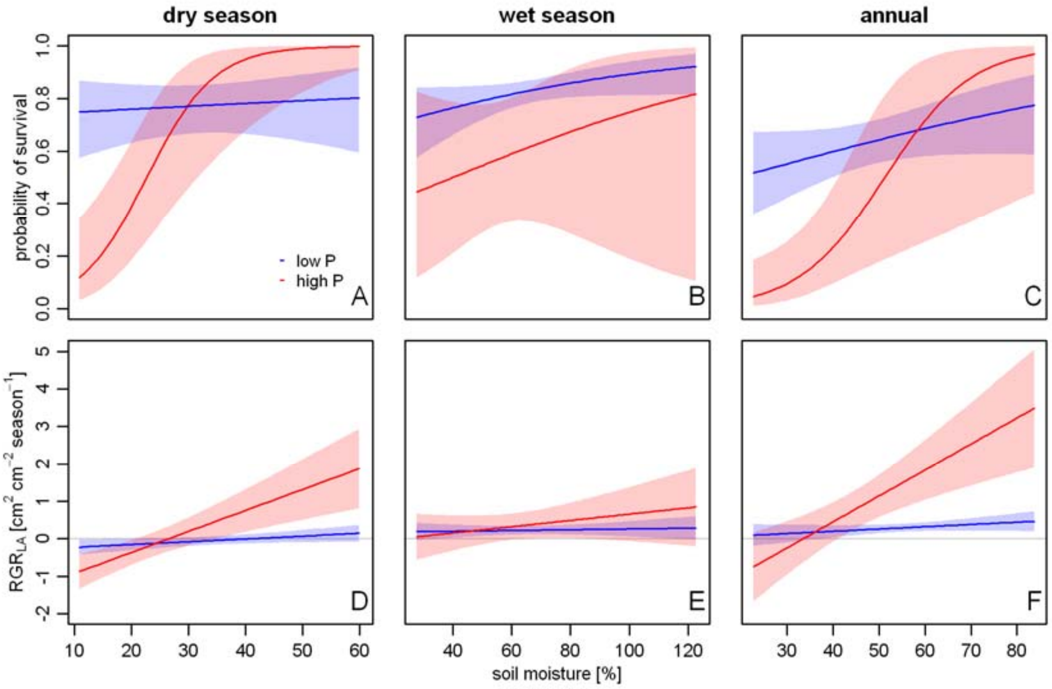

Figure 2: Relation of seedling performance to soil moisture under high vs. low phosphorus conditions. Survival (A, B, C) and growth (RGR_{LA}, D, E, F) are given over the dry season (A, D), the wet season (B, E), and the entire year (C, F). For clarity, soil phosphorus content is represented as a discrete factor (high/low) instead of continuous variable, as analyzed (Table 1). Shaded bands show the 95% confidence intervals (CI) for high (red) and low (blue) phosphorus conditions, respectively. For significance of the main effects and interactions see Table 1.

Opposite to our expectations, dry origin species were not more nutrient-demanding than wet origin species, as indicated by the lack of significant interactions between origin and the response to soil phosphorus for all performance parameters and seasons.

Light

The influence of light availability on species performance depended on the season: during the wet season, when we expected light to be most limiting, higher light conditions indeed significantly increased growth, but not survival (Table 1 b). In contrast, during the dry season, light had a negative effect on RGR_{LA} and survival (Table 1 a), indicating that light exacerbated the negative effects of drought. Over the whole year, light availability had a significant negative effect on survival, and a positive, though non-significant, effect on RGR_{LA} (Table 1 c).

Opposite to our expectations, light did not affect dry origin species more than species from wet origins, reflecting differences in light requirements, as the interaction origin x canopy openness was not significant for any of the performance parameters and seasons (Table 1).

Effects of drought resistance and origin on seedling performance

Drought resistance

Seedling drought resistance had previously been independently experimentally assessed for the species (Engelbrecht et al. 2007). Dry origin species were more drought resistant, as indicated by a significant positive relation between species distribution index and seedling drought resistance of the species ($r_{pearson} = 0.4$). With species drought resistance, dry season survival and growth increased (Table 1 a). During the wet season and over the whole year, only survival but not growth was positively related to drought resistance (Table 1 b, c).

As expected, drought resistant species tended to be less affected by lower water availabilities in the experiment than drought-sensitive species, as indicated by a negative slope of the interaction between soil moisture and drought resistance for all parameters and seasons (Table 1). However, the interaction was only significant for dry season and annual RGR_{LA} (Table 1 a and c).

Origin: do species have a home advantage?

Based on water limitation in the dry season, limiting performance of wet origin species in drier sites, and on light and/or nutrient limitation in the wet season, limiting performance of dry origin species in wetter sites, we had expected a performance rank reversal with species having a higher seedling performance in their home range than foreign species. We indeed found higher survival of dry origin species during the dry season (Table 1 a, significant increase of survival with origin index, indicative of dry forest association), and a slightly higher survival of wet origin species in the wet season (Table 1 b, marginally significant decrease of survival with origin index). However, after one year none of the origins had higher survival in their home range than foreign species, as indicated by a non-significant origin x moisture interaction (Table 1 c and Figure 3 A). Instead all species, regardless of their origin, survived better under moister conditions, indicating no home advantage of the origins at early seedling stages.

Regarding yearly growth, we found a higher increase in leaf area with increasing moisture for dry origin species compared to wet origin species (origin x moisture interaction, Table 1 c and Figure 3 B). This interaction was opposite to our expectations, thus origins also did not have a home advantage in terms of leaf area increase.

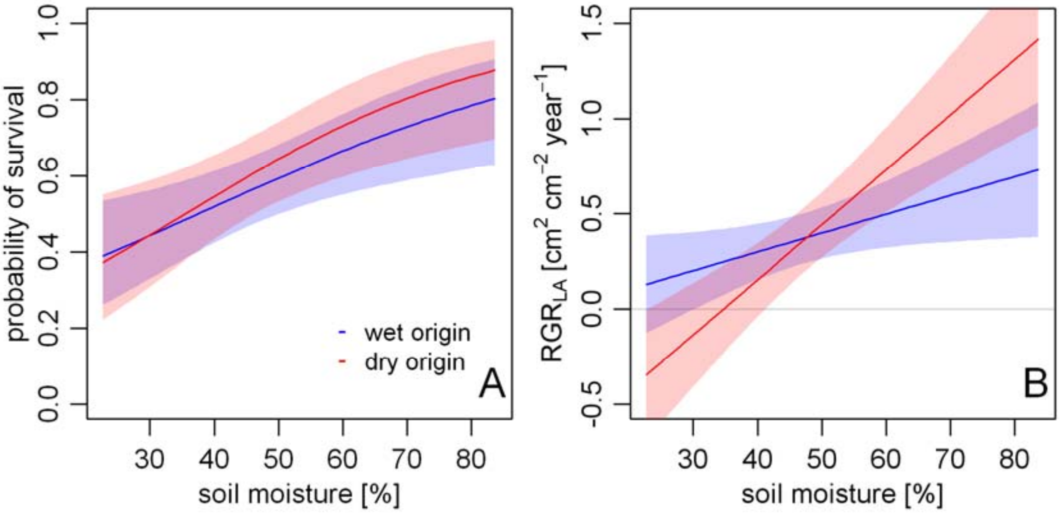

Figure 3: Relation of annual survival (A) and growth (RGR_{LA}, B) of seedling of dry and wet origin species to soil moisture. For clarity, origin is represented as a discrete factor (dry/wet) instead of continuous variable, as analyzed (Table 1). The shaded bands show the 95% Confidence Intervals (CI) for dry (red) and wet (blue) origin species, respectively. For significance of the main effects and interactions see Table 1.

Discussion

In this study, we aimed to disentangle the factors that shape species distribution patterns, leading to the high variation in beta diversity that can be found across tropical rainfall gradients. By analyzing the effects of environmental factors on performance of first-year seedlings with contrasting origins, we tried to understand which factors exclude dry-origin species form wet forests and *vice versa*. While drought has received strong support as the main factor excluding wet origin species from dry sites (summarized in Comita and Engelbrecht 2013), previous studies have failed to explain the mechanism that excludes dry origin species from wet sites. Also, in contrast to other studies in the area which have compared two or maximum three sites at the extremes (Brenes-Arguedas et al. 2009, 2011, Spear et al. 2015), here we used six sites spanning a rainfall gradient. This allowed us to assess the effects more rigorously, since we covered a broader variance of environmental conditions. Additionally, by using a transplant experiment across a gradient instead of experimental manipulations of environmental factors in a common garden (e.g. Bunker and Carson 2005, Engelbrecht et al. 2007), we were able to cover the natural conditions and combinations of environmental factors the seedlings have to face.

