

## Understanding the controls over forest carbon use efficiency on small spatial scales: Effects of forest disturbance and tree diversity

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### ARTICLE INFO

#### Keywords:

Carbon cycle

Tropical trees

Allocation

Forest disturbance

Functional diversity

### ABSTRACT

Tropical forests are characterized by their high photosynthetic activity. Results from pantropical studies show that on average only 30% of the products from photosynthesis are allocated to new biomass compared to on average 50% in temperate forest systems. Why those forests have a low carbon use efficiency (CUE) remains unknown. We present a synthesis of studies from two tropical sites with a similar methodological setup allowing the evaluation of the ecophysiological responses of tree communities to diversity effects and forest disturbance. Here we use xylem sap flux derived gross primary productivity (GPP) that was modeled with eddy covariance data for a seven-year-old planted forest with plots of varying tree species diversity in Panama and an old-growth forest with a distinct disturbance gradient in an Amazonian moist lowland forest. Additionally, net primary productivity (NPP) was calculated from inventory data. We found evidence that GPP is relatively stable with forest disturbance but GPP increases with tree species richness. Lowest CUE with only 35% of GPP allocated to growth was present in undisturbed forest patches. Light forest disturbance had a positive effect on CUE and 62% of GPP was allocated to grow new biomass. Monospecific stands had a higher CUE than diverse tree stands. We conclude that two factors are controlling CUE in tropical forests leading to low CUE. First, forest disturbance leads to a higher allocation of carbon towards growth. Second, a great fraction of GPP is used for processes dedicated to species complementarity. This improves the overall performance of diverse forest stands compared to monocultures.

### 1. Introduction

The quantification of the carbon flow through terrestrial ecosystems has been a major and broad research area over the last decades. Carbon studies concerning climate change have gained much attention and especially studies that quantify how much carbon can be sequestered by forests (e.g. Lewis et al., 2009; Luyssaert et al., 2008; Malhi et al., 2009). The potential of sequestering carbon is partly reflected by the productivity of a forest ecosystem. The productivity of forest ecosystems can be divided into the gross and the net primary production (GPP and NPP) respectively defined as the total amount of carbon fixed by an ecosystem through photosynthesis and as the fraction of GPP allocated to grow new biomass (Clark et al., 2001). The ratio of carbon acquired

from the atmosphere by plants to carbon allocated to growth is broadly defined as the carbon use efficiency ( $GPP/NPP = CUE$ ) (DeLucia et al., 2007). CUE gives a measure of how much carbon is respired by a plant as autotrophic respiration ( $R_a$ ). Consequently, a higher CUE refers to a higher biomass gain per unit carbon assimilated by the plant. CUE is thus a critical physiological parameter describing the carbon storage and allocation processes in forest ecosystems.

It has initially been thought that CUE would be relatively stable among the different forests and that a constant fraction of about half of the GPP is being allocated towards NPP (Waring et al., 1998). Considerable deviation from this relationship has been reported by DeLucia et al. (2007) indicating a strong age relationship, in which declining CUE has been observed in older forests. For example, Vanninen and

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Mäkelä (2005) found that carbon allocation pattern of photosynthetic products towards growth in Scots pine decreased from 0.65 in 13 years old trees to 0.45 in over 200 years old trees. In addition to tree age, CUE changes with environmental conditions e.g. with increasing temperatures a decrease in CUE can be observed (Bradford and Crowther, 2013). For example, a considerable deviation of CUE from the 0.5 values has been found from tropical old-growth forests indicating that tropical forests are at the lower end of observed CUE (Chambers et al., 2004; Doughty et al., 2018; Fernández-Martínez et al., 2014; Malhi, 2012). In tropical old-growth forests, only 30% of the assimilated carbon is allocated to growth. The other 70% of the assimilated carbon is dedicated to metabolic processes such as maintenance or growth respiration and is respired back to the atmosphere. Hence, one driving question is, why are old-growth forest trees in the Amazon so inefficient in converting assimilated photosynthetic products into new plant biomass (see e.g. Malhi, 2012)? Explanations for low CUE range from low nutrient availability enhancing the production of root exudates, unaccounted processes within NPP (Goulden et al., 2011; Vicca et al., 2012) or that water availability and high temperature are influencing autotrophic and heterotrophic respiration (Chambers et al., 2004; Kato and Tang, 2008). It is a general assumption, that lower CUE might be explained by higher temperatures in tropical climates and thus a higher stimulation of plant respiratory activities than stimulation of photosynthesis (Dewar et al., 1999; Hartley et al., 2006). Forests in lower latitudes should be characterized by a higher respiratory carbon loss and thus lower CUE than temperate forests or even boreal forests. In contrary to this assumption, CUE decreases from temperate forest towards colder climates in boreal forests (DeLucia et al., 2007; Fernández-Martínez et al., 2014). On smaller scales on in a tropical biome, Doughty et al. (2018) confirmed that CUE actually tended to decrease with lower mean temperatures along a rainfall and fertility gradient in the Amazon forest. The CUE was strongest (positive) correlated to fertility and increased with biomass residence time (Doughty et al., 2018). Most studies are comparing CUE as a property changing along a large gradient of parameters and even with spatial distribution.

Concluding from those observation, there are two major gradients in CUE. The first one is a forest-age-gradient meaning that younger forests – or, in other words, forests recovering from forest disturbance – more photosynthetic products are allocated towards growth. The second gradient is a latitudinal gradient coinciding with a significant increase in tree species diversity in lower latitudes towards the equator.

Methodical limitations prevented testing those two gradients up to now, as there were only two main approaches to estimate CUE of forest ecosystems both relying on GPP and NPP measurements. The first approach, the so-called “top-down” approach, is based on measurements of the net ecosystem carbon fluxes between the forest ecosystem and the atmosphere. GPP is accordingly the net ecosystem exchange (-NEE) of carbon plus all ecosystem respiratory processes. NPP is assumed to be -NEE plus all heterotrophic ecosystem respiration ( $R_h$ ). This approach gives good information on an ecosystem scale, but often lacks sufficient spatial resolution to actually relate the finding to specific ecosystem properties on the individual tree level (Tang et al., 2006). The second approach, the “bottom-up” approach assesses different autotrophic respiratory processes ( $R_a$ ) on an individual tree level by measuring  $\text{CO}_2$  effluxes from leaves, woody tissue and roots. The bottom-up approach is bonded to upscaling artifacts and thus require large sample sizes or at least strong allometric equations.

A third and relatively new approach to get a higher spatial resolution has been proposed by Tang et al. (2006) and combines bottom-up and top-down data. This method allows the derivation of high-resolution GPP of single trees and forest plots by combining data from xylem sap flux measurements and GPP measurements from eddy covariance ecosystem measurement (see Tatarinov et al., 2017 for detailed protocol). The method assumes that xylem sap flux measurements serve to directly estimate photosynthesis by using the strong correlation of water use efficiency (WUE) and vapor pressure deficit ( $D$ ) (Tang et al.,

2006; Tatarinov et al., 2017).  $D$  is accordingly one of the main factors controlling WUE calculated out of canopy transpiration ( $E_c$ ) from xylem sap flux measurements and GPP from the eddy flux measurements (Tatarinov et al., 2017). In the following, this relationship can be used to model GPP of certain trees or plots that are actually contributing to the initial canopy transpiration. Accordingly, this approach accounts for environmental heterogeneity on fine spatial scales in the canopy boundary layer.

