

Effects of tree species diversity on insect herbivory and leaf defences in *Cordia dodecandra*

SILVIA ROSADO-SÁNCHEZ,¹ VÍCTOR PARRA-TABLA,¹
 DAVID BETANCUR-ANCONA,² XOAQUÍN MOREIRA³
 and LUIS ABDALA-ROBERTS¹ ¹Departamento de Ecología Tropical, Campus de Ciencias Biológicas y Agropecuarias, Universidad Autónoma de Yucatán, Mérida, Mexico, ²Facultad de Ingeniería Química, Universidad Autónoma de Yucatán, Mérida, Mexico and ³Forest Ecology, Misión Biológica de Galicia (MBG-CSIC), Pontevedra, Spain

Abstract. 1. It is broadly accepted that plant diversity influences plant–herbivore interactions, yet few studies have separated damage by different herbivore species and simultaneously tested for effects on herbivory and plant defences to elucidate the mechanisms underlying such effects.

2. This study tested for effects of tree species diversity on leaf damage by beetle larvae and caterpillars on *Cordia dodecandra* in a diversity experiment, and measured leaf chemical and physical traits to assess whether differences in herbivory underlie changes in plant traits.

3. Four year-old trees were sampled in 21 × 21-m plots classified as monocultures of *C. dodecandra* or mixtures of this species plus three other tree species. Herbivory by beetle larvae and caterpillars was estimated, and we subsequently collected undamaged leaves to quantify total phenolics and condensed tannins, trichome density, and specific leaf area.

4. A significant effect of diversity on herbivory by beetle larvae was found, with mixtures exhibiting an 80% lower mean value than monocultures. By contrast, there was no diversity effect on caterpillar herbivory. Additionally, a positive relationship was found between beetle larvae (but not caterpillar) damage and plant chemical defences, yet there was no effect of diversity on defences. Specific leaf area was higher and trichome density tended to be lower in mixtures than in monocultures, but neither trait was associated with herbivory, and damage did not explain diversity effects on these traits.

5. These findings emphasise the importance of separating diversity effects on different herbivore species and measuring plant traits associated with herbivory to better understand controls of diversity on plant–herbivore interactions.

Key words. Associational resistance, defences, diversity, herbivory, phenolic compounds.

Introduction

Over the last decade, manipulative experiments have shown that increasing plant species diversity leads to concomitant increases in consumer abundance and diversity (reviewed by Cardinale *et al.*, 2011), particularly in the case of arthropods

(e.g. Koricheva *et al.*, 2000; Haddad *et al.*, 2009). In parallel, studies have also shown that plant diversity drives changes in plant–consumer and consumer–consumer interactions such as herbivory and predation (reviewed by Agrawal *et al.*, 2006; Barbosa *et al.*, 2009; Moreira *et al.*, 2016). Such effects on consumer diversity and species interactions have been attributed to higher primary productivity and greater habitat complexity with increasing diversity, which increase resource availability and multidimensional niche space for consumers, respectively (Langellotto & Denno, 2004; Moreira *et al.*, 2016).

Correspondence: Luis Abdala-Roberts, Departamento de Ecología Tropical, Campus de Ciencias Biológicas y Agropecuarias, Universidad Autónoma de Yucatán, Carretera Mérida-Xmatkuil Km 15.5, C.P. 97100, Mérida, Yuc., Mexico. E-mail: abdala.luis@yahoo.com

The influence of plant species diversity on insect herbivory has received much attention (reviewed by Moreira *et al.*, 2016). One dominant body of theory predicts that, in the case of dietary specialist herbivores, abundance and plant damage should decrease with increasing plant diversity, because preferred host plants are less abundant and therefore more difficult to locate in mixtures (Resource Concentration Hypothesis; Root, 1973; Otway *et al.*, 2005; Salazar *et al.*, 2013). Yet diversity effects on herbivory may vary in magnitude and sign, depending on the herbivore species or guild and how each responds to changes in plant biomass, quality or habitat heterogeneity with increasing plant diversity (Moreira *et al.*, 2016). Hence the importance of separating the effects of plant diversity on different herbivore species in order to understand the underlying mechanisms driving species- or guild-specific patterns. In addition, addressing the effects of plant diversity on herbivory is crucial not only for predicting effects of habitat heterogeneity on trophic interactions, but also because such effects may ultimately influence ecosystem function. For instance, effects of diversity on herbivory may in turn influence plant growth and therefore determine the magnitude and sign of diversity effects on primary productivity (Schuldt *et al.*, 2015). Accordingly, understanding the controls of plant diversity over herbivory is necessary in order to gain a complete understanding of plant diversity–ecosystem function relationships.