Below we first discuss the direct effects of the environmental factors moisture, phosphorus and light on seedling performance. Then we relate performance differences to species' drought resistance and origin, and discuss their role for shaping distribution patterns along rainfall gradients.

Species responses to variation in moisture, phosphorus and light

Moisture

Across sites, increasing moisture led to considerably higher survival and growth in the dry season, and survival in the wet season (Table 1 a and b). Our results therefore support that seedling performance in moist tropical forests is directly limited by soil water availability, in congruence with previous experimental and observational studies (Bunker and Carson 2005, Daws et al. 2005, Engelbrecht et al. 2007, Brenes-Arguedas et al. 2009, Comita and Engelbrecht 2009, 2013, Baltzer and Davies 2012, Amissah et al. 2015). Across seasons, seedling growth and survival were considerably higher in the wet than the dry season, consistent with previous studies (reviewed in Comita and Engelbrecht 2013). Lower light conditions, higher herbivore pressure and lower nutrient availability in the wet than in the dry season are potentially limiting seedling wet season performance, and it has been suggested that seedling may thus perform better in the dry season (Wright and Van Schaik 1994). Higher survival and growth in this study clearly indicate that overall these factors are less limiting than the direct effects of low water availability in the dry season. The direct role of drought in limiting seedling survival and growth is also supported by a number of experimental irrigation studies that led to higher performance with irrigation (reviewed in Comita and Engelbrecht 2013), underlining the importance of water availability for seedling regeneration in moist tropical forests, and the potential for excluding drought-sensitive species and thus shaping species local and regional distribution patterns.

Phosphorus

Across sites, seedling performance did not increase with soil phosphorus concentrations (Table 1), as we had expected, suggesting that phosphorus was not directly limiting seedling performance. Fertilization studies under high light

conditions in the greenhouse or in common gardens often lead to increased growth and survival in tropical seedlings (Gunatilleke et al. 1997, Yavitt and Wright 2008). However, in forests understory conditions, results are much less clear, depending on the nutrient considered and depending on other factors like herbivory (Santiago et al. 2012). Lack of immediate and pronounced responses to fertilization in the forest understory, and a lack of response to spatial variation of soil phosphorus in our study, indicates that other factors are more limiting, and nutrient relations are complex.
Instead, survival and growth overall even decreased with increasing phosphorus. This was due to a strong negative effect in the dry season and in the driest sites (Table 1 a, Figure 2 A and D), and this effect was strong enough to also be reflected in growth and survival over the whole year (see interaction soil moisture x phosphorus Table 1 a, Figure 2 C and F). Thus high phosphorus concentrations enhanced negative effects of drought.

It is unlikely that this effect was due to phosphorus toxicity. Soils in our plots had overall relatively low phosphorus contents, compared to other forests in the area (see Condit et al. (2013), and compared to fertilization experiments in tropical seedlings that showed performance increases (Gunatilleke et al. 1997). Additionally, most species are able to efficiently down-regulate their phosphorus uptake (Hawkesford et al. 2012). Instead, we believe that indirect effects of phosphorus on allocation patterns and/or interactions with mycorrhiza may have made the plants more susceptible to drought. Specifically, under high phosphorus conditions, a shift of biomass allocation towards aboveground parts at the expense of lower biomass allocation to roots (Chapin III 1980, Wright et al. 2011, Poorter et al. 2012), and/or reduced investment into mycorrhiza (Treseder 2004, Johnson 2010) may have impaired water uptake (Read 1991), and thus lead to reduced growth and survival under low soil moisture conditions.

Light

We had expected that higher light availabilities increases seedling performance, congruent with the general notion that light availability is limiting in the understory of tropical forests (Whitmore 1996), where light availability is very low, typically under 5% of overstory light availability (Harms et al. 2004, Brenes-Arguedas et al. 2011).

Species partitioning along light gradients has been shown to occur even along very small gradients of 0.2-6.5% transmittance within the understory (Montgomery and Chazdon 2002). However, in our study yearly seedling growth and survival did not increase with light (Table 1 c), indicating that light was not a main factor limiting seedling regeneration. A weak limitation of leaf area increase was observed in the wet season (Table 1 b). However, in the dry season light had negative effects on both survival and RGR_{LA} (Table 1 a), which goes together with the observation that water availability was lower in high light sites (Table S4). Thus, negative effects of drought were exacerbated under high light conditions in the dry season. Similar effects have been found in other studies in tropical forests (Gerhardt 1996, McLaren and McDonald 2003, Brenes-Arguedas et al. 2011), and may be due to increased temperature and evaporation under high light conditions.

Overall, our study provides strong support that seedling regeneration in tropical forests is directly limited by soil moisture availability. In contrast, neither soil phosphorus concentrations nor light was limiting seedling performance in our study. Instead, high phosphorus and high light conditions exacerbated drought effects in the dry season, and this effect was strong enough to be reflected in performance over the whole year.

In the following we will consider whether effects of soil moisture, phosphorus or light on seedling performance shape species distributions across the moisture gradient, and evaluate the physiological tolerance hypothesis and the nutrient distribution and light distribution hypotheses in the light of our results.

Physiological tolerance hypothesis

A growing body of literature shows that woody species in wetter or moister habitats are more drought-sensitive than species in drier sites and that drought acts as a filter excluding wet forest species from dry sites (Bunker and Carson 2005, Daws et al. 2005, Engelbrecht et al. 2007, Brenes-Arguedas et al. 2009, Comita and Engelbrecht 2009, 2013, Baltzer and Davies 2012, Amissah et al. 2015). Our results are overall consistent with these studies: species' previously independently experimentally quantified drought sensitivity (Engelbrecht et al. 2007) increased with species association to wet sites (see results), and more drought-sensitive species had lower annual survival in drier sites (significant moisture x drought resistance interaction, Table 1 c). Nevertheless, such effects were not strong enough to manifest themselves in higher survival of dry origin species in the drier sites over the dry season or the year, i.e. dry forest species did not gain an explicit home advantage over wet forest species during the course of this study. A home advantage in terms of survival across moisture gradients has previously been shown in some experiments (Comita and Engelbrecht 2009, Baltzer and Davies 2012), but not in others (Brenes-Arguedas et al. 2011, Spear et al. 2015). It has also been shown that the effect of drought varies considerably across years with different dry season intensity, with effects being most pronounced in extremely dry years, and weak in wetter years (Comita and Engelbrecht 2013). The dry season during the experimental year was relatively weak and shorter than normal (see methods), with more than twice as much rainfall in March, April and May compared to the long-term average (Paton 2008, ESP 2015). This may have precluded the expression of a survival advantage of drought resistant dry origin species in their home range, as had been previously found in other studies (Bunker and Carson 2005, Daws et al. 2005, Engelbrecht et al. 2007, Brenes-Arguedas et al. 2009, Comita and Engelbrecht 2009, 2013, Baltzer and Davies 2012, Amissah et al. 2015), and underlines the importance of pronounced and extreme dry season events for excluding wet origin species from dry forests.

Contrary to our expectations, dry origin species also did not have a home advantage in terms of leaf area increase, as yearly growth was lower in drier sites (Figure 3 B), mimicking dry season responses to drought (more negative growth rates (RGR_{LA}) of dry origin species compared to wet origin species in the driest places, see Table 1 a and Figure 1 D). This is likely to reflect facultative leaf shedding in response to drought in dry origin species, whereas wet origin species tended to maintain their leaves even under very dry conditions (Figure 1 D). Facultative leaf shedding in seedlings under drought has been shown even for species whose adults are not known to be deciduous (Engelbrecht and Kursar 2003). In our study, the species that were a priori known to be dry season deciduous were all associated to dry sites (Table S2), likely showing an adaptation to drought: Higher leaf loss under dry conditions may improve survival under dry conditions (Poorter and Markesteijn 2008, but see Engelbrecht and Kursar 2003, Comita and Engelbrecht 2013). The negative growth (representing leaf shedding) we found for dry origin species in drier sites may increase their dry season survival, indirectly giving them a home advantage. This relation changes among years with differing rainfall, being stronger in drier years (Comita and Engelbrecht 2013). In drier years the ability of leaf shedding may be the decisive factor that excludes non-leaf shedding, wet origin species from dry sites.

Thus, our results are overall consistent with the physiological tolerance hypothesis, and add to the increasing amount of studies that support the direct role of drought for species distributions along moisture availability gradients (Engelbrecht et al. 2005, 2007, Brenes-Arguedas et al. 2009, Comita and Engelbrecht 2009, 2013, Kursar et al. 2009, Baltzer and Davies 2012, Condit et al. 2013). Overall, the exclusion of wet origin species from dry forests can be explained by drought and differences in drought tolerance between species, especially during exceptionally dry years.