With this new approach being able to derived GPP data on small spatial scales, we wanted to test if it could confirm the two gradients mentioned above. The goal of this study was to assess if CUE is significantly different between a mature, old-growth forest with different degrees of disturbance and if CUE decreases with increasing species richness in a planted forest. Therefore, we hypothesized that differences in CUE can be explained on small spatial scales by forest disturbance, hence forest age. Further, we assumed that species diversity plays a role in controlling CUE and that it decreases with increasing species diversity. We combined xylem sap flux and eddy covariance datasets from two different tropical study sites to derive GPP values for small forest monitoring plots characterized by different forest disturbance intensity and species diversity.

## 2. Material and methods

### 2.1. Study sites

The data for this study were obtained from one site in the Central Amazon and one in Central Panama. Both study sites can be classified as tropical moist lowland forests (Kunert, 2016). Their annual precipitation is about 2550 and 2350 mm with mean temperatures of 25.8 °C and 26.2 °C for the site in the Amazon and in Panama, respectively (Kunert et al., 2017, 2010).

The Amazon dataset comes from a species-rich old growth forest characterized by a distinct disturbance gradient caused by a forest edge which was created about 25 years ago (Kunert et al., 2015b). The Panama dataset comes from an experimental planted forest designed to test for functional biodiversity (Scherer-Lorenzen et al., 2005). The main difference between the two sites was the forest age, with the trees in Panama being only seven years old at the time when the measurements were conducted in 2007/2008.

The old growth forest in the Central Amazon is located northwest of the city of Manaus in the TACAPE monitoring plots (Tree Assimilation Carbon Allocation Physiology Experiment, 02°38'22.54"S 60°09'51.34"W). The TACAPE monitoring plots are part of the Experimental Station for Tropical Silviculture (ZF-2) of the Brazilian National Institute of Amazonian Research (INPA). The tropical moist forest in the area is considered to be a hyperdiverse forest with more than 250 different species per hectare (only accounting for tree larger than 10 cm in diameter). Due to this high diversity all estimates of tree properties are thus not given on species level. The study site is along the access road, which is ~6 m wide with an approximately 9 m wide buffer zone on each side to avoid access blockage by falling trees. The road was built in the 1980s when the field station was established. A 250 m-long transect was established perpendicular to the road and extending into the forest. A set of four plots (20 × 50 m) were established at distances of 10 m, 50 m, 100 m, and 250 m from the forest edge. Field measurements were conducted from January 2013 to December 2013 (Kunert et al., 2015b).

The experimental planted forest is located in Sardinilla, Central Panama (9°19' N, 79°38' W), which is approximately 50 km north of Panama City. It was established in 2001 with six different tree species planted as a monoculture, 3-species mixtures and 6-species mixtures. Trees were planted in 3 × 3 m spacings (Scherer-Lorenzen et al., 2005). One species *Cordia alliodora* (Ruiz & Pavon) Oken) did not establish, probably due to missing mycorrhiza at the beginning of the experiment and the fact that the site was used as pasture site for about 50 years

(Kunert and Mercado Cardenas, 2015). However, the other five species *Anacardium excelsum* ((Bert. & Balb. ex Kunth) Skeels), *Cedrela odorata* (L.), *Luehea seemannii* (Triana & Planch), *Hura crepitans* (L.) and *Tabebuia rosea* ((Bertol.) DC.) developed well over the years reaching medium tree heights of up to 13 m at the time of the present experiment (Kunert et al., 2012). The species were chosen out of the variety relative growth rates on Barro Colorado Island (relative growth rates of species on Barro Colorado Island: *Luehea seemannii* 9.1%; *Cordia alliodora* 7.0%; *Anacardium excelsum* 5.9%; *Hura crepitans* 4.0%; *Tabebuia rosea* 3.4%; *Cedrela odorata* 2.3%; for more details see Scherer-Lorenzen et al. (2005)). The failed establishment of one species resulted in the development of five monocultural, three 2-species mixtures, three 3-species mixtures and four 5-species mixtures plots ( $n = 15$  plots). The field measurements used for this analysis were conducted during June 2007 and July 2008 when trees were between 6 and 7 years old (for details see Kunert et al., 2012; Wolf et al., 2011). A closed canopy had been established already at the time of study (compare crown projection areas in Kunert et al., 2012). Trees for the sap flux measurements were chosen in subplots with similar soil properties and water table (compare Kunert et al., 2012).

## 2.2. Xylem sap flux derived gross primary productivity

At both sites, the same set up of Granier type thermal dissipation probes was used to measure xylem sap flux densities ( $J_s$ ,  $\text{g cm}^{-2} \text{d}^{-1}$ ). We used the standard Granier calibration equation (Granier, 1985) to calculate xylem sap flux density from the thermocouple readings. We assumed a good fit of this equation, as Granier et al. (1996) tested the calibration equation on trees in the same biome and on trees from the same families and even tree species. All our investigated tree species are diffuse porous trees (Aparecido et al., 2015), which seem to be in good agreement with the standard calibration equation (Bush et al., 2010). Further, Kunert et al. (2015a) and Kunert et al. (2013) already showed that a large variety of arborescent plants from tropical moist lowland forest are in good agreement with the Granier equation. Xylem sap flux measurements were conducted at four individual trees of each given species within the mixtures. We used the species and mixture specific sap wood and diameter at breast height (DBH, cm) relationships in the experimental plantation developed by Kunert et al. (2012). Briefly, the equation was derived from profile measurements at different xylem depths to assess changes of xylem sap flux densities across the stem cross section (for details see Kunert et al., 2012). Further, we applied the allometric equation to calculate the conductive xylem area for the Amazonian forest presented by Kunert et al. (2017) to estimate the conductive sap wood area ( $A$ ,  $\text{cm}^2$ ) for the trees equipped with xylem sap flux sensors. Kunert et al. (2017) showed that the use of the allometric equation caused only minor and non-significant errors in the upscaling process. We calculated tree water use rate ( $Q$ ,  $\text{kg d}^{-1}$ ) by multiplying mean species and mixture specific  $J_s$  and  $A$  of a given tree. Canopy transpiration ( $E_C$ ,  $\text{mm d}^{-1}$ ) is given as the sum of water use rates of all trees for a given plot. We used total daily gross primary production ( $\text{GPP}_{\text{eddy}}$ ) from eddy covariance flux measurements (for details on the eddy flux measurements and analysis see Kunert et al., 2017; Wolf et al., 2011) and  $E_C$  to estimate daily or monthly mean water use efficiency (WUE,  $\text{mg C g}^{-1} \text{H}_2\text{O}$ ) per plot, which is defined as  $\text{GPP}_{\text{eddy}}/E_C$  (Tatarinov et al., 2017). The relationship between WUE and vapor pressure deficit ( $D$ ) was described by an exponential decay function (Tang et al., 2006):

$$\text{WUE} = a_0 + a_1 e^{-a_2 D} \quad (1)$$

where the parameter  $a_0$ ,  $a_1$  and  $a_2$  are fitted coefficients (Figs. 1 and 2). WUE reaches its minimum at  $a_0$  when  $D$  approaches  $\infty$ . Hence, daily  $\text{GPP}_m$  values were derived as (Tang et al., 2006):