Plant diversity may also influence plant traits such as chemical or physical defences against herbivores (Mraja *et al.*, 2011; Moreira *et al.*, 2014, 2017; Kostenko *et al.*, 2017), and these effects may contribute to explain effects of diversity on plant–herbivore interactions. Diversity effects on plant traits may be due to changes in abiotic conditions (e.g. soil nutrients or light availability; Rosado-Sánchez *et al.*, 2018), in plant–plant interactions (e.g. competition; Kim & Underwood, 2015), in plant–herbivore interactions (e.g. McArt & Thaler, 2013), or in plant endogenous resource allocation processes (e.g. higher growth leads to decreased levels of defences when these functions trade off; McArt & Thaler, 2013). For instance, in the case of plant–herbivore interactions, differences in the amount of herbivory in monocultures versus mixtures are expected to lead to concomitant differences in the strength of induction and defence levels in attacked plant tissues. To date, however, few studies have simultaneously measured the effects of plant diversity on herbivory and plant defensive traits (but see Mraja *et al.*, 2011; McArt & Thaler, 2013; Moreira *et al.*, 2014; Kostenko *et al.*, 2017), therefore limiting our understanding of whether effects of diversity on herbivory are mediated by changes in specific plant traits and if there are feedbacks between induced plant responses and herbivory shaping these interactions. Further, even fewer studies have reported on the concurrent influences of multiple herbivore species under realistic scenarios where plants are under attack by more than one herbivore species (but see Kostenko *et al.*, 2017).

This study was aimed at evaluating the effects of tree species diversity on insect herbivory and plant defences in the tropical tree *Cordia dodecandra* A. DC. (Boraginaceae) planted in a forest diversity experiment in southern Mexico. We chose *C. dodecandra* as the focal species because it exhibits high levels of herbivory by leaf-chewing insects which reach outbreak

levels on this tree species in the study system (up to 50% of leaf area removed), therefore representing a conspicuous and ecologically important interaction. Most of the leaf damage recorded was caused by one species of beetle and two caterpillar species which are common herbivores of this tree species in managed or disturbed sites in the study region. We measured leaf damage caused by these herbivores and subsequently collected leaves to quantify chemical (total phenolics and condensed tannins) and physical [trichome density, specific leaf area (SLA)] traits potentially associated with resistance to insect herbivores. Specifically, we sought to answer the following questions: (i) is there an effect of tree diversity on insect herbivory in *C. dodecandra*, and does the magnitude or sign of such an effect vary between the two types of herbivores; and (ii) is there an effect of diversity on leaf chemical and physical traits associated with herbivore resistance, and do differences in herbivory explain these plant phenotypic changes as well as changes in plant growth? The studied species of beetle and caterpillars only feed on *C. dodecandra* in this study system, i.e. they are dietary specialists on this species, so following predictions by the Resource Concentration Hypothesis, we expected lower levels of herbivory by all three insect species in polyculture than in monoculture and, in turn, lower levels of defences in polyculture due to weaker induced responses to damage in plants found in mixtures. In measuring the effects of plant diversity on multiple herbivore species and plant defensive traits, this study moves a step forward towards understanding the influence of producer diversity on plant phenotypes and phytophagous insects associated with trees.

Materials and methods

Study site and tree species

The study was conducted at the Autonomous University of Yucatan (UADY) Tree Diversity Experiment (20°24'44"N, 89°45'13"W), located near the town of Muna, Yucatán (México). The experiment is found within the 'Uxmal Experimental Site', a field station owned and administered by the Instituto Nacional de Investigaciones Forestales Agrícolas y Pecuarias (INIFAP). Mean annual temperature at the study site is 26.6 °C and mean annual precipitation is 1300 mm, with most of the rainfall occurring from May to October (Orellana *et al.*, 1999). The experiment contains the following species of tropical trees: *Swietenia macrophylla* King (Meliaceae), *Enterolobium cyclocarpum* Jacq. (Leguminosae), *Tabebuia rosea* Bertol. (Bignoniaceae), *Ceiba pentandra* L. (Malvaceae), *Piscidia piscipula* L. (Leguminosae), and *Cordia dodecandra*. All these species are long-lived and most occur in sympatry in tropical wet and dry forests of the Yucatan Peninsula (Pennington & Sarukhán, 2005). Seeds of each species were collected from adult individuals found in southern Quintana Roo (México) during January and March 2011 (Abdala-Roberts *et al.*, 2015).