However, the factors that exclude dry origin species from wet forests remain uncertain. In this study, we explicitly addressed the role of nutrients and light.

Nutrient availability distribution hypothesis

According to the nutrient availability distribution hypothesis, wet forests should be nutrient poorer than dry forests due to leaching. However, in contrast to other areas where nutrients and rainfall are strongly correlated (Hall and Swaine 1976, Swaine 1996, Austin and Vitousek 1998, Schuur and Matson 2001), in our study there was no simple decline of soil phosphorus concentrations with rainfall. Indeed, our wettest site, situated on limestone, had the highest phosphorus concentrations (Figure S2). Other studies in the area also did not find strong relations between rainfall and soil nutrient content (Engelbrecht et al. 2007, Turner and Engelbrecht 2011), due to the complex geology of the Isthmus of Panama (Pyke et al. 2001).

Additionally, we found no indication that dry origin species are more nutrient-demanding than wet forest species, as indicated by the lack of interactions between the effect of soil phosphorus content and origin for seedling performance (Table 1). A greenhouse study in the area similarly did not support that species associated to drier forests have higher nutrient requirements than those associated with wet forests (Brenes-Arguedas et al. 2008), as required by the nutrient availability distribution hypothesis.

While nutrients have been shown to be important for shaping species distributions across local and regional nutrient gradients in tropical forests in Panama and other tropical areas (Fine et al. 2004, John et al. 2007, Condit et al. 2013), ours and previous results (Brenes-Arguedas et al. 2008) do not support that effects of phosphorous availability on seedling regeneration are driving regional distribution patterns along rainfall gradients. Indeed, even at the adult level the occurrence of strong species turn-over (Condit et al. 2002) without a corresponding pronounced correlation of phosphorus or other nutrients with rainfall (Turner and Engelbrecht 2011, Condit et al. 2013), implies that nutrients are not a main factor excluding dry forest species from wet forests in the area.

Light availability distribution hypothesis

It has long been assumed that light availability in the understory of tropical forests decreases with rainfall (Smith and Huston 1989), which is the basis for the light availability distribution hypothesis. However, studies that reported lower light conditions in wetter forests were based either on comparisons between single forest pairs (Santiago et al. 2004, Brenes-Arguedas et al. 2011), or were not designed for direct comparisons of forests along rainfall gradients with standardized methods (Parker et al. 2005, Peña-Claros et al. 2012). In this study across six sites along the regional rainfall gradient, we were able to better catch the variation in light availability. However, there was only a very weak decrease of light availability with increasing rainfall (Table S4), and it was only significant in the dry season. The highest light availabilities were actually measured in the dry season at the wettest site, and may have been a consequence of a high proportion of deciduous species in the high nutrient site on limestone (Bohlman 2010). Additionally, variation of light within the understory of the sites was high (Figure S2). Our study adds to others who did not find lower light availabilities with increasing rainfall (Engelbrecht 1998, Harms et al. 2004), and underline that processes shaping forest structure - and through that understory light conditions - of tropical lowland forests are complex (Toledo et al. 2011). They challenge the view that wetter forests are inherently darker.

Our results also do not support that dry origin species are more light-requiring than wet forest species, as required by the light availability distribution hypothesis. Brenes-

Arguedas et al. (2011) similarly found no evidence that dry origin species are more light-demanding than wet origin species in a separate species set in a transplant experiment to two sites in the area (5 species overlap). To our knowledge, there is to date no robust support for the assumption that dry origin species are generally more light-demanding than wet origin species. Higher light requirements for dry forest species have originally been hypothesized based on a trade-off between drought and shade tolerance, due to a carbon allocation trade-off to roots vs. leaves (Huston 1994). However, in tropical seedlings there is no robust evidence for a trade-off between drought and shade tolerance (Engelbrecht et al. 2007, Markesteijn and Poorter 2009), and a comprehensive study in a dry forest in Bolivia even showed convergence between drought and shade tolerance and the underlying traits (Markesteijn et al. 2011).

In summary, there is no evidence that higher light requirements of seedlings of dry forest species are excluding them from darker forests.

What excludes dry forest species from wet forests?

We had expected a performance rank reversal with species having a higher seedling performance in their home range. This home-range performance advantage should lead to the exclusion of species from foreign sites and explain how the combined effects of water, light and nutrients influence species distribution patterns.

Indeed, survival was higher for dry origin species in the dry season, and slightly higher for wet origin species in the wet season (Table 1 a and b). However after one year, species did not exhibit a home advantage regarding survival (Figure 3 A), as all survived better under moister conditions, regardless of their origin. Furthermore, neither phosphorus nor light had a differential effect on seedling performance of species with contrasting origins in the wet season (no significant origin x phosphorus or origin x light interaction, Table 1 a and b and Figure 1 B, C, E and F), consistent with previous studies (Brenes-Arguedas et al. 2008, 2011). This implies that at the seedling stage we considered here, neither nutrients, nor light limited seedling performance in wet sites to an extent that may lead to species exclusion.

It has also been suggested that herbivores and pathogens (summarized as pests) are important in excluding dry forest species from wet forests, with pest pressure being hypothesized to be higher in wet forests, and thus excluding poorly defended dry forest species (Baltzer and Davies 2012). We would have expected that in the wet season, when they are most abundant (Wolda 1978), pests reduce seedling performance of dry origin species in wet sites (i.e. a significant negative moisture x origin interaction), if pest pressure on seedlings is important excluding dry forest species from wet sites. Although we did not specifically quantify pest damage in this study, the lack of a moisture x origin interaction (Table 1 b) implies that effects of pest pressure on seedling performance do also not drive the exclusion of dry forest species from wet forests.

Of the studies that specifically tested the pest pressure gradient hypothesis through seedling transplant experiments, so far none found convincing evidence, i.e. lower performance of seedlings of dry forest species in wetter forests due to a combination of increased pest pressure in wetter sites, and lower defenses of dry forest species (Gaviria and Engelbrecht 2015, Brenes-Arguedas et al. 2009, Baltzer and Davies 2012, Spear et al. 2015). However, in the experiment of Spear et al. (2015), higher susceptibility to die after pathogen attack of dry forest species was suggested as an alternative mechanism that excludes dry forest species from wet sites. Also in a laboratory feeding trial with caterpillars, Gaviria et al. (unpublished) found a higher rejection rate of tree species associated to wetter sites, an indicative of higher defenses, as expected from the pest pressure gradient hypothesis. The role of pests in shaping tree species distribution patterns therefore remains under debate.

Together, these results indicate that effects of phosphorus or light availability, or pest pressure on the performance of established, young seedlings do not exclude dry forest species from wet forests, and thus do not drive the high beta diversity across rainfall gradients. There are two other mechanisms that may explain the exclusion of dry origin species from wet forests: differential growth rates of dry and wet origin species, and different responses to environmental conditions at earlier or later life stages. These mechanisms are depicted in more detail below.

After one year, dry origin species had overall significantly lower RGR_{LA} than wet origin species (Table 1 c). This is consistent with previous studies that found higher growth rates in wet compared to dry origin species both in the area and other tropical forests (Gaviria and Engelbrecht 2015, Baltzer et al. 2007, Brenes-Arguedas et al. 2008, 2009, Baltzer and Davies 2012), suggesting that it is a widespread pattern. These differences in growth between origins may be related to a drought tolerance-growth trade-off, based on differential investment in organs that increase drought tolerance (roots, dense wood, cavitation-resistant xylem, etc.) vs. organs that increase growth (leaves) (Smith and Huston 1989). If this is a general pattern, slow growing dry origin species may be overgrown by wet origin species and in the long term excluded from wet forests. However, while the existence of this trade-off is widely accepted and many traits that are related to drought tolerance are also related to lower growth rates (Poorter et al. 2010, O'Brien et al. 2015), direct experimental evidence at the whole-plant performance level is scarce and contradictory (Fernandez and Reynolds 2000, Polley et al. 2002, Wikberg and Ögren 2004).

Our study, as well as several previous ones, focused on the performance of first-year seedlings to examine the processes driving species distributions across tropical rainfall gradients (Gaviria and Engelbrecht 2015, Brenes-Arguedas et al. 2008, 2009, 2011, Baltzer and Davies 2012), because early regeneration stages are considered the most vulnerable to environmental stressors and are a bottleneck in population dynamics (Harper 1977, Daws et al. 2005, Brenes-Arguedas et al. 2009). However, neither ours nor previous studies supported that this life stage is crucial for excluding species associated to dry sites from wet sites, i.e. seedlings of dry forest species did not have lower performance in wet sites than wet forest species (Figure 3). It has been shown that in tropical forests, trees have developed the distribution patterns they exhibit as adults by the time they reach 1 cm diameter at breast height (Baldeck et al. 2013). This strongly suggests that life stages other than established first year seedlings may be more important for excluding dry forest species from wetter forests.