$$\text{GPP}_m = E_C(a_0 + a_1 e^{-a_2 D}) \quad (2)$$

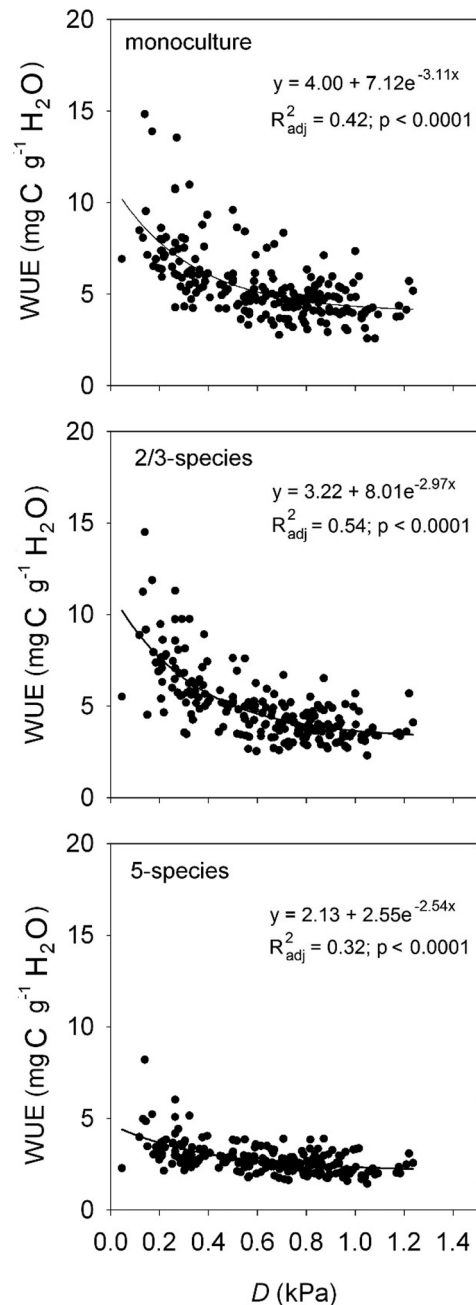
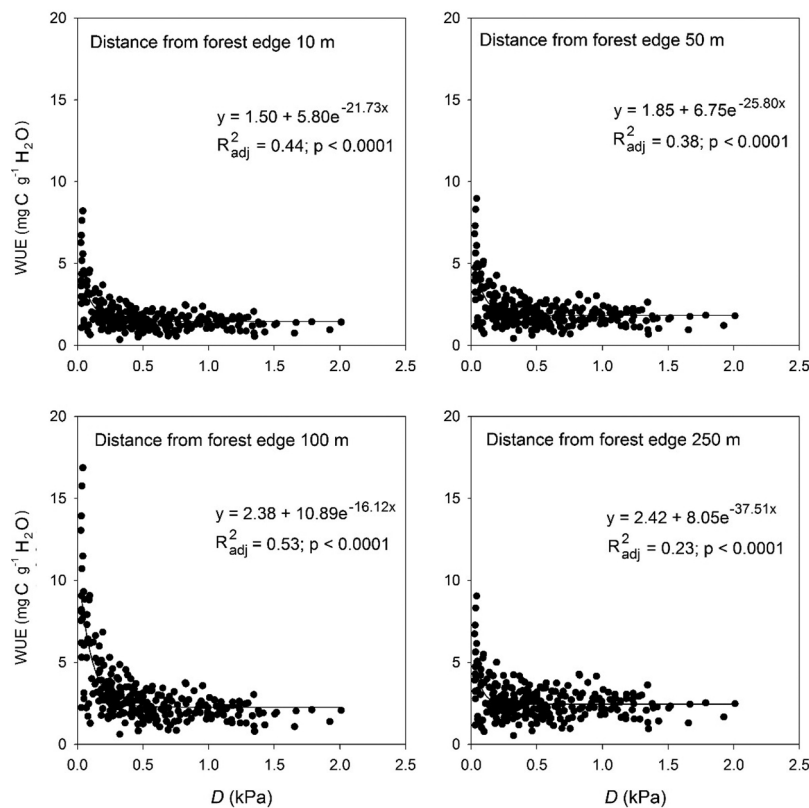


Fig. 1. Response curves of daily water use efficiency (WUE) to daily mean vapor pressure deficit ( $D$ ) in the different subplots in the Amazon. Fitted curves are exponential decay functions.

## 2.3. Estimation of NPP and CUE

The net primary productivity (NPP) was calculated based on the annual increment of the trees. In Panama, the increment of the studied trees was measured on a bi-monthly basis with measuring tape during the study period. In the Central Amazon, lab-made metal increment bands were used and increment was measured at the beginning and at the end of the study. At both sites, we had specific details on the tree biomass. For the Amazon, we used the site-specific allometric equation for calculating tree biomass and carbon stock (Chambers et al., 2001). In Panama, tree species- and site-specific allometric equations were used (Oelmann et al., 2010). CUE was then calculated as the ratio of ecosystem-scale GPP (from eddy covariance fluxes) and NPP from site-specific allometric relationships of the trees.



**Fig. 2.** Water use efficiency (WUE) response curves to daily mean vapor pressure deficit ( $D$ ) in the different mixtures in the experimental tree plantation in Panama. Fitted curves are exponential decay functions.

#### 2.4. Statistical analysis

An analysis of variance (One-way ANOVA followed by a Brown-Forsythe test) was performed to test for homogeneity of variables across the different treatments. A Monte Carlo analysis was performed to calculate the uncertainty around the final estimate of  $GPP_m$ . We used the estimated standard errors of the coefficients of the different exponential decay functions (Eq. (1) and Figs. 1 and 2) and assumed normal distributions for the averages. Random numbers were sampled from the distribution of  $E_C$  and  $D$  and then summed up to produce an estimate of the  $GPP_m$ . We repeated the procedure 1000 times and performed a sensitivity analysis to test the effect of correlation between variables. We used the standard deviation of the distribution of the averages (i.e. the standard error of the mean) as an estimate of the uncertainty of each treatment. Statistical analyses were carried out using the open source statistical program JASP 0.8.2 (The JASP Team, 2017. Computer software, <https://jasp-stats.org/>). Curve fittings for exponential decay functions were done with Sigmaplot (version 10.0, SPSS Inc., Chicago, IL, USA). The Monte Carlo was performed in an excel matrix and the covariance analysis with Analysis ToolPak (Version 2016, Microsoft Corporation, USA).