Cordia dodecandra, the focal tree species in this study, is a deciduous tree native to southern Mexico and Central America, which grows up to 12 m tall and 60 cm in diameter at breast height. Leaves are simple, have an undulated edge, and are characterised by a high density of trichomes on both sides. The

abaxial side of the leaf has simple glandular trichomes whereas the adaxial surface has starred trichomes (S. Rosado-Sánchez, pers. obs.). Previous studies have reported that *Cordia* species induce defences in response to insect herbivore damage, including both chemical (e.g. phenolic compounds) and physical (e.g. leaf toughness, thickness) traits correlated with herbivore resistance (Frederickson *et al.*, 2013). Species from this genus have also been shown to induce indirect defensive traits such as domatia and extrafloral nectar to attract predatory ants (Frederickson *et al.*, 2013). However, ant abundance on *C. dodecandra* has been low during previous years in this study system (Rosado-Sánchez *et al.*, 2018) as well as during this study's sampling season, and we therefore focused exclusively on defensive traits conferring direct defence.

In the study system, *C. dodecandra* is heavily attacked by the beetle *Eurypepla brevilineata* (Boheman 1854, Chrysomelidae), mainly by skeletonising larvae and, to a lesser extent, by adults of this species. Both adults and larvae aggregate on leaves and reach peak abundance in June and July (S. Rosado-Sánchez, pers. obs.). This species is a common pest on *C. dodecandra* in managed or disturbed sites in Yucatan (L. Abdala-Roberts pers. obs.), indicating that this interaction is potentially widespread and relevant from a management perspective. On the other hand, leaves are also consumed by larvae of the moth species *Cropia connecta* (Smith 1894, Noctuidae) and *Ethmia delliella* (Fernald 1891, Depressariidae). The latter has been previously reported to feed on species of *Cordia* (e.g. *C. alliodora*; Phillips-Rodríguez *et al.*, 2014), whereas the former may achieve pest levels feeding on species such as *Cordia alliodora*, *Cordia panamensis* and *Cordia boissieri* (Janzen, 1981). All three insect species feed exclusively on *C. dodecandra* in the study system (L. Abdala-Roberts, unpublished) and therefore can be considered specialists on this species for the purposes of this experiment. In this study, we focused exclusively on leaf damage caused by larvae of these insects.

Experimental design and plot selection

The tree diversity experiment was established in December 2011 and includes a total of 74 plots, each 21 × 21 m, covering an area of 7.2 ha. Distance between plots is 6 m and distance between plants within plots is 3 m. Plot size, planting density, and the separation between plots fall within the range of design features of other tree diversity experiments that have measured effects on productivity and plant–insect interactions (Scherer-Lorenzen *et al.*, 2005; Grossman *et al.*, 2018), and significant effects of diversity have been found previously for insect communities associated with this system (e.g. Abdala-Roberts *et al.*, 2015; Campos-Navarrete *et al.*, 2015). Plots are classified as either monocultures ($N = 42$, two plots per species except for mahogany for which a greater number of monoculture plots were planted to test for effects of genotypic diversity within this species; see Abdala-Roberts *et al.*, 2015) or polycultures ($N = 32$, random combinations of four out of the six species) (see Fig. S1 in the Supplementary Material). Each plot contains 64 plants, making a total of 4780 plants at the start of the experiment; cumulative tree mortality was less than 5% at the time this study was conducted. Of this total, 448 plants

correspond to *C. dodecandra* (128 in monoculture and 320 in polyculture), and for this study we used the two monoculture plots where this species is found, as well as four out of 20 polycultures where the species is planted (Fig. S1). We sampled 29 plants across both monocultures (14 or 15 plants per plot) and 44 plants in polyculture (10–12 plants per plot), avoiding plants located on the outer rows of the plots to reduce edge effects.