Species responses to environmental conditions change with ontogeny (Comita et al. 2007): Light requirements of species may increase with ontogeny and plant size (Lusk et al. 2008), given the cost of additional supporting tissue (stems, branches) a bigger plant has to invest in. Differential species responses to low light availability may therefore be more pronounced at older life stages (Lusk et al. 2008).

Most shade-tolerant understory species have big seeds, which make them more independent of other resources like soil nutrients during the early regeneration stage

(Milberg et al. 1998). Therefore, at this stage differential nutrient requirements of species play probably a minor role. Indeed, in temperate forests it has been shown that nutrient requirements of trees are higher for adults than for saplings and much higher than for seedlings, leading to shifts towards nutrient-rich niches with ontogeny (Bertrand et al. 2011). If this process occurs also in tropical forests, the role of nutrients for species discrimination would only be visible at later life stages.

Plant defenses also change with ontogeny, with younger seedlings being generally less defended (Boege and Marquis 2005, Barton and Koricheva 2010). Especially the germination phase might be susceptible to pathogens and herbivory, and thus limit the establishment of poorly defended dry forest species in wet forests with high pest pressure. However, previous studies did not find a differential effect of pest pressure on species mortality at the germination and seed-to-seedling transition stage (Gaviria and Engelbrecht 2015, Spear et al. 2015). In contrast, defenses of older seedlings and saplings have been shown to increase with species association to wet forests (Gaviria et al. unpublished). Processes excluding dry forest species from wetter forests may therefore act at later life stages than the first year seedlings we considered in our study (Comita et al. 2007), and may also accumulate over time.

Conclusions

Our results are consistent with previous studies that show that drought effects on young, established seedlings are important for excluding wet forest species from dry forests. The rainfall gradient examined in this study covered a wide range of the precipitation conditions in moist tropical forests, and supports that this applies to wide areas of tropical forests. Effects of drought on seedling performance vary with strength of the dry season (Comita and Engelbrecht 2013), and more pronounced or extreme dry seasons may exacerbate the observed effects. Due to climate change, frequency and intensity of exceptionally dry years, like during El Niño events, are likely to increase (Hulme and Viner 1998), altering the effects of drought on species regeneration, distribution and consequently community composition.

On the other hand, our results indicate that effects of phosphorus or light availability, or pest pressure on the performance of established, young seedlings do not exclude dry forest species from wet forests, and thus do not drive the high beta diversity across rainfall gradients. However, other (later) life stages than the ones considered here may respond differentially to these environmental factors, and be more important for the exclusion of dry origin species from wet forests.

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References

Alvarez-Clare, S., M. C. Mack, and M. Brooks. 2013. A direct test of nitrogen and phosphorus limitation to net primary productivity in a lowland tropical wet forest. Ecology 94:1540–1551.

Amissah, L., G. M. J. Mohren, B. Kyereh, and L. Poorter. 2015. The effects of drought and shade on the performance, morphology and physiology of Ghanaian tree species. PloS One 10:e0121004.

Austin, A. T., and P. Vitousek. 1998. Nutrient dynamics on a precipitation gradient in Hawai'i. Oecologia 113:519–529.

Baldeck, C. A., K. E. Harms, J. B. Yavitt, R. John, B. L. Turner, R. Valencia, H. Navarrete, S. Bunyavejchewin, S. Kiratiprayoon, A. Yaacob, M. N. N. Supardi, S. J. Davies, S. P. Hubbell, G. B. Chuyong, D. Kenfack, D. W. Thomas, and J. W. Dalling. 2013. Habitat filtering across tree life stages in tropical forest communities. Proceedings of the Royal Society B: Biological Sciences 280:20130548.

Baltzer, J. L., and S. J. Davies. 2012. Rainfall seasonality and pest pressure as determinants of tropical tree species' distributions. Ecology and Evolution 2:2682–2694.

Baltzer, J. L., S. J. Davies, N. S. M. Noor, A. R. Kassim, and J. V. LaFrankie. 2007. Geographical distributions in tropical trees: can geographical range predict performance and habitat association in co-occurring tree species? Journal of Biogeography 34:1916–1926.

Barton, K. E., and J. Koricheva. 2010. The ontogeny of plant defense and herbivory: characterizing general patterns using meta-analysis. The American Naturalist 175:481–493.

Bates, D., M. Maechler, B. Bolker, and S. Walker. 2013. lme4: Linear mixed-effects models using Eigen and S4. R package version 1.0-5.

Bertrand, R., J.-C. Gégout, and J.-D. Bontemps. 2011. Niches of temperate tree species converge towards nutrient-richer conditions over ontogeny. Oikos 120:1479–1488.

Boege, K., and R. J. Marquis. 2005. Facing herbivory as you grow up: the ontogeny of resistance in plants. Trends in Ecology & Evolution 20:441–448.

Bohlman, S. A. 2010. Landscape patterns and environmental controls of deciduousness in forests of central Panama. Global Ecology and Biogeography 19:376–385.

Bongers, F., M. P. E. Parren, and D. Traore. 2005. Forest Climbing Plants of West Africa - Diversity, Ecology and Management. CABI, London.

Brenes-Arguedas, T., P. D. Coley, and T. A. Kursar. 2009. Pests vs. drought as determinants of plant distribution along a tropical rainfall gradient. Ecology 90:1751–1761.

Brenes-Arguedas, T., M. Rios, G. Rivas-Torres, C. Blundo, P. D. Coley, and T. A. Kursar. 2008. The effect of soil on the growth performance of tropical species with contrasting distributions. Oikos 117:1453–1460.

Brenes-Arguedas, T., A. B. Roddy, P. D. Coley, and T. A. Kursar. 2011. Do differences in understory light contribute to species distributions along a tropical rainfall gradient? Oecologia 166:443–456.

Bunker, D. E., and W. P. Carson. 2005. Drought stress and tropical forest woody seedlings: effect on community structure and composition. Journal of Ecology 93:794–806.

Chapin III, F. S. 1980. The mineral nutrition of wild plants. Annual Review of Ecology and Systematics 11:233–260.

Comita, L. S., R. Condit, and S. P. Hubbell. 2007. Developmental changes in habitat associations of tropical trees. Journal of Ecology 95:482–492.

Comita, L. S., and B. M. Engelbrecht. 2009. Seasonal and spatial variation in water availability drive habitat associations in a tropical forest. Ecology 90:2755–2765.

Comita, L. S., and B. M. Engelbrecht. 2013. Drought as a driver of tropical tree species regeneration dynamics and distribution patterns. Pages 261–308 *in* Coomes, David A and Burslem, David FRP and Simonson, William D, editor. Forests and Global Change. Cambridge University Press, Cambridge, UK.

Condit, R., B. M. Engelbrecht, D. Pino, R. Pérez, and B. L. Turner. 2013. Species distributions in response to individual soil nutrients and seasonal drought across a community of tropical trees. Proceedings of the National Academy of Sciences of the United States of America 110:5064–5068.

Condit, R., S. P. Hubbell, and R. B. Foster. 1995. Mortality rates of 205 neotropical tree and shrub species and the impact of a severe drought. Ecological Monographs 65:419–439.

Condit, R., N. Pitman, E. G. Leigh, J. Chave, J. Terborgh, R. B. Foster, P. Nunez, S. Aguilar, R. Valencia, G. Villa, H. C. Muller-Landau, E. Losos, and S. P. Hubbell. 2002. Beta-diversity in tropical forest trees. Science 295:666–669.

Croat, T. B. 1978. Flora of Barro Colorado Island. Stanford University Press, Stanford, California, USA.

Currie, D. J., G. G. Mittelbach, H. V. Cornell, R. Field, J.-F. Guégan, B. A. Hawkins, D. M. Kaufman, J. T. Kerr, T. Oberdorff, E. O'Brien, and J. Turner. 2004. Predictions and tests of climate-based hypotheses of broad-scale variation in taxonomic richness. Ecology Letters 7:1121–1134.

Davidar, P., J. Puyravaud, and E. Leigh. 2005. Changes in rain forest tree diversity, dominance and rarity across a seasonality gradient in the Western Ghats, India. Journal of Biogeography 32:493–501.