### 3. Results

#### 3.1. Meteorological conditions

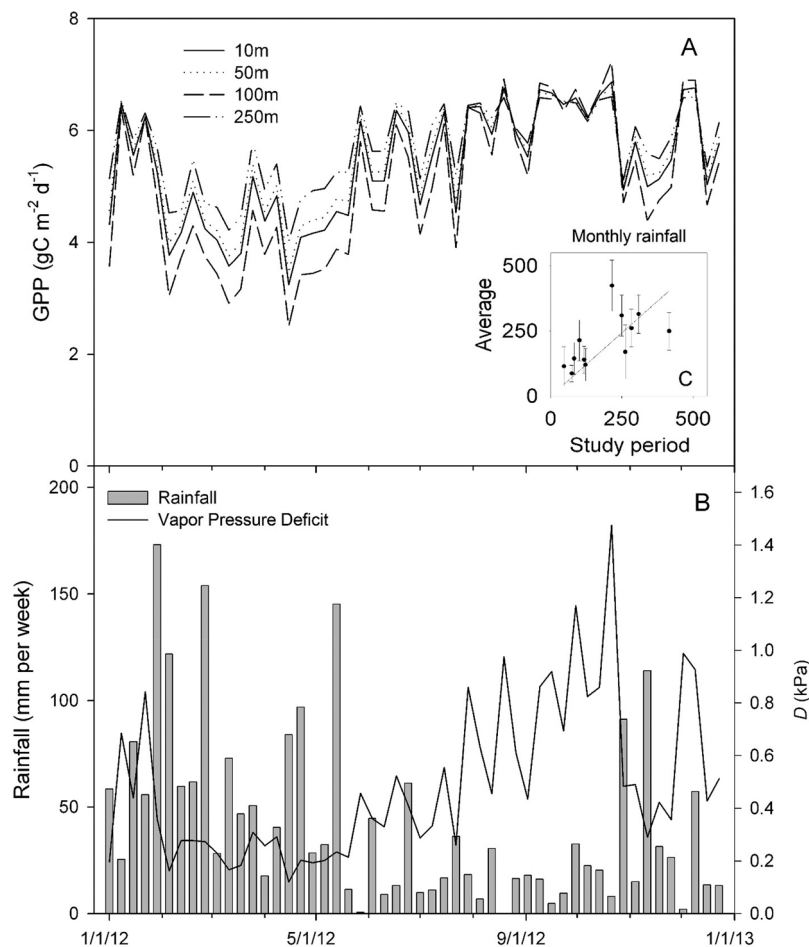
During the study period, the meteorological conditions did not show any significant anomalies for both study sites (Fig. 3c and 4f). At both sites, the relationship between monthly averages over the last 12 years and monthly rainfall during the study period was close to the 1:1 line. In the year 2013 the study site in the Amazon received 2300 mm of rainfall, what is about 10% less rain than the long-term average in the area recorded by a long-term climate station. However, the TACAPE

monitoring plots are about 16 km away from the long-term climate station and receive on average 11% less rainfall per year due to a natural rainfall gradient. During the study period, less than 100 mm of rainfall fell in the three months June, August and September (Fig. 3b). At the long-term climate station, monthly rainfall drops below 100 mm only in August. However, up to four months with less than 100 mm of rainfall per month is nothing uncommon. We did not observe any meteorological abnormalities during the study period at the site in Panama (Fig. 4d). The plantation received 2260 mm of rainfall during the 12-month periods and the dry season length of approximately 116 days was within the average dry season length and its standard deviation of  $131 \pm 19$  days.

#### 3.2. Gross primary productivity in relation to species richness and forest disturbance

High GPP was observed in the undisturbed old-growth forest in the Amazon (Table 1). The undisturbed old forest assimilated  $31.1 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$ , slightly more than in the disturbed subplots closer to the forest edge, however, this difference was not significant ( $p = 0.151$ , ANOVA). In the young planted forest in Panama, GPP ranged from 2.3 to  $10.1 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$  in the monospecific stands with a mean and standard deviation of  $4.9 \pm 3.1 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$  (Table 2). This was on average lower than in the mixed species plots. The 3-species mixtures assimilated  $8.1 \pm 1.9 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$  and the 5-species mixtures up to  $9.3 \pm 0.4 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$ . The 2-species mixtures were lower in their GPP and only assimilated  $2.8 \pm 1.0 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$ .

Annual pattern of GPP followed similar courses in the Amazon old-growth forest and in the disturbed subplots (Fig. 3). Dry season GPP was almost equal in all plots, but diverged during the wet months. The undisturbed plot (i.e., 250 m inside the forest) showed the highest GPP during this time and the difference possibly explains the overall higher annual GPP of this plot. In the young planted forest in Panama, the



**Fig. 3.** Annual course of A) gross primary production (GPP) in the different subplots in the Amazon and B) weekly rainfall data and weekly mean vapor pressure deficit ( $D$ ). C) mean monthly rainfall (mm, error bars indicated the standard deviation) observed over the last 12 years at the nearest long term climate station at the LBA tower versus the monthly rainfall during the study period (the slope is 0.97).

annual patterns in GPP was as diverse as its species diversity (Fig. 4). Planted in monoculture, *Luehea seemanii* had the highest GPP with just one short drop at the end of the dry season coinciding with the species short leaf shedding period. Despite a considerable scatter, GPP of the other species was following a very similar course during the wet season and diverged during the dry season depending on the leaf phenology of a given species. Dry season GPP tended to be lower than in the wet season for all the species growing in monoculture. The GPP in mixture plots was characterized by an averaging-effect and followed all similar courses over the year depending on the species they contain. Most pronounced was this averaging effect in the 5-species mixtures showing all the same pattern throughout the year. GPP was highest in those plots during the wet season. Despite some drop in GPP in the 5-species mixtures, they maintained a high GPP even throughout the dry season. All 2-species mixtures had lower GPP during the dry season, mainly due to the proportion of deciduous species in those plots. GPP was lowest in the 2-species mixtures, due to the reduced number of individuals in those plots (since *Cordia alliodora* did not establish). Exceptional was the higher GPP in the 3-species mixtures during the dry season compared to the wet season GPP (Fig. 4b).

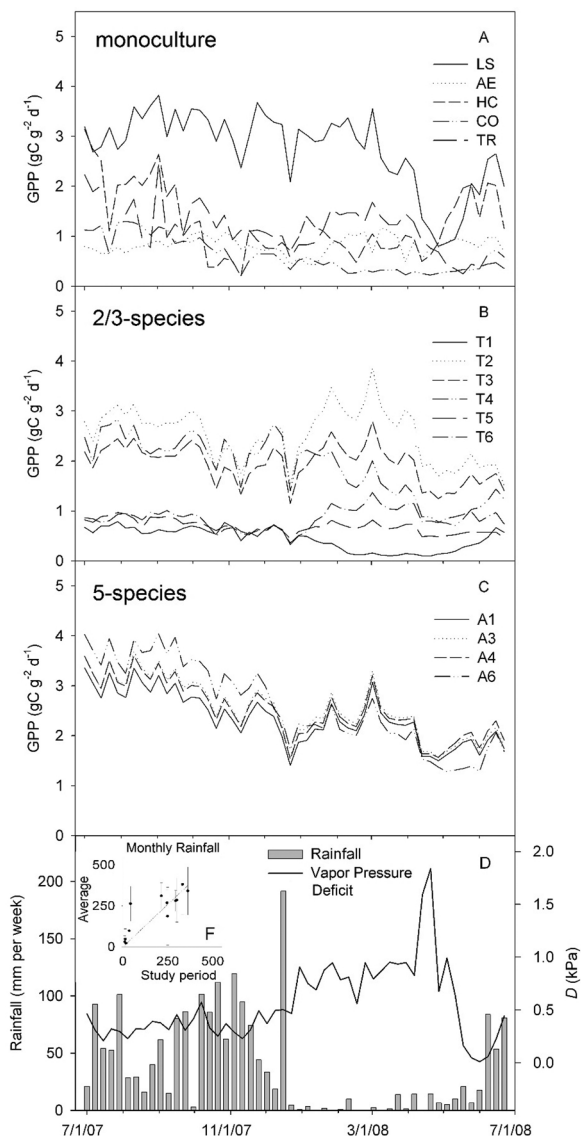
### 3.3. Sources of error in the GPP estimates

The standard error of estimates calculated for the exponential decay functions describing the relationship between WUE and  $D$  were in general low. In the Amazonian *terra firme* forest, the standard error of estimate of the function for the plot 10 m from the forest edge was 0.83

and 0.93 at 50 m from the forest edge. The standard error of estimate was highest with 1.61 for the function of plot the at 100 m from the forest edge. The standard error of estimate for the function for the forest plot at 250 m was lying at 1.05. The uncertainty of final GPP estimates was accordingly 1.4, 1.4, 1.6 and 1.2% for the plot at 10 m, 50 m, 100 m and 250 m, respectively. In the planted forests in Panama, the largest standard error of estimates was present in function for the monocultures and was 1.44. The standard error of estimates for the 2/3-species mixtures was with 1.29 lower and was only 0.58 for the 5-species mixtures. This resulted in an uncertainty of the final GPP values of only 2.7, 2.7 and 2.1% for the monocultures, 2/3-species and 5-species mixtures, respectively.