Measurements of herbivory and leaf sampling

In mid-July 2015, two weeks after the peak in herbivore abundance on *C. dodecandra*, we estimated leaf herbivory by beetle larvae and caterpillars. Particularly strong herbivory outbreaks on *C. dodecandra* were observed in 2014 and 2015 (year of this study). The fact that these interactions were intense the previous year of this study motivated the present work with the expectation that subsequent outbreaks would (and in fact did) occur. The studied beetle larvae are skeletonisers and produce a pattern of damage that is clearly distinguishable from that caused by caterpillars. Observations of other species of leaf chewers on *C. dodecandra* were rare during the study, and other groups of phytophagous insects (miners, gallers, sap-suckers) were also extremely rare (S. Rosado-Sánchez, pers. obs.). Therefore, virtually all of the herbivory in leaves (and therefore overall herbivory across plant tissues) recorded during this study was caused by the three species of focal herbivores. For each plant, we selected two branches of similar length (1–1.5 m), leaf number, and height above ground level (1–1.5 m). For each branch, we counted the total number of leaves, the number of leaves attacked by beetle larvae, and the number of leaves attacked by caterpillars. Individual leaves were rarely simultaneously attacked by beetle larvae and caterpillars, such that estimates of herbivory effectively represented separate measures of damage inflicted by each type of herbivore. Leaf longevity of *C. dodecandra* spans several months (> 3 months.) and leaves that are heavily damaged are not dropped immediately by the plant; rather they remain attached for a considerable amount of time (L. Abdala-Roberts, pers. obs.). Accordingly, we measured leaf damage right after the insect outbreak took place, so the chance of missing heavily damaged leaves because they were previously abscised is unlikely. For each branch, we calculated the proportion of leaves attacked by each type of herbivore and used the mean value across branches per plant for statistical analyses. We note that measurements of proportion of damaged leaves may not always correlate with the amount of leaf area consumed, and should therefore be interpreted exclusively as a measure of frequency of attack (Moreira *et al.*, 2018). In addition to herbivory, we also previously measured plant height in May 2014 to account for effects of initial plant size (surrogate of apparency or detectability) on herbivory (Castagneyrol *et al.*, 2013).

In early October 2015, 2.5 months after herbivore measurements were taken and the outbreak subsided, we collected seven to eight new, fully expanded leaves from each plant for quantification of chemical and physical traits. Leaves were collected from different branches from those used to estimate herbivory, and were chosen based on the following criteria: that leaves were from a single branch (to avoid branch-to-branch

variation in leaf traits), that branches were at a similar height above ground level (1–1.5 m), and that leaves were collected at a similar position along the branch and had no evidence of insect damage or disease. By sampling healthy, undamaged leaves, our goal was to reduce variation in leaf traits resulting from localised induction, therefore allowing a better estimate of systemic induction in response to overall levels of damage experienced by the plant as a whole (Moreira *et al.*, 2014, 2018). A focus on systemic induction is crucial in this study, as caterpillar and beetle abundance have consistently reached outbreak levels during the last three years in the system (L. Abdala-Roberts, pers. obs.). Such high levels of damage probably result in systemic responses at the whole-plant level, a phenomenon that is typical of communities characterised by insect outbreaks which may contribute to feedbacks and temporal fluctuations in plant–herbivore interactions (Kessler *et al.*, 2012). Leaves were placed in paper bags and transported in coolers to the laboratory where they were immediately dried at 40–45 °C until a constant weight was achieved.

We quantified leaf chemical defences by measuring the concentration of total phenolics and hydrolysable tannins, both of which are considered broad insect feeding deterrents that confer resistance against insect herbivores due to their toxicity and anti-nutritional qualities (Salminen & Karonen, 2011; Mithöfer & Boland, 2012). In addition, they have also been shown to confer resistance against leaf-chewing insects in the genus *Cordia* (Frederickson *et al.*, 2013). Although qualitative changes (e.g. compound relative abundances or composition) in response to damage may also take place, long-lived trees such as *Cordia* spp. are exposed to numerous herbivore species throughout their life and are therefore expected to rely heavily on quantitative defences for herbivore resistance. To extract phenolic compounds, we diluted 0.3 g of ground leaf tissue in 10 ml of a 50% methanol/water solution with a pH of 2, and then taken to an ultrasound bath (Branson Ultrasonics, Danbury, Connecticut) at ambient temperature for 15 min using 50-ml Falcon tubes. Samples were centrifuged for 15 min, and the supernatant was collected and stored in a freezer at –20 °C. Quantification of total phenolics was done colorimetrically with the Folin-Ciocalteu reactive (Makkar, 2003; Moreira *et al.*, 2014), using 5 µl of the methanolic extract, 45 µl of the methanol:water solution (50%, pH = 2), 500 µl of the Folin-Ciocalteu reactive at 1 N, and 400 µl of Na₂CO₃ at 20%. Absorbance values were measured with a spectrophotometer (VE-5100UV, Velab, Madrid, Spain) at 765 nm, and gallic acid was used as standard. To quantify condensed tannins, we used the proanthocyanidin method modified by Moreira *et al.* (2014). Briefly, we prepared a reactive with 0.7 g of FeSO₄·7H₂O in 50 ml of concentrated HCl which was gauged with n-butanol up to 1 litre. Using tubes with screw-in tops we added 2.8 ml of the reactive extract and 0.2 ml of the methanolic extract. Samples were then submerged in water at 95 °C for 40 min. Absorbances were measured at 550 nm and we used commercial quebracho as standard. Concentrations of total phenolics and condensed tannins were expressed as mg equivalents to the standard g⁻¹ dry tissue (mg g⁻¹).