Davidar, P., B. Rajagopal, D. Mohandass, J.-P. Puyravaud, R. Condit, S. Wright, and E. Leigh. 2007. The effect of climatic gradients, topographic variation and species traits on the beta diversity of rain forest trees. Global Ecology and Biogeography 16:510–518.

Daws, M. I., T. R. Pearson, D. F. P. Burslem, C. E. Mullins, and J. W. Dalling. 2005. Effects of topographic position, leaf litter and seed size on seedling demography in a semi-deciduous tropical forest in Panama. Plant Ecology 179:93–105.

Dormann, C. F., J. Elith, S. Bacher, C. Buchmann, G. Carl, G. Carré, J. R. G. Marquéz, B. Gruber, B. Lafourcade, P. J. Leitão, and others. 2013. Collinearity: a review of methods to deal with it and a simulation study evaluating their performance. Ecography 36:027–046.

Engelbrecht, B. M. J. 1998. Ökologie und Ökophysiologie von koexistierenden Piper-Arten im Unterwuchs tropischer Regenwälder: "...wo der Pfeffer wächst..." Technische Universität Darmstadt, Darmstadt, Germany.

Engelbrecht, B. M. J., L. S. Comita, R. Condit, T. A. Kursar, M. T. Tyree, B. L. Turner, and S. P. Hubbell. 2007. Drought sensitivity shapes species distribution patterns in tropical forests. Nature 447:80–83.

Engelbrecht, B. M. J., and H. M. Herz. 2001. Evaluation of different methods to estimate understorey light conditions in tropical forests. Journal of Tropical Ecology 17:207–224.

Engelbrecht, B. M. J., and T. A. Kursar. 2003. Comparative drought-resistance of seedlings of 28 species of co-occurring tropical woody plants. Oecologia 136:383–393.

Engelbrecht, B. M., T. A. Kursar, and M. T. Tyree. 2005. Drought effects on seedling survival in a tropical moist forest. Trees 19:312–321.

ESP. 2015. Data sets provided by the Terrestrial-Environmental Sciences Program of the Smithsonian Tropical Research Institute. Available at: http://stri.si.edu/sites/esp/

Fernandez, R. J., and J. F. Reynolds. 2000. Potential growth and drought tolerance of eight desert grasses: lack of a trade-off? Oecologia 123:90–98.

Fine, P. V., I. Mesones, and P. D. Coley. 2004. Herbivores promote habitat specialization by trees in Amazonian forests. Science 305:663–665.

Frazer, G. W., C. Canham, and K. Lertzman. 1999. Gap Light Analyzer (GLA), Version 2.0: Imaging software to extract canopy structure and gap light transmission indices from true-colour fisheye photographs, users manual and program documentation.

Gaviria, J., and B. Engelbrecht. 2015. Effects of drought, pest pressure and light availability on seedling establishment and growth: their role for distribution of tree species across a tropical rainfall gradient. PloS One 10:e0143955.

Gaviria, J., B. Reineking, I. Steffan-Dewenter, and B. M. Engelbrecht. (unpublished). Herbivore defenses increase along a tropical rainfall gradient.

Gerhardt, K. 1996. Effects of root competition and canopy openness on survival and growth of tree seedlings in a tropical seasonal dry forest. Forest Ecology and Management 82:33–48.

Grubb, P. J. 1977. The maintenance of species-richness in plant communities: the importance of the regeneration niche. Biological Reviews 52:107–145.

Gunatilleke, C., I. Gunatilleke, G. Perera, D. Burslem, and P. Ashton. 1997. Responses to nutrient addition among seedlings of eight closely related species of Shorea in Sri Lanka. Journal of Ecology 85:301–311.

Hall, J., and M. Swaine. 1976. Classification and ecology of closed-canopy forest in Ghana. Journal of Ecology 64:913–951.

Harms, K. E., J. S. Powers, and R. A. Montgomery. 2004. Variation in small sapling density, understory cover, and resource availability in four Neotropical forests. Biotropica 36:40–51.

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

Hawkesford, M., W. Horst, T. Kichey, H. Lambers, J. Schjoerring, I. S. Møller, and P. White. 2012. Chapter 6 - Functions of Macronutrients. Pages 135 – 189 *in* Marschner, Petra, editor. Marschner’s Mineral Nutrition of Higher Plants (Third Edition). Academic Press, San Diego.

Hidalgo, H. G., J. A. Amador, E. J. Alfaro, and B. Quesada. 2013. Hydrological climate change projections for Central America. Journal of Hydrology 495:94–112.

Hijmans, R. J., S. E. Cameron, J. L. Parra, P. G. Jones, and A. Jarvis. 2005. Very high resolution interpolated climate surfaces for global land areas. International Journal of Climatology 25:1965–1978.

Hulme, M., and D. Viner. 1998. A climate change scenario for the tropics. Climatic Change 39:145–176.

Huston, M. A. 1994. Biological Diversity: The Coexistence of Species. Cambridge University Press, Cambridge, UK.

IPCC. 2013. Climate Change 2013: The Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change. Page 1535 (Stocker, T.F., Qin, D., Plattner, G.-K., Tignor, M., Allen, S.K., Boschung, J., Nauels, A., Xia, Y., Bex, V., Midgley, P.M., Ed.). Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.

John, R., J. W. Dalling, K. E. Harms, J. B. Yavitt, R. F. Stallard, M. Mirabello, S. P. Hubbell, R. Valencia, H. Navarrete, M. Vallejo, and R. B. Foster. 2007. Soil nutrients influence spatial distributions of tropical tree species. Proceedings of the National Academy of Sciences of the United States of America 104:864–869.

Johnson, N. C. 2010. Resource stoichiometry elucidates the structure and function of arbuscular mycorrhizas across scales. New Phytologist 185:631–647.

Kursar, T. A., B. M. Engelbrecht, A. Burke, M. T. Tyree, B. EI Omari, and J. P. Giraldo. 2009. Tolerance to low leaf water status of tropical tree seedlings is related to drought performance and distribution. Functional Ecology 23:93–102.

Kuznetsova, A., P. B. Brockhoff, and R. H. B. Christensen. 2014. lmerTest: Tests for random and fixed effects for linear mixed effect models (lmer objects of lme4 package). R package version 2.0-6.

Lusk, C., D. S. Falster, C. Jara-Vergara, M. Jimenez-Castillo, and A. Saldaña-Mendoza. 2008. Ontogenetic variation in light requirements of juvenile rainforest evergreens. Functional Ecology 22:454–459.

Malhi, Y., L. E. O. C. Aragão, D. Galbraith, C. Huntingford, R. Fisher, P. Zelazowski, S. Sitch, C. McSweeney, and P. Meir. 2009. Exploring the likelihood and mechanism of a climate-change-induced dieback of the Amazon rainforest. Proceedings of the National Academy of Sciences of the United States of America 106:20610–20615.

Markesteijn, L. 2010. Drought Tolerance of Tropical Tree Species: Functional Traits, Trade-offs and Species Distribution. Wageningen, NL.

Markesteijn, L., and L. Poorter. 2009. Seedling root morphology and biomass allocation of 62 tropical tree species in relation to drought-and shade-tolerance. Journal of Ecology 97:311–325.

Markesteijn, L., L. Poorter, F. Bongers, H. Paz, and L. Sack. 2011. Hydraulics and life history of tropical dry forest tree species: coordination of species' drought and shade tolerance. New Phytologist 191:480–495.

McLaren, K., and M. McDonald. 2003. The effects of moisture and shade on seed germination and seedling survival in a tropical dry forest in Jamaica. Forest Ecology and Management 183:61–75.

Mehlich, A. 1984. Mehlich 3 soil test extractant: A modification of Mehlich 2 extractant. Communications in Soil Science and Plant Analysis 15:1409–1416.

Milberg, P., M. A. Pérez-Fernández, and B. B. Lamont. 1998. Seedling growth response to added nutrients depends on seed size in three woody genera. Journal of Ecology 86:624–632.

Montgomery, R., and R. Chazdon. 2002. Light gradient partitioning by tropical tree seedlings in the absence of canopy gaps. Oecologia 131:165–174.

O'Brien, M. J., D. F. R. P. Burslem, A. Caduff, J. Tay, and A. Hector. 2015. Contrasting nonstructural carbohydrate dynamics of tropical tree seedlings under water deficit and variability. New Phytologist 205:1083–1094.

Parker, G., C. Tinoco-Ojanguren, A. Martínez-Yrízar, and M. Maass. 2005. Seasonal balance and vertical pattern of photosynthetically active radiation within canopies of a tropical dry deciduous forest ecosystem in Mexico. Journal of Tropical Ecology 21:283–295.

Paton, S. 2008. 2007 Meteorological and hydrological summary for Barro Colorado Island. Smithsonian Tropical Research Institute, Panama City, Panama.