### 3.4. Net primary productivity in relation to species richness and forest disturbance

Large differences in NPP were observed along the disturbance gradient in the Central Amazon plots. The plots in the forest interior with only intermediate to low disturbance pattern grew  $9.31$  and  $11.0$  Mg C ha<sup>-1</sup> yr<sup>-1</sup> (at 100 m and 250 m distance to the forest edge, respectively). The two plots at the forest edge were characterized by a high increment and a NPP of  $14.5$  and  $18.6$  Mg C ha<sup>-1</sup> yr<sup>-1</sup> at 10 m from the edge and at 50 m from the edge, respectively (see Table 1). In the Panama study site, NPP was highest in the 3-species plots with trees accumulating  $5.0 \pm 3.4$  Mg C ha<sup>-1</sup> yr<sup>-1</sup>. Lowest NPP was observed in the 2-species mixtures, where  $2.1 \pm 0.9$  Mg C ha<sup>-1</sup> was allocated to growth. For the monocultures and the 5-species mixtures, annual NPP



**Fig. 4.** Annual course of gross primary production (GPP) in the different plots in Sardinilla, Panama. A) weekly GPP in the five different monocultures, B) in the 2/3-species mixtures and C) in the 5-species mixtures. D) weekly rainfall and mean weekly vapor pressure deficit (*D*) is shown in the lower panel. F) mean monthly rainfall (mm, error bars indicated the standard deviation) observed over the last 12 years at the nearest long term climate station in Salamanca versus the monthly rainfall during the study period (the slope is 0.87).

**Table 1**

Summary of the data set from the Central Amazon. Values of xylem sap flux derived annual gross primary productivity ( $GPP_m$ ), net primary productivity (NPP) and carbon use efficiency (CUE) for different tree species mixtures. CUE is given as the ratio between GPP and NPP.

Distance from forest edge	Disturbance intensity	$GPP_m$	NPP	CUE
		Mg C ha <sup>-1</sup>	Mg C ha <sup>-1</sup>	
10	High	29.2	14.5	0.50
50	High	29.8	18.6	0.62
100	Intermediate	27.3	9.31	0.34
250	Low	31.1	11.0	0.35

**Table 2**

Summary of the data set from the planted forest in Panama. Values of xylem sap flux derived annual gross primary productivity ( $GPP_m$ ), net primary productivity (NPP) and carbon use efficiency (CUE) for different tree species mixtures. CUE is given as the ratio between GPP and NPP.

Plot No	Mixture	Species <sup>a</sup>	$GPP_m$ Mg C ha <sup>-1</sup>	NPP Mg C ha <sup>-1</sup>	CUE
LS1	monoculture	LS	10.1	8.4	0.83
AE2	monoculture	AE	3.0	2.2	0.73
HC1	monoculture	HC	4.7	3.7	0.78
CO2	monoculture	CO	2.3	2.0	0.86
TR1	monoculture	TR	4.4	3.1	0.71
T1	2-species	CO;HC	1.6	1.1	0.67
T2	3-species	TR;AE;LS	10.3	8.8	0.85
T3	3-species	LS;CO;AE	7.2	4.2	0.58
T4	3-species	LS;CO;HC	6.9	2.1	0.31
T5	2-species	HC;TR	3.5	2.5	0.71
T6	2-species	TR;AE	3.1	2.7	0.86
A1	5-species	LS;AE;HC;CO;TR	8.7	2.4	0.28
A3	5-species	LS;AE;HC;CO;TR	9.4	5.2	0.55
A4	5-species	LS;AE;HC;CO;TR	9.3	5.1	0.55
A5	5-species	LS;AE;HC;CO;TR	9.6	3.0	0.31

<sup>a</sup> LS: *Luehea seemannii*; AE: *Anacardium excelsum*; HC: *Hura crepitans*; CO: *Cedrela odorata*; TR: *Tabebuia rosea*.

was almost the same with  $3.9 \pm 2.6$  Mg C ha<sup>-1</sup> yr<sup>-1</sup> and  $3.9 \pm 1.4$  Mg C ha<sup>-1</sup> yr<sup>-1</sup>, respectively.

### 3.5. Carbon use efficiency in relation to species richness and forest disturbance

In the Amazonian forest, a clear pattern was present for the CUE in relation to forest disturbance. Edge effects on CUE disappeared at a 100 m distance from the edge during the wet season (January to May and October to December); accordingly, CUE was almost equally low at 100 m and 250 m (0.34 and 0.35). During the dry season, edge effects penetrated deeper into the forest, increasing the CUE of the trees at 100 m distance from the forest edge to 0.39 (Fig. 5c). The plots next to the forest disturbance were characterized by the highest CUE, independently of seasonality (0.50 at 10 m from the forest edge and 0.62 at 50 m from the forest edge) (see Table 1).

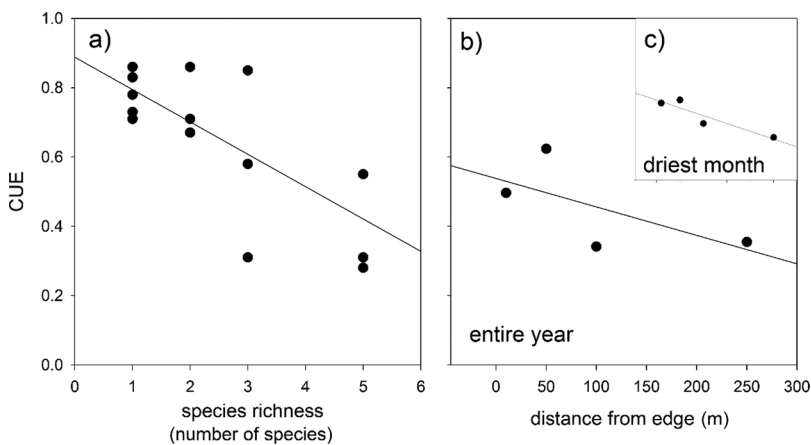
In the planted forest in Panama, there was a clear pattern of CUE across the different species mixtures. CUE was on average highest in the monocultures ( $0.78 \pm 0.06$ ) followed by the 2-species mixtures ( $0.75 \pm 0.10$ ). CUE in the 3-species mixtures was on average  $0.58 \pm 0.27$ . Accordingly, the 5-species plots with the highest species number were the least efficient plots and the ratio between NPP to GPP was only  $0.42 \pm 0.15$ .