In the case of leaf physical traits, for each plant we estimated trichome density by selecting one of the leaves used for chemical (above) and counting the number of trichomes present

in each of two 0.20-cm² discs per leaf using a stereoscopic microscope. These values were averaged to obtain a single value per plant for statistical analyses. Counts were performed exclusively on the abaxial surface (simple, glandular trichomes), as it was not possible to confidently quantify trichomes on the adaxial surface (starred trichomes). Values were extrapolated to 1 cm² and trichome density was expressed as the number of trichomes cm⁻². Glandular trichomes are frequently associated with herbivore resistance against insect herbivores (Dalin *et al.*, 2008), and previous work has shown that trichome density is associated with resistance against leaf-chewing insects in *Cordia* (Frederickson *et al.*, 2013). In addition, we used the same leaf disks to measure SLA and expressed this trait as the amount of leaf surface (cm²) per g of dry leaf tissue (cm² g⁻¹). Although SLA is usually related to plant function and resource use, it is also correlated with leaf toughness and is therefore associated with mechanical resistance to herbivory (Onoda *et al.*, 2011; Marquis *et al.*, 2012).

Finally, we measured diameter at breast height (DBH, cm) in March 2016 and 2017 for all plants. These values were transformed to basal area (BA, in cm²) using $BA = \pi \left(\frac{DBH}{2} \right)^2$ and we then calculated proportional growth rate based on these data $[(BA_{2017} - BA_{2016})/BA_{2016}]$, to test whether any such previous differences in herbivory (2015) influenced subsequent plant growth.

Statistical analyses

We ran general linear mixed models (GLMMs) using PROC MIXED in SAS 9.2 (SAS Institute Inc., 2008) testing for an effect of diversity (fixed, two levels) on the proportion of leaves attacked by caterpillars, the proportion of leaves attacked by beetle larvae, the concentration of total phenolics, the concentration of condensed tannins, SLA, trichome density, and plant growth rate. In all these models, we included plot and the plot × diversity interaction, both treated as random effects. The effect of plot was included to account for spatial heterogeneity (i.e. block effect) whereas the interaction allowed us to test the effect of diversity using plot as the unit of replication (Littell *et al.*, 2006). For the herbivory models, we also included plant height (2014) as a covariate to control for residual variation in herbivory due to previous differences in plant size not accounted for by diversity.

For all leaf traits and plant growth rate, we subsequently ran models adding the proportion of leaves attacked by each group of herbivores as covariates. By comparing results between models without and models with these covariates (hereafter ‘models 1 and 2’, respectively) we were able to determine whether the effect of diversity on a given trait or growth was mediated by herbivory. If this were the case, a significant effect of diversity in model 1 (without covariates) should become non-significant after accounting for leaf damage in model 2 (see Abdala-Roberts *et al.*, 2016). Including leaf damage as a covariate also allowed us to test for an association between herbivory and leaf defences (suggestive of plant induction in response to damage) and plant growth (i.e. diversity feedbacks on plant growth via herbivory). In all cases, residuals were normally distributed and we report least-square means and standard errors as descriptive statistics. Trichome density, total phenolics, and condensed tannins

Table 1. Results from general linear mixed models testing for effects of tree species diversity on the proportion of leaves attacked by beetle larvae (*Eurypepla brevilineata*) and caterpillars (*Cropia connecta* and *Ethmia delliella*) feeding on *Cordia dodecandra* in a tree diversity experiment in Yucatán (Mexico)

Effect	Beetle herbivory			Caterpillar herbivory		
	d.f.	<i>F</i>	<i>P</i>	d.f.	<i>F</i>	<i>P</i>
Diversity	1.4	14.59	0.018	1.4	0.01	0.926
Tree height	1.62	2.56	0.115	1.62	0.95	0.332

Significant effects ($P < 0.05$) are in bold.

were all significantly positively correlated (Pearson's $r \geq 0.24$, $P \leq 0.04$), whereas SLA was negatively correlated with trichome density ($r = -0.52$, $P < 0.0001$) and tended to be negatively correlated with total phenolics ($r = -0.23$, $P = 0.06$).

Results

Leaf herbivory

We found a significant effect of tree species diversity on the proportion of damaged leaves by beetle larvae (Table 1), for which the mean value in polyculture was 81% lower (0.032 ± 0.021) than in monoculture (0.170 ± 0.028 ; Fig. 1). In contrast, there was no effect of diversity on the proportion of leaves attacked by caterpillars (monoculture, 0.250 ± 0.024 ; polyculture, 0.253 ± 0.019 ; Table 1; Fig. 1). Plant height did not significantly influence the proportion of damaged leaves by either group of herbivores (Table 1).