Peña-Claros, M., L. Poorter, A. Alarcón, G. Blate, U. Choque, T. S. Fredericksen, M. J. Justiniano, C. Leaño, J. C. Licona, W. Pariona, and others. 2012. Soil effects on forest structure and diversity in a moist and a dry tropical forest. Biotropica 44:276–283.

Polley, H. W., C. R. Tischler, H. B. Johnson, and J. D. Derner. 2002. Growth rate and survivorship of drought: CO2 effects on the presumed tradeoff in seedlings of five woody legumes. Tree Physiology 22:383–391.

Poorter, H., K. J. Niklas, P. B. Reich, J. Oleksyn, P. Poot, and L. Mommer. 2012. Biomass allocation to leaves, stems and roots: meta-analyses of interspecific variation and environmental control. New Phytologist 193:30–50.

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

Poorter, L., I. McDonald, A. Alarcón, E. Fichtler, J.-C. Licona, M. Peña-Claros, F. Sterck, Z. Villegas, and U. Sass-Klaassen. 2010. The importance of wood traits and hydraulic conductance for the performance and life history strategies of 42 rainforest tree species. New Phytologist 185:481–492.

Pyke, C. R., R. Condit, S. Aguilar, and S. Lao. 2001. Floristic composition across a climatic gradient in a neotropical lowland forest. Journal of Vegetation Science 12:553–566.

R Core Team. 2013. R: a language and environment for statistical computing.

Read, D. J. 1991. Mycorrhizas in ecosystems. Experientia 47:376–391.

Santiago, L. S., K. Kitajima, S. J. Wright, and S. S. Mulkey. 2004. Coordinated changes in photosynthesis, water relations and leaf nutritional traits of canopy trees along a precipitation gradient in lowland tropical forest. Oecologia 139:495–502.

Santiago, L. S., S. J. Wright, K. E. Harms, J. B. Yavitt, C. Korine, M. N. Garcia, and B. L. Turner. 2012. Tropical tree seedling growth responses to nitrogen, phosphorus and potassium addition. Journal of Ecology 100:309–316.

Schuur, E. A., and P. A. Matson. 2001. Net primary productivity and nutrient cycling across a mesic to wet precipitation gradient in Hawaiian montane forest. Oecologia 128:431–442.

Smith, T., and M. Huston. 1989. A theory of the spatial and temporal dynamics of plant communities. Vegetatio 83:49–69.

Spear, E. R., P. D. Coley, and T. A. Kursar. 2015. Do pathogens limit the distributions of tropical trees across a rainfall gradient? Journal of Ecology 103:165–174.

ter Steege, H., N. Pitman, D. Sabatier, H. Castellanos, P. Van der Hout, D. C. Daly, M. Silveira, O. Phillips, R. Vasquez, T. Van Andel, J. Duivenvoorden, A. A. De Oliveira, R. Ek, R. Lilwah, R. Thomas, J. Van Essen, C. Baider, P. Maas, S. Mori, J. Terborgh, P. N. Vargas, H. Mogollon, and W. Morawetz. 2003. A spatial model of tree alpha-diversity and tree density for the Amazon. Biodiversity and Conservation 12:2255–2277.

Swaine, M. D. 1996. Rainfall and soil fertility as factors limiting forest species distributions in Ghana. Journal of Ecology 84:419–428.

Swaine, M. D., and P. Becker. 1999. Woody life-form composition and association on rainfall and soil fertility gradients in Ghana. Plant Ecology 145:167–173.

Swaine, M., and T. Whitmore. 1988. On the definition of ecological species groups in tropical rain forests. Vegetatio 75:81–86.

Toledo, M., L. Poorter, M. Peña-Claros, A. Alarcón, J. Balcázar, C. Leaño, J. C. Licona, and F. Bongers. 2011. Climate and soil drive forest structure in Bolivian lowland forests. Journal of Tropical Ecology 27:333–345.

Tremblay, A., D. University, J. Ransijn, and U. of Copenhagen. 2013. LMERConvenienceFunctions: A suite of functions to back-fit fixed effects and forward-fit random effects, as well as other miscellaneous functions. R package version 2.5.

Treseder, K. K. 2004. A meta-analysis of mycorrhizal responses to nitrogen, phosphorus, and atmospheric CO2 in field studies. New Phytologist 164:347–355.

Turner, B. L., and B. M. Engelbrecht. 2011. Soil organic phosphorus in lowland tropical rain forests. Biogeochemistry 103:297–315.

Vitousek, P. M. 1984. Litterfall, nutrient cycling, and nutrient limitation in tropical forests. Ecology 65:285–298.

Whitmore, T. 1996. A review of some aspects of tropical rain forest seedling ecology with suggestions for further enquiry. Pages 3–40 *in* Swaine, MD, editor. The Ecology of Tropical Forest Tree Seedlings. UNESCO and The Parthenon Publishing Group, Paris, France.

Wikberg, J., and E. Ögren. 2004. Interrelationships between water use and growth traits in biomass-producing willows. Trees 18:70–76.

Wolda, H. 1978. Seasonal fluctuations in rainfall, food and abundance of tropical insects. Journal of Animal Ecology 47:369–381.

Wright, S. J. 1992. Seasonal drought, soil fertility and the species density of tropical forest plant communities. Trends in Ecology & Evolution 7:260–263.

Wright, S. J., and C. P. Van Schaik. 1994. Light and the phenology of tropical trees. The American Naturalist 143:192–199.

Wright, S. J., J. B. Yavitt, N. Wurzburger, B. L. Turner, E. V. Tanner, E. J. Sayer, L. S. Santiago, M. Kaspari, L. O. Hedin, K. E. Harms, and others. 2011. Potassium, phosphorus, or nitrogen limit root allocation, tree growth, or litter production in a lowland tropical forest. Ecology 92:1616–1625.

Yavitt, J. B., and S. J. Wright. 2008. Seedling growth responses to water and nutrient augmentation in the understorey of a lowland moist forest, Panama. Journal of Tropical Ecology 24:19–26.

Zuur, A. F. 2009. Mixed Effects Models and Extensions in Ecology with R. Springer, New York, USA.

Supporting information

Effects of drought, nutrient and light availability on tropical tree distribution: Results from a regional multisite, multispecies seedling transplant experiment

Julian Gaviria and Bettina M. J. Engelbrecht

Figure S1: Gravimetric soil water content in 6 forest sites across the isthmus over one year.

Figure S2: Gravimetric soil moisture, soil phosphorus content and light availability measured as percent canopy openness for the six experimental sites and the two seasons.

Table S1: Site characteristics of the six 1 ha forest sites across the Isthmus of Panama, sorted by rainfall.

Table S2: Study species and their family, drought resistance, origin and deciduousness.

Table S3: Parameters used to calculate the leaf area of each species, following the formula leaf area = a x (length x width) + b, with length and width the measure of a focal leaf.

Table S4: Spearman rank correlation coefficients (r) among the environmental factors.