## 4. Discussion

In the current study, we could confirm that an undisturbed tropical old-growth forest was characterized by a low CUE; however, CUE changes due to forest disturbance on small spatial scales. Despite lower CUE, GPP was higher in the undisturbed forest mostly driven by higher wet season productivity. Overall, species richness had a positive effect on GPP in the planted forest. GPP in mixtures exceed the GPP in most monospecific stands. This was driven by higher dry season productivity of the mixed species stands compared to monocultures. However, the higher overall gross gain in carbon in the mixtures could not be equally allocated into a higher net productivity and resulted in a lower CUE in mixed species plots.

### 4.1. Forest disturbance increases carbon use efficiency

We observed the lowest CUE in trees growing at least 100 m away from the forest disturbance where edge effects have been found to be



**Fig. 5.** Linear relationship between carbon use efficiency (CUE) and controlling variables such as a) tree species richness ( $R^2 = 0.56$ ;  $p < 0.001$ ) and b) forest disturbance indicated as edge effect ( $R^2 = 0.44$ ;  $p = 0.334$ ). Note: a) and b) indicate annual values, whereas c) indicates the linear relationship between CUE and forest disturbance ( $R^2 = 0.86$ ;  $p < 0.05$ ) in the driest month during the study period (September 2013), when edge effects penetrated deeper into the forest interior.

minimal (Kunert et al., 2015b). Studies presenting low CUE from tropical forests usually consider the forests to have been in a ‘close-to-stable-state’ over a longer period and were not affected by any major disturbance (Chambers et al., 2004; Malhi, 2012; Malhi et al., 2009). According to a global assessment of the variation of CUE among different forest ecosystems, DeLucia et al. (2007) found that CUE decreases with forest age and tropical forests were considered to be the oldest forests.

In general, a common explanation why tropical trees – and especially old trees – are characterized by a lower CUE is that they need to invest more energy into defense compounds against herbivory and infestation (Coley and Barone, 1996). Further, we can consider that tropical forest are very old and at a low disturbance rate over a long time period or are at least containing a high fraction of ancient tree individuals (Chambers et al., 1998). This means, that trees are on average relatively old in an undisturbed old-growth forest. As tree growth tends to slow with tree age and tree size (Luyssaert et al., 2008; Peñuelas, 2005), we can assume that the average tree in an old-growth forest is probably characterized by a rather slow growth rate (e.g. da Silva et al. (2002) give a mean annual diameter increment of only 1.64 mm per year at our study site). On the other side, younger tropical trees in forests recovering from disturbance or faster-growing tree species have higher CUE (Doughty et al., 2018) and early successional species more abundant at the forest edge than in the forest interior (Kunert et al., 2015b). Accordingly, faster growth is more important for younger trees in terms of competition than defense compounds. Unfortunately, there is only limited information available on the quantities of carbon allocated to plant defense compounds (Schultz et al., 2013). However, the allocation to defense compounds explains probably only a part of the lower CUE and deviation from the 0.5 value observed in temperate forests. In other words, if tropical forests would have the same CUE as temperate forests, roughly 20% more assimilated carbon would be allocated to growth accounting for more or less  $6 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$ . Even if the energetic carbon invested in the synthesis of those products is very high, such a high amount of carbon cannot disappear in the production process of secondary defense compounds.

Another explanation could be the idling hypotheses: tropical old-growth forest trees have a lot of excess carbon available and maintain a high rate of idle respiration so that they can respond quickly after disturbance or sudden availability of additional resources (Chambers and Silver, 2004; Doughty et al., 2018). The hypothesis is based on the assumption that plants need to respire carbon to maintain their living cells or to grow new tissue (maintenance-growth-paradigm, Amthor, 2000). Besides beneficial maintenance-growth respiration, some respiration cannot be attributed to neither maintenance nor growth and thus occurs without any benefit to the plant. In this pathway, photosynthetic products are wasted by the plant (maintenance-growth-wastage-paradigm, Amthor, 2000) for example by hydrolysis of ATP

explaining a higher respiration or a lower yield rate of plants (Tanaka, 1972). Especially older plant tissue is thought to use such alternative pathways (Beever, 1970). Considering that tropical old-growth forest are very old (Chambers et al., 1998) or contain a fraction of old individuals a higher fraction of GPP might be lost in such overflow pathways. Another possibility is that a high fraction of GPP is allocated to plant parts (e.g. storage, belowground processes) hard to identify with our approach.

In our study, sap-flow derived GPP was not significantly different in the disturbed plots at the forest edge compared to the undisturbed forest interior (Table 2). Forest disturbance induced a clear and significant effect on allocation processes and a higher fraction of carbon was allocated to growth. NPP almost doubled, comparing between plots at 50 and 100 m distance from the forest edge. Forest disturbance in the form of edge effect had thus a clear effect on the carbon allocation.

One study found that anthropogenic disturbance of forests by forest management had a significant effect on forest CUE in general (Fernández-Martínez et al., 2014). Forest disturbance caused by timber harvesting reduced biomass stocks and was lowering the competition for light, water, and nutrients (Fernández-Martínez et al., 2014). Hence, forest disturbance can be seen as a key for increasing NPP and might be the main explaining factor for decreasing CUE with forest age.

#### 4.2. Carbon a limited resource?

In our data from Panama, the on average highest GPP during the observed year was in the plots with the highest species richness (Table 1). Those plots were also characterized by the on average faster growth rates and thus highest NPP. The positive species mixture effect on the net primary production has been attributed to be clearly dependent on complementarity effects on water use characteristics (Kunert et al., 2012) and soil water uptake pattern (Schwendenmann et al., 2015). Further, mixtures can perform photosynthesis for a longer time when soil water is limited (Kunert and Mercado Cardenas, 2015). Nevertheless, higher GPP and NPP does not mean that the trees in the plots with the highest species richness did allocate GPP to NPP most efficiently. In general, it is assumed that higher productivity of diverse plant communities, compared to single-species communities, comes from a high resource use efficiency (Kelty, 2006). This is induced by a reduction in competition for light, water and nutrients by complementarity and niche facilitation (Forrester and Albrecht, 2014; Sapijanskas et al., 2014, 2012). Our results suggest that this does not apply for carbon as a resource. Trees probably assimilate more carbon than they are able to allocate to growth. Accordingly this over production needs to be disposed in an overflow pathway via wastage respiration (Amthor, 2000). Mixed species stands might be at the maximum capacity of the species for net biomass production and might be simply not able to allocate more carbon towards growth. Another speculative

explanation could be that the excess carbon is used to access limited nutrients and is leading to observed higher growth rates of species-rich forests. A fraction of the carbohydrates might be distributed to mycorrhiza enabling trees in mixtures to retain nutrients (Read, 1991).

Tropical trees in old-growth forests, but even in young forests with high species diversity, are demonstrating significant luxury consumption of carbon, and ‘waste’ carbon via idling respiration. This could mean that forests may be more resistant than expected to changing environmental conditions, and may exhibit low sensitivity to elevated atmospheric CO<sub>2</sub> as carbon does not seem to be a limiting resource. Information on responses of tropical trees to elevated CO<sub>2</sub> only exists for tree seedlings or lianas (Marvin et al., 2015; Raj et al., 2014). Information on the effect of elevated atmospheric CO<sub>2</sub> on old-growth tropical forest trees is – to our knowledge – still absent from the literature, however the AmazonFACE experiment (Norby et al., 2016) might shed light on this question. Additional studies focused on the carbon metabolism of individual trees under changing resource supply are needed to better understand forest response to a changing environment. We especially need an improved understanding on which mechanism is responsible for the low CUE in tropical trees since the implications for predicting forest response to a changing environment are highly critical. If the low CUE is the result of carbon oversupply, tropical trees might be more resistant to a changing atmospheric CO<sub>2</sub> concentration.