Leaf traits and effects of herbivory on traits

We found no effect of diversity on either the concentration of total leaf phenolics (monoculture = $5.84 \pm 1.07 \text{ mg g}^{-1}$, polyculture = $3.91 \pm 0.77 \text{ mg g}^{-1}$) or condensed tannins (monoculture = $1.84 \pm 0.05 \text{ mg g}^{-1}$, polyculture = $1.87 \pm 0.04 \text{ mg g}^{-1}$) (model 1 in Table 2; Fig. 2a,b). The effect of diversity remained non-significant in both cases after accounting for leaf herbivory by each group of insects (model 2 in Table 2). In both of these models with covariates, we found a significant (positive) effect of leaf damage by beetle larvae on the concentration of total phenolics and condensed tannins, whereas leaf damage by caterpillars was not significantly associated with either of these traits (Table 2).

For leaf physical traits, there was a marginally significant effect ($P = 0.057$) of diversity on trichome density (Table 2), where the mean value in monoculture was 24% greater ($1467.56 \pm 85.93 \text{ trichomes cm}^{-2}$) than in polyculture ($1180.03 \pm 66.28 \text{ trichomes cm}^{-2}$) (Fig. 2c). This effect remained unchanged (marginally significant) after accounting for leaf damage by beetle larvae and caterpillars, and neither of these covariates was significantly associated with this trait (model 2 in Table 2). In addition, we found a significant effect of diversity on SLA (Table 2), with polycultures exhibiting a 39% greater mean value ($167.80 \pm 8.46 \text{ cm}^2 \text{ g}^{-1}$) than monocultures ($120.48 \pm 11.07 \text{ cm}^2 \text{ g}^{-1}$) (Fig. 2d). Again, the effect of diversity remained significant after including leaf damage by beetle

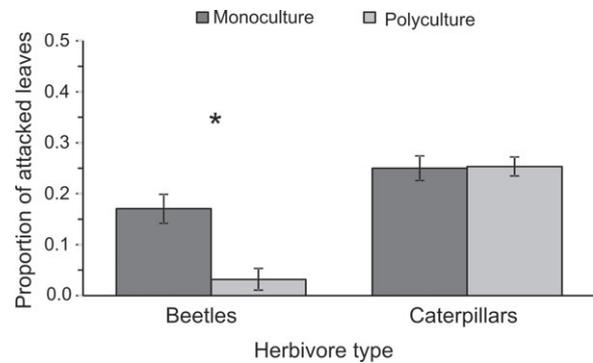


Fig. 1. Effects of tree species diversity on the proportion of leaves of *Cordia dodecandra* damaged by beetle larvae and caterpillars. Values are least-square means and standard errors for the proportion of leaves attacked by each type of herbivore from a general linear mixed model testing for the effect of diversity (fixed) and including plot and the plot \times diversity interaction as random effects. * $P < 0.05$.

Table 2. Results from general linear mixed models testing for the effect of tree species diversity on the concentration (mg g^{-1}) of leaf total phenolics, leaf condensed tannins, leaf trichome density (trichomes cm^{-2}), specific leaf area ($\text{cm}^2 \text{ g}^{-1}$), and growth rate (percentage change in stem basal area from 2016 to 2017) for *Cordia dodecandra* in a tree diversity experiment in Yucatán (Mexico). All models included previous plant height (2014) to account for initial differences in plant size

Effects	Model 1			Model 2		
	d.f.	<i>F</i>	<i>P</i>	d.f.	<i>F</i>	<i>P</i>
Total phenolics						
Diversity	1.4	2.14	0.218	1.4	0.81	0.419
Beetle damage	–	–	–	1.61	6.53	0.013
Caterpillar damage	–	–	–	1.61	0.96	0.330
Condensed tannins						
Diversity	1.4	0.18	0.694	1.4	1.70	0.262
Beetle damage	–	–	–	1.61	4.38	0.041
Caterpillar damage	–	–	–	1.61	0.03	0.858
Trichomes						
Diversity	1.4	7.02	<i>0.057</i>	1.4	5.59	<i>0.077</i>
Beetle damage	–	–	–	1.62	0.43	0.512
Caterpillar damage	–	–	–	1.62	0.09	0.762
Specific leaf area						
Diversity	1.4	11.53	0.027	1.4	9.60	0.036
Beetle damage	–	–	–	1.62	0.51	0.479
Caterpillar damage	–	–	–	1.62	0.06	0.809
Growth rate						
Diversity	1.4	1.47	0.293	1.4	1.56	0.280
Beetle damage	–	–	–	1.61	0.23	0.630
Caterpillar damage	–	–	–	1.61	0.71	0.402

Significant ($P < 0.05$) and marginally significant ($0.05 < P < 0.10$) values are in bold and italics, respectively.

larvae and caterpillars in model 2, and neither of these covariates was significantly associated with this trait (model 2 in Table 2). Finally, we found no effect of diversity on plant growth rate (monoculture, 0.185 ± 0.044 ; polyculture, 0.118 ± 0.035) and neither beetle larvae nor caterpillar herbivory was significantly associated with growth (model 2 in Table 2).