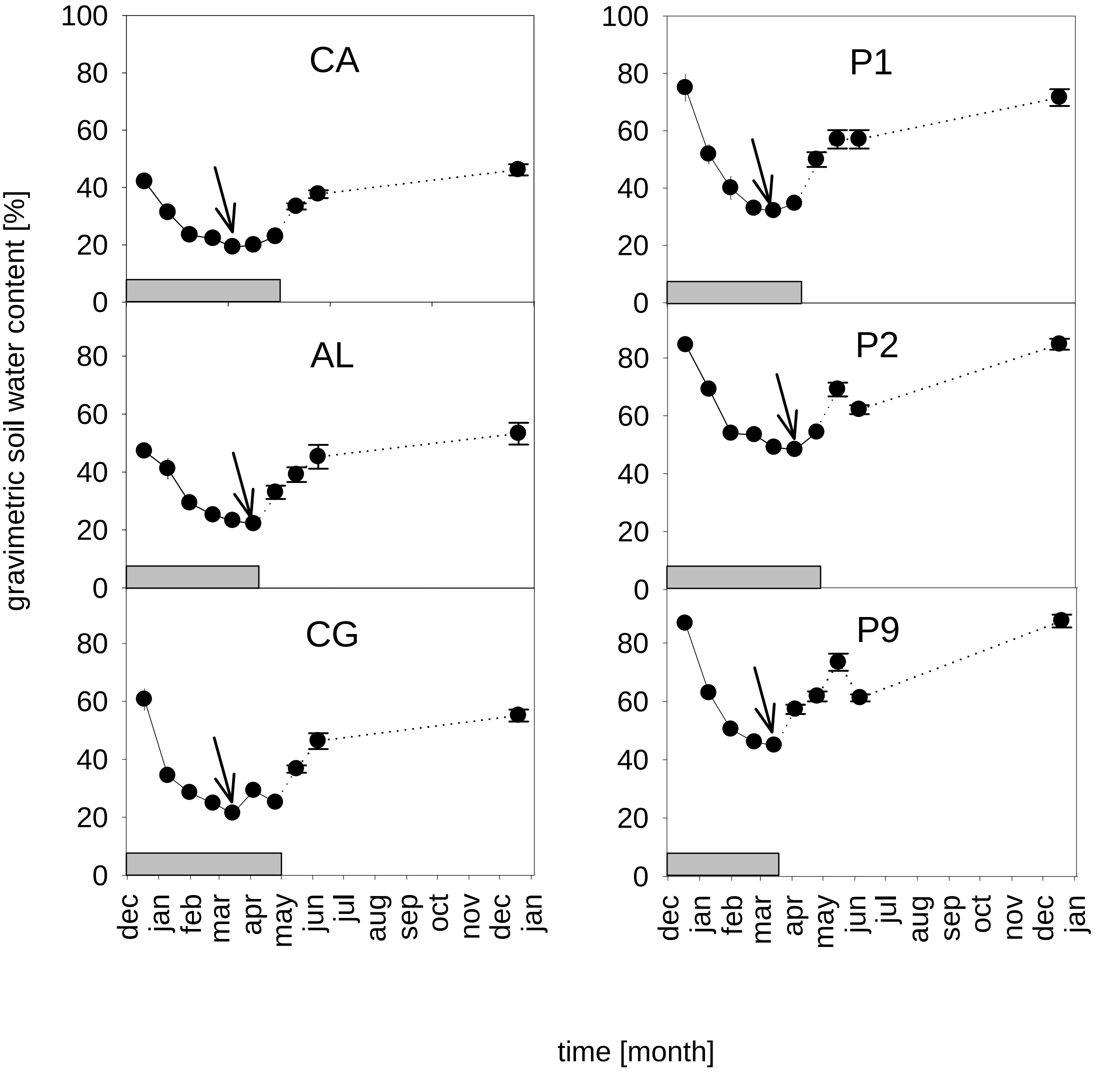

Figure S1: Gravimetric soil water content in 6 forest sites across the isthmus over one year. Each point is the average of 25 systematic plots; standard errors are mostly smaller than the symbols and not visible. Arrows mark the minimum gravimetric soil water content reached in the dry season, and the gray bars mark the dry season – the time from an initial pronounced decrease of soil water content to the last census after which an increase of soil moisture of at least 10% was observed, followed by a further soil moisture increase.

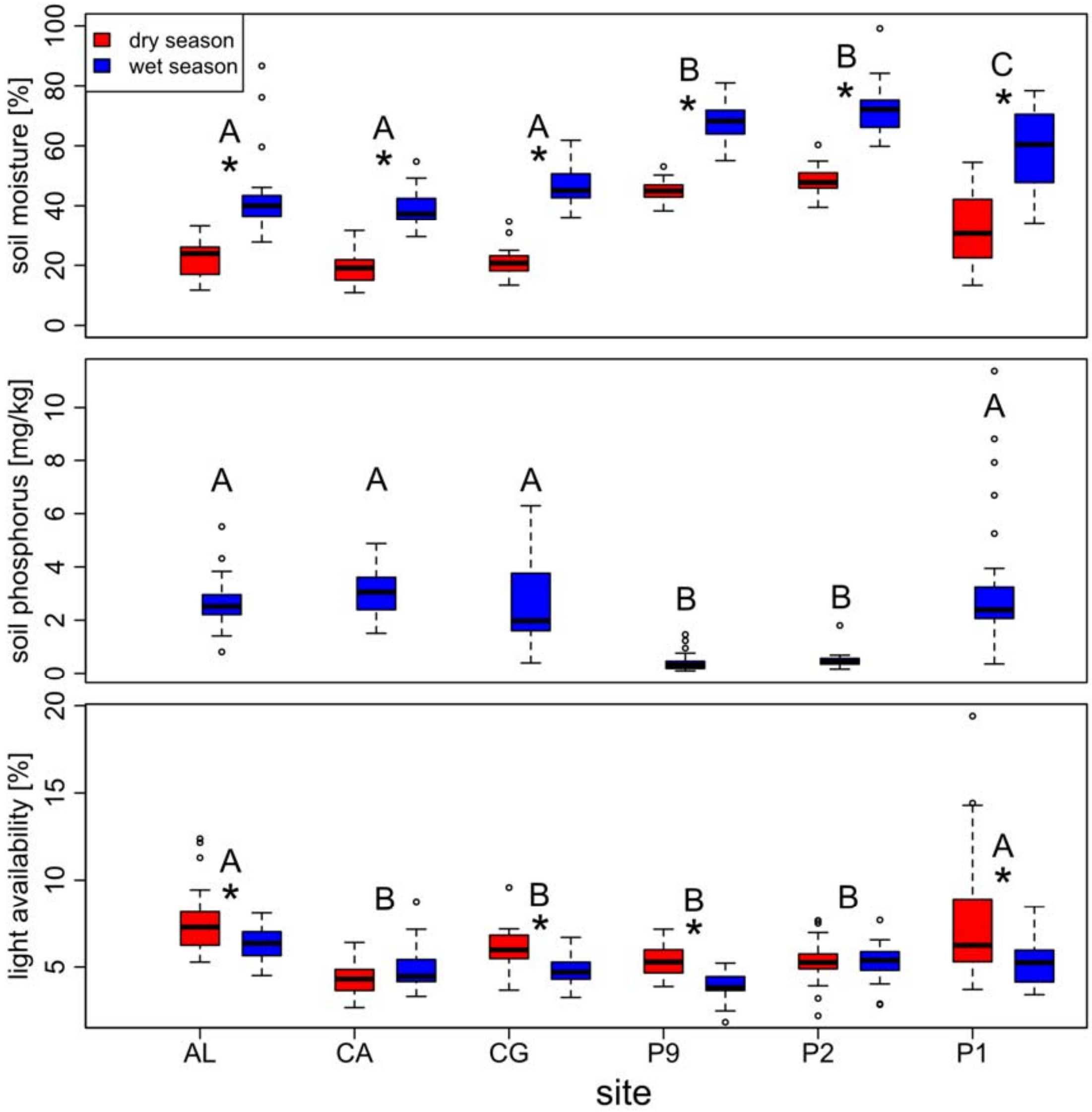

Figure S2: Gravimetric soil moisture, soil phosphorus content and light availability measured as percent canopy openness for the six experimental sites and the two seasons. Soil phosphorus was measured only in the wet season. Stars represent significant differences between the seasons within each site; letters represent significant differences between the sites (Anova with sequential Bonferroni as post-hoc test). Sites are sorted by annual rainfall (see Table S1).

Table S1: Site characteristics of the six 1 ha forest sites across the Isthmus of Panama, sorted by rainfall. Average yearly rainfall is based on BIOCLIM data (Hijmans et al. 2005). Species number per plot refers to woody plants with a diameter at breast height (dbh) > 10 cm. More information on soil types, nutrients, forest composition, forest age and geology of the plots can be found in Pyke et al. (2001), Condit et al. (2013) and Turner and Engelbrecht (2011).

Site name	ID	Utm.x	Utm.y	Altitude	Rainfall	Species number
Albrook	AL	657865.5	992602.5	60	1864	23
Cardenas	CA	657395.0	994229.0	70	1891	15
Cerro Galera	CG	651376.0	987044.0	330	2310	48
P9	P9	638364.8	1013754.2	190	2562	107
P2	P2	613985.4	1030725.4	100	3283	84
P1	P1	614857.0	1031786.4	50	3286	63

Table S2: Study species and their family, drought resistance, origin and deciduousness. Drought resistance is based on irrigation experiments in the forest understory, and origin on occurrence on 122 plots along the rainfall gradient (Engelbrecht et al. 2007). Deciduousness is included from various sources (Croat 1978, Engelbrecht and Kursar 2003); only species specifically described as deciduous are indicated.

Species	Code	Family (-aceae)	Drought resistance	Origin	Deciduous-ness
Beilschmiedia pendula (Sw.) Hemsl.	BEILPE	Laur	0	-1.801	
Brosimum alicastrum Sw.	BROSAL	Mor	86.16	0.572	deciduous
Brosimum utile (Kunth) Pittier	BROSUT	Mor	40.29	-3.564	
Calophyllum longifolium Willd.	CALOLO	Clusi	30	-0.596	
Capparis frondosa Jacq.	CAPPFR	Cappar	91.89	0.157	
Cordia alliodora (Ruiz & Pav.) Oken	CORDAL	Boragin	74.23	1.650	deciduous
Faramea occidentalis (L.) A. Rich.	FARAOC	Rubi	91.89	0.428	
Garcinia intermedia (Pittier) Hammel	GAR2IN	Clusi	98.33	0.008	
Herrania purpurea (Pittier) R.E. Schult.	HERRPU	Malv	53.34	2.349	deciduous
Hybanthus prunifolius (Humb. & Bonpl. ex Roem. & Schult.) Schulze-Menz	HYBAPR	Viol	78.66	6.306	deciduous
Inga sapindoides Willd.	INGASA	Fab	71.82	-0.210	
Lacistema aggregatum (P.J. Bergius) Rusby	LACIAG	Lacistemat	53.33	0.139	
Lacmellea panamensis (Woodson) Markgr.	LACMPA	Apocyn	86.67	-0.563	
Mouriri myrtilloides (Sw.) Poir.	MOURMY	Melastomat	88.92	-0.463	
Myrcia gatunensis Standl.	MYRCGA	Myrt	61.11	0.085	
Posoqueria latifolia (Rudge) Roem. & Schult.	POSOLA	Rubi	89.02	0.860	
Pouteria reticulate (Engl.) Eyma	POUTRE	Sapot	65.38	-0.002	
Psychotria horizontalis Sw.	PSYCHO	Rubi	96.43	0.368	
Pterocarpus rohrii Vahl	PTERRO	Fab	70.83	-0.001	deciduous
Sorocea affinis Hemsl.	SOROAF	Mor	53.33	0.676	
Tabebuia rosea (Bertol.) A. DC.	TAB2RO	Bignoni	48.02	0.627	deciduous
Tetragastris panamensis (Engl.) Kuntze	TET2PA	Burser	70.47	-0.007	
Thevetia ahouai (L.) A. DC.	THEVAH	Apocyn	76.63	0.004	deciduous
Trichilia tuberculata (Triana & Planch.) C. DC.	TRI2TU	Meli	75.86	0.154	
Virola surinamensis (Rol. ex Rottb.) Warb.	VIROSU	Myristic	14.18	-0.993	
Vochysia ferruginea Mart.	VOCHFE	Vochysi	36.69	-0.075	

Table S3: Parameters used to calculate the leaf area of each species, following the formula leaf area = a x (length x width) + b, with length and width the measure of a focal leaf. Species codes follow Table S2. R² denotes the coefficient of determination for the relation of each species, and n is the number of leaves used to determine the parameters a and b.

Species code	a	b	R²	n (Leaves)
BEILPE	0.006	0.3681	0.9802	19
BROSAL	0.0058	-0.9092	0.9635	20
BROSUT	0.0069	0.0164	0.9929	19
CALOLO	0.0073	-2.635	0.9809	20
CAPPFR	0.0062	-1.355	0.9153	20
CORDAL	0.0056	0.1436	0.9783	20
FARAOC	0.0062	0.0359	0.9953	20
GAR2IN	0.0068	-0.649	0.986	19
HERRPU	0.00561	1.5076	0.941	20
HYBAPR	0.0058	0.0364	0.9908	20
INGASA	0.0056	0.7064	0.972	20
LACIAG	0.0061	-0.1669	0.9837	19
LACMPA	0.0055	0.151	0.9805	20
MOURMY	0.0066	0.0466	0.9926	20
MYRCGA	0.0063	0.0291	0.9926	20
POSOLA	0.00668	-0.1336	0.998	40
POUTRE	0.0051	0.3293	0.9868	20
PSYCHO	0.0065	-0.423	0.9932	17
PTERRO	0.0037	3.1377	0.8344	19
SOROAF	0.0061	-0.3617	0.9843	20
TAB2RO	0.0064	-0.2536	0.9894	19
TET2PA	0.00643	0.2199	0.995	20
THEVAH	0.0055	-0.1694	0.9901	20
TRI2TU	0.0062	-0.0785	0.9951	20
VIROSU	0.0063	-0.4286	0.9859	20
VOCHFE	0.00602	0.11924	0.9913	10

Table S4: Spearman rank correlation coefficients (r) among the environmental factors. Statistically significant correlations are in boldface. Values of |r|<0.7 are typically low enough to prevent collinearity from affecting the model estimation (Dormann et al. 2013).

		Soil moisture			Soil	Canopy openness			Mean
		dry season	wet season	annual	phosphorus content	dry season	wet season	annual	annual rainfall
Soil moisture	dry season	1.00							
	wet season	**0.84**	1.00						
	annual	**0.92**	**0.94**	1.00					
Soil phosphorus content		**-0.66**	**-0.52**	**-0.61**	1.00				
Canopy openness	dry season	**-0.16**	-0.02	-0.06	**0.16**	1.00			
	wet season	**-0.16**	-0.10	-0.06	**0.37**	**0.44**	1.00		
	annual	**-0.18**	-0.06	-0.07	**0.28**	**0.87**	**0.81**	1.00	
Mean annual rainfall		**0.56**	**0.62**	**0.61**	**-0.36**	-0.02	**-0.19**	-0.08	1.00

List of manuscripts and specification of own contributions

Manuscript 1

Authors	Julian Gaviria, Bettina M.J. Engelbrecht
Title	Effects of drought, pest pressure and light availability on seedling establishment and growth: Their role for distribution of tree species across a tropical rainfall gradient
Status	Published (*Gaviria, J., and B. M. J. Engelbrecht. 2015. Effects of drought, pest pressure and light availability on seedling establishment and growth: Their role for distribution of tree species across a tropical rainfall gradient. PloS One 10:e0143955.)*
Contributions	The study was designed by JG and BMJE. The experiment was set up and data were collected by JG, with some support of field assistants and colleagues (see acknowledgements). Statistical models were designed and conducted by JG, with advice from BMJE. Results were interpreted and discussed by JG and BMJE. Figures and tables were created by JG, with suggestions by BMJE. JG wrote the first draft of the manuscript, and it was revised and edited for submission by BMJE and JG. JG is acting as communicating author.

Manuscript 2

Authors	Julian Gaviria, Björn Reineking, Ingolf Steffan-Dewenter, Bettina M.J. Engelbrecht
Title	Herbivore defenses of tree species increase across a tropical rainfall gradient
Status	Submitted to PNAS, Nature, Nature Communications, Current Biology, and Ecology Letters (partly with or without review) Currently in preparation for Proceedings of the Royal Society B: Biological Sciences
Contributions	Concept and study design were developed by JG, with advice from BR and BMJE. Field and laboratory work was conducted by JG, with support for species identification and caterpillar collection by a field assistant and local students (see acknowledgements). The data were analyzed by JG with advice from BR and Bernhard Hoiß (see acknowledgements). The

results were discussed by JG, BR, BMJE and ISD. Figures and tables were created by JG, with advice from BR, BMJE and ISD. JG wrote the first draft of the manuscript. The manuscript was revised and rewritten by BMJE and JG, with advice and input from BR and ISD. JG is acting as communicating author.

Manuscript 3

Authors	Julian Gaviria, Bettina M.J. Engelbrecht
Title	Effects of drought, nutrient and light availability on tropical tree distribution: Results from a regional multisite, multispecies seedling transplant experiment
Status	In preparation for publication
Contributions	The study was originally designed by BMJE, and BMJE coordinated and supervised experimental set up and data collection and input by several assistants (see acknowledgements). Ben Turner analyzed soil nutrient samples (see acknowledgements). JG redesigned the models and conducted the analyses of the data. JG and BMJE interpreted and discussed the results. Figures and tables were created by JG, with advice from BMJE. JG wrote the first draft of the manuscript. Revision and rewriting of the manuscript was done by BMJE and JG.

Additional publication not included in this thesis

Hoiss, B., J. **Gaviria**, A. Leingärtner, J. Krauss, and I. Steffan-Dewenter. 2013. Combined effects of climate and management on plant diversity and pollination type in alpine grasslands. Diversity and Distributions 19:386–395.

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(Eidesstattliche) Versicherungen und Erklärungen

(§ 8 S. 2 Nr. 6 PromO)

Hiermit erkläre ich mich damit einverstanden, dass die elektronische Fassung meiner Dissertation unter Wahrung meiner Urheberrechte und des Datenschutzes einer gesonderten Überprüfung hinsichtlich der eigenständigen Anfertigung der Dissertation unterzogen werden kann.

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Hiermit erkläre ich eidesstattlich, dass ich die Dissertation selbständig verfasst und keine anderen als die von mir angegebenen Quellen und Hilfsmittel benutzt habe.

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Bayreuth, 18.11.15

Julian Gaviria