## 5. Conclusion

We conclude that forest disturbance and species diversity are two important variables controlling carbon use efficiency in tropical forests. Undisturbed tropical forests are characterized by a low CUE and forest disturbance increases CUE. Trees in disturbed forest have an enhanced carbon allocation towards growth. Tree diversity seems to highly favor forest net primary productivity and to an even larger portion gross primary productivity. CUE in diverse stands is accordingly lower and a great fraction of GPP is probably used for processes supporting species complementarity. Developing an improved mechanistic understanding of CUE variability in tropical forests is critical for predicting forest response to changing environmental conditions.

## Acknowledgements

We acknowledge support from Canada Natural Science and Engineering Research Council and of the Smithsonian Tropical Research Institute for the maintenance of the Sardinilla site. The German Research Foundation and the Max Planck Society funded the sap flux studies. Special thanks to the LBA-project for providing eddy flux data for the study site in Manaus. Eddy flux measurements at the Sardinilla site were funded by the North-South Centre of ETH Zurich. Further, Norbert Kunert would like to thank Richard Norby to point out the value of this data set during a brief planning meeting in the jungle of the Amazon. Finally, we would like to thank two anonymous reviewer for their valuable suggestions on how to improve the manuscript.

## References

Amthor, J.S., 2000. The McCree–de Wit–penning de vries–thornley respiration paradigms: 30 years later. *Ann. Bot.* 86, 1–20.

Aparecido, L.M.T., dos Santos, J., Higuchi, N., Kunert, N., 2015. Ecological applications of differences in the hydraulic efficiency of palms and broad-leaved trees. *Trees* 29, 1431–1445.

Beevers, H., 1970. *Respiration in Plants and Its Regulation*. Pudoc, Wageningen.

Bradford, M.A., Crowther, T.W., 2013. Carbon use efficiency and storage in terrestrial ecosystems. *New Phytol.* 199, 7–9.

Bush, S.E., Hultine, K.R., Sperry, J.S., Ehleringer, J.R., Philips, N., 2010. Calibration of thermal dissipation sap flow probes for ring and diffuse porous trees. *Tree Physiol.* 30, 1545–1554.

Chambers, J.Q., Silver, W.L., 2004. Some aspects of ecophysiological and biogeochemical

responses of tropical forests to atmospheric change. *Philos. Trans. R. Soc. Lond. B Biol. Sci.* 359, 463–476.

Chambers, J.Q., Higuchi, N., Schimel, J.P., 1998. Ancient trees in Amazonia. *Nature* 391, 135.

Chambers, J.Q., Santos, Jd., Ribeiro, R.J., Higuchi, N., 2001. Tree damage, allometric relationships, and above-ground net primary production in central Amazon forest. *For. Ecol. Manage.* 152, 73–84.

Chambers, J.Q., Tribuzy, E.S., Toledo, L.C., Crispim, B.F., Higuchi, N., Santos, Jd., Araújo, A.C., Kruijt, B., Nobre, A.D., Trumbore, S.E., 2004. Respiration from a tropical forest ecosystem: partitioning of sources and low carbon use efficiency. *Ecol. Appl.* 14, 72–88.

Clark, D.A., Brown, S., Kicklighter, D.W., Chambers, J.Q., Thomlinson, J.R., Ni, J., 2001. Measuring net primary production in forests: concepts and field methods. *Ecol. Appl.* 11, 356–370.

Coley, P.D., Barone, J.A., 1996. Herbivory and plant defenses in tropical forests. *Annu. Rev. Ecol. Syst.* 27, 305–335.

da Silva, R.P., dos Santos, J., Tribuzy, E.S., Chambers, J.Q., Nakamura, S., Higuchi, N., 2002. Diameter increment and growth patterns for individual tree growing in Central Amazon, Brazil. *For. Ecol. Manage.* 166, 295–301.

DeLucia, E.H., Drake, J.E., Thomas, R.B., Gonzalez-Meler, M., 2007. Forest carbon use efficiency: is respiration a constant fraction of gross primary production? *Glob. Change Biol.* 13, 1157–1167.

Dewar, R.C., Medlyn, B.E., McMurtrie, R.E., 1999. Acclimation of the respiration/photosynthesis ratio to temperature: insights from a model. *Glob. Change Biol.* 5, 615–622.

Doughty, C.E., Goldsmith, G.R., Raab, N., Girardin, C.A.J., Farfan-Amezquita, F., Huaraca-Huasco, W., Silva-Espejo, J.E., Araujo-Murakami, A., da Costa, A.C.L., Rocha, W., Galbraith, D., Meir, P., Metcalfe, D.B., Malhi, Y., 2018. What controls variation in carbon use efficiency among Amazonian tropical forests? *Biotropica* 50, 16–25.

Fernández-Martínez, M., Vicca, S., Janssens, I.A., Luysaert, S., Campioli, M., Sardans, J., Estiarte, M., Peñuelas, J., 2014. Spatial variability and controls over biomass stocks, carbon fluxes, and resource-use efficiencies across forest ecosystems. *Trees* 28, 597–611.

Forrester, D.I., Albrecht, A.T., 2014. Light absorption and light-use efficiency in mixtures of *Abies alba* and *Picea abies* along a productivity gradient. *For. Ecol. Manage.* 328, 94–102.

Goulden, M., Mcmillan, M., Winston, G., 2011. Patterns of NPP, GPP, respiration, and NEP during boreal forest succession. *Glob. Change Biol.* 17, 855–871.

Granier, A., 1985. Une nouvelle méthode pour la mesure du flux de sève brute dans le tronc des arbres. *Ann. For. Sci.* 42, 193–200.

Granier, A., Biron, P., Breda, N., Pontailler, J.Y., Saugier, B., 1996. Transpiration of trees and forest stands: short and long-term monitoring using sapflow methods. *Glob. Change Biol.* 2, 265–274.

Hartley, I.P., Armstrong, A.F., Murthy, R., Barron-Gafford, G., Ineson, P., Atkin, O.K., 2006. The dependence of respiration on photosynthetic substrate supply and temperature: integrating leaf, soil and ecosystem measurements. *Glob. Change Biol.* 12, 1954–1968.

Kato, T., Tang, Y., 2008. Spatial variability and major controlling factors of CO<sub>2</sub> sink strength in Asian terrestrial ecosystems: evidence from eddy covariance data. *Glob. Change Biol.* 2333–2348.

Kelty, M.J., 2006. The role of species mixtures in plantation forestry. *For. Ecol. Manage.* 233, 195–204.

Kunert, N., 2016. Curious relationship revealed by looking at long term data sets—the geometry and allometric scaling of diel xylem sap flux in tropical trees. *J. Plant Physiol.* 205, 80–83.

Kunert, N., Mercado Cardenas, A., 2015. Are diverse tropical tree plantations more resistant and resilient to drought than monocultures? *Forests* 6, 2029–2046.