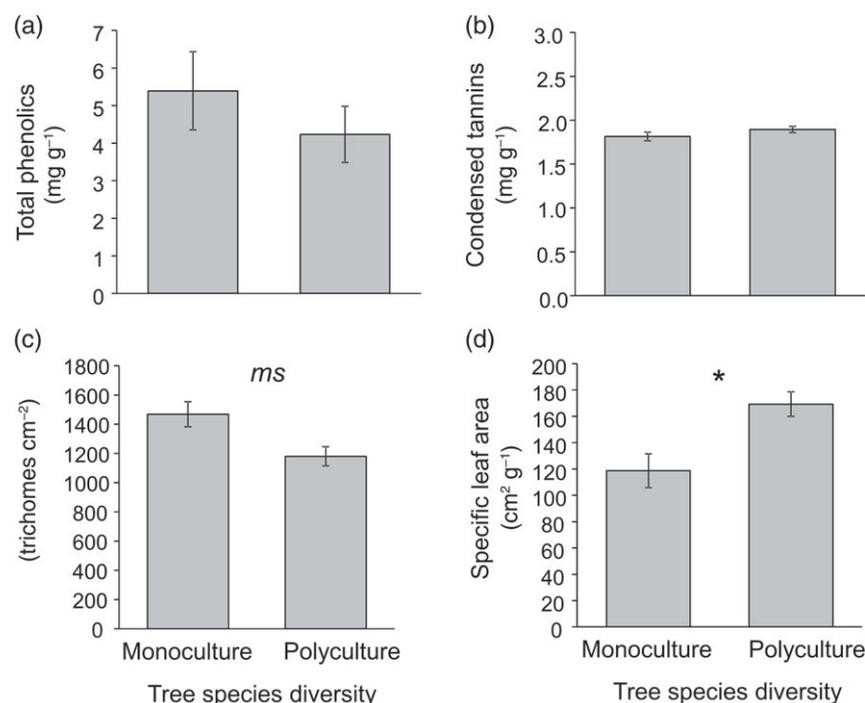


Fig. 2. Effects of tree species diversity on *Cordia dodecandra* chemical and physical leaf traits. Values are least-square means and standard errors from general linear mixed models testing for diversity as fixed effect and including plot and plot \times diversity as random factors. Traits shown in each panel are as follows: the concentration (mg g^{-1}) of total phenolics (a) and condensed tannins (b), trichome density (trichomes cm^{-2}) (c), and specific leaf area ($\text{cm}^2 \text{g}^{-1}$) (d). * $P < 0.05$; ms, marginally significant ($P = 0.057$).

Discussion

The results showed contrasting effects of tree species diversity on leaf damage by beetle larvae and caterpillars associated with *C. dodecandra*. Whereas the incidence of leaf damage by beetle larvae was substantially reduced in polyculture relative to monoculture, we found no detectable effect of diversity on caterpillar herbivory. In addition, we found effects of diversity on physical traits, namely lower trichome density and greater SLA in polyculture than in monoculture, but no effect on chemical traits. Further, there were significant positive associations between beetle larvae (but not caterpillar) damage and chemical defences, but no associations between either type of herbivory and leaf physical traits. Thus, in cases where diversity influenced leaf traits such trait changes were not associated with damage (trichomes, SLA), and in cases where herbivory was associated with traits, there was no effect of diversity on such traits to start with (total phenolics, tannins). These findings indicate that changes in herbivory did not mediate diversity effects on the studied leaf traits, presumably due to weak plant-induced responses to herbivory or other factors counteracting effects of diversity on plant traits via herbivory.

Effects of tree diversity on *Cordia dodecandra* insect herbivory

We observed a strong negative effect of tree species diversity on the frequency of leaf damage by larvae of the chrysomelid beetle *E. brevilineata*. This finding is in agreement with the Resource Concentration Hypothesis which predicts that

reductions in host plant–insect encounter rates as preferred host plants become increasingly rare in more diverse plant patches should lead to lower rates of herbivory by specialist herbivores (Root, 1973). Empirical support for this prediction is relatively strong based on a number of studies conducted over the last decade showing a similar pattern for specialist herbivores (e.g. Jactel & Brockerhoff, 2007; Salazar *et al.*, 2013; Abdala-Roberts *et al.*, 2015; reviewed by Moreira *et al.*, 2016). In contrast, there was no detectable effect of tree diversity on herbivory by moth caterpillars, despite the fact that *C. connecta* and *E. deliella* can also be considered specialists on *C. dodecandra* in this system. These contrasting effects of diversity on attack by beetle larvae versus caterpillars suggest that other traits besides diet breadth explained these contrasting responses. We next speculate on several potential explanations for these patterns. Insect dispersal ability, for example, may condition plant diversity effects such that herbivores with low mobility tend to exhibit weaker responses to plant diversity, particularly in the case of larger-scale experiments, as they do not move long enough distances and their numbers do not segregate between monocultures and mixtures (Bommarco & Banks, 2003). Following this interpretation, ovipositing female moths would be expected to have a lower dispersal ability than adult beetles, therefore weakening diversity effects on caterpillar damage. Alternatively, greater efficiency in host plant location by moths could have made them overcome reduced host densities in polycultures (Abdala-Roberts *et al.*, 2015). Interestingly, the fact that adult beetles feed in groups (possibly due to the release of aggregation pheromones for mating) may have resulted in

greater attraction of adults to monocultures and, in turn, higher larvae abundance. This mechanism, however, explains patterns of beetle but not caterpillar herbivory. Overall, these results emphasise that closer attention to these as well as other traits (e.g. ovipositing behaviour) is necessary in order to elucidate the mechanisms underlying observed patterns of herbivory.

It should be noted that our estimate of herbivory (frequency of damaged leaves) may differ from measurements of the amount of leaf area consumed (Moreira *et al.*, 2018). Nonetheless, in the case of herbivory by beetle larvae, the amount of leaf damage per leaf was consistently high (> 50% of leaf area lost), which suggests that counts of damaged leaves were probably well correlated with the amount of leaf area lost or damaged by this insect. In the case of caterpillars, however, we are less certain whether our measurements were correlated with the amount of leaf tissue consumed. Regardless, data for frequency of attacked leaves are, in their own right, highly informative regarding underlying changes in plant–herbivore interactions (e.g. Tack *et al.*, 2010; see discussion in Moreira *et al.*, 2018) and results based on such measurement undoubtedly revealed contrasting effects of diversity on patterns of attack by beetles and caterpillars.

Effects of tree diversity on Cordia dodecandra leaf traits and growth

The lack of diversity effects on *C. dodecandra* total phenolics and condensed tannins in leaves is somewhat surprising considering that: (i) both traits were significantly associated with leaf damage by beetle larvae (suggestive of damage driving an increase in defence levels via plant induction); and (ii) damage by beetle larvae was strongly affected by diversity. Previous work with *C. nodosa* similarly found that folivory by another species of chrysomelid is associated with increases in total phenolics, also suggesting induced responses to herbivory in this species (Frederickson *et al.*, 2013). Based on the observed patterns, we would have expected lower herbivory by beetle larvae in polyculture to lead to weaker induction and therefore lower levels of chemical defences, whereas plants in monoculture would presumably be highly induced. Results instead agree with previous work in this system showing no association between damage by stem-boring caterpillars and differences in chemical defence levels between monoculture and polyculture for big-leaf mahogany *S. macrophylla* (Moreira *et al.*, 2014). A possible explanation for these patterns, particularly in the case of *C. dodecandra* for which damage was correlated with defences, is that some other factor counteracted potential differences in the magnitude of plant induction by beetle damage between monoculture and polyculture. For example, differences in plant–plant interactions or abiotic conditions (e.g. soil nutrients) between plot types could have offset differences in defence induction by beetle damage (Mraja *et al.*, 2011; Roscher *et al.*, 2011). We also note that these compounds were present at low concentrations and their inducibility might be limited in *C. dodecandra* (i.e. low plasticity; Sinimbu *et al.*, 2012), or at least not strong enough for substantial differences in herbivory to produce concomitant differences in defence levels. On the other hand, it is also possible that other chemical defences not measured (e.g. saponins, terpenes) or specific groups of phenolic compounds

(e.g. lignins, flavonoids, etc.) responded more strongly to herbivory. A broader and more comprehensive assessment of leaf chemical defences is therefore desirable in future work.

One important consideration in interpreting results for chemical defences is that we lack measurements prior to herbivory (i.e. at the start of the growing season), which complicates interpretations about potential plant induced responses and resulting levels of defence after damage. For instance, it is possible that prior to the studied insect outbreak, leaf chemical defences differed between polyculture and monoculture due to some other factor (e.g. abiotic conditions). If chemical defences were higher in polyculture at this moment, then once the outbreak