Kunert, N., Schwendenmann, L., Hölscher, D., 2010. Seasonal dynamics of tree sap flux and water use in nine species in Panamanian forest plantations. *Agric. For. Meteorol.* 150, 411–419.

Kunert, N., Schwendenmann, L., Potvin, C., Hölscher, D., 2012. Tree diversity enhances tree transpiration in a Panamanian forest plantation. *J. Appl. Ecol.* 49, 135–144.

Kunert, N., Barros, P., Higuchi, N., 2013. Do palm water use characteristics explain the spatial distribution of palms in the Central Amazon? *Acta Hort.* 991, 197–204.

Kunert, N., Aparecido, L.M.T., Barros, P., Higuchi, N., 2015a. Modeling potential impacts of planting palms or tree in small holder fruit plantations on ecohydrological processes in the Central Amazon. *Forests* 6, 2530–2544.

Kunert, N., Aparecido, L.M.T., Higuchi, N., Santos, Jd., Trumbore, S., 2015b. Higher tree transpiration due to road-associated edge effects in a tropical moist lowland forest. *Agric. For. Meteorol.* 213, 183–192.

Kunert, N., Aparecido, L.M.T., Wolff, S., Higuchi, N., Santos, Jd., Araujo, A.Cd., Trumbore, S., 2017. A revised hydrological model for the Central Amazon: the importance of emergent canopy trees in the forest water budget. *Agric. For. Meteorol.* 239, 47–57.

Lewis, S.L., Lopez-Gonzalez, G., Sonke, B., Affum-Baffoe, K., Baker, T.R., Ojo, L.O., Phillips, O.L., Reitsma, J.M., White, L., Comiskey, J.A., K. M.-N.D., Ewango, C.E.N., Feldpausch, T.R., Hamilton, A.C., Gloor, M., Hart, T., Hladik, A., Lloyd, J., Lovett, J.C., Makana, J.-R., Malhi, Y., Mbago, F.M., Ndangalasi, H.J., Peacock, J., Peh, K.S.H., Sheil, D., Sunderland, T., Swaine, M.D., Taplin, J., Taylor, D., Thomas, S.C., Votere, R., Woll, H., 2009. Increasing carbon storage in intact African tropical forests. *Nature* 457, 1003–1006.

Luysaert, S., Schulze, E.D., Börner, A., Knohl, A., Hessenmöller, D., Law, B.E., Ciais, P., Grace, J., 2008. Old-growth forests as global carbon sinks. *Nature* 455, 213.

Malhi, Y., 2012. The productivity, metabolism and carbon cycle of tropical forest vegetation. *J. Ecol.* 100, 65–75.



- Malhi, Y., Aragão, L.E.O.C., Metcalfe, D.B., Paiva, R., Quesada, C.A., Almeida, S., Anderson, L., Brando, P., Chambers, J.Q., Da Costa, A.C.L., Hutyra, L.R., Oliveira, P., Patiño, S., Pyle, E.H., Robertson, A.L., Teixeira, L.M., 2009. Comprehensive assessment of carbon productivity, allocation and storage in three Amazonian forests. *Glob. Chang. Biol.* 15, 1255–1274.
- Marvin, D.C., Winter, K., Burnham, R.J., Schnitzer, S.A., 2015. No evidence that elevated CO<sub>2</sub> gives tropical lianas an advantage over tropical trees. *Glob. Change Biol.* 21, 2055–2069.
- Norby, R.J., De Kauwe, M.G., Domingues, T.F., Duursma, R.A., Ellsworth, D.S., Goll, D.S., Lapola, D.M., Luus, K.A., MacKenzie, A.R., Medlyn, B.E., Pavlick, R., Rammig, A., Smith, B., Thomas, R., Thonicke, K., Walker, A.P., Yang, X., Zaehle, S., 2016. Model–data synthesis for the next generation of forest free-air CO<sub>2</sub> enrichment (FACE) experiments. *New Phytol.* 209, 17–28.
- Oelmann, Y., Potvin, C., Mark, T., Werther, L., Tapernon, S., Wilcke, W., 2010. Tree mixture effects on aboveground nutrient pools of trees in an experimental plantation in Panama. *Plant Soil* 326, 199–212.
- Peñuelas, J., 2005. A big issue for trees. *Nature* 437, 965.
- Raj, E.E., Buvanewaran, C., Menason, E., Vijayalakshmi, C., 2014. Physiological response of tropical tree species to elevated CO<sub>2</sub> levels at seedling stage. *Indian J. Ecol.* 41, 16–22.
- Read, D.J., 1991. Mycorrhizas in ecosystems. *Experientia* 47, 376–391.
- Sapijanskas, J., Potvin, C., Loreau, M., 2012. Beyond shading: litter production by neighbors contributes to overyielding in tropical trees. *Ecology* 94, 941–952.
- Sapijanskas, J., Paquette, A., Potvin, C., Kunert, N., Loreau, M., 2014. Tropical tree diversity enhances light capture through crown plasticity and spatial and temporal niche differences. *Ecology* 95, 2479–2492.
- Scherer-Lorenzen, M., Potvin, C., Koricheva, J., Schmid, B., Hector, A., Bornik, Z., Reynolds, G., Schulze, E.-D., 2005. The design of experimental tree plantations for functional biodiversity research. In: Scherer-Lorenzen, M., Körner, C., Schulze, E.-D. (Eds.), *Forest Diversity and Function: Temperate and Boreal Systems*. Springer Berlin Heidelberg, Berlin, Heidelberg, pp. 347–376.
- Schultz, J., Appel, H., Ferrieri, A., Arnold, T., 2013. Flexible resource allocation during plant defense responses. *Front. Plant Sci.* 4.
- Schwendenmann, L., Pendall, E., Sanchez-Bragado, R., Kunert, N., Hölscher, D., 2015. Tree water uptake in a tropical plantation varying in tree diversity: interspecific differences, seasonal shifts and complementarity. *Ecohydrology* 8, 1–12.
- Tanaka, A., 1972. Efficiency of Respiration, Rice Breeding - International Rice Research Institute, Los Banos, Philippines, pp. 483–498.
- Tang, J., Bolstad, P.V., Ewers, B.E., Desai, A.R., Davis, K.J., Carey, E.V., 2006. Sap flux–upscaled canopy transpiration, stomatal conductance, and water use efficiency in an old growth forest in the Great Lakes region of the United States. *J. Geophys. Res. Biogeosci.* 111 n/a-n/a.
- Tatarinov, F., Rotenberg, E., Yakir, D., Klein, T., 2017. Forest GPP calculation using sap flow and water use efficiency measurements. *Bioprotocol* 7, e2221.
- Vanninen, P., Mäkelä, A., 2005. Carbon budget for Scots pine trees: effects of size, competition and site fertility on growth allocation and production. *Tree Physiol.* 25, 17–30.
- Vicca, S., Luysaert, S., Peñuelas, J., 2012. Fertile forests produce biomass more efficiently. *Ecol. Lett.* 520–526.
- Waring, R.H., Landsberg, J.J., Williams, M., 1998. Net primary production of forests: a constant fraction of gross primary production? *Tree Physiol.* 18, 129–134.
- Wolf, S., Eugster, W., Potvin, C., Turner, B.L., Buchmann, N., 2011. Carbon sequestration potential of tropical pasture compared with afforestation in Panama. *Glob. Change Biol.* 17, 2763–2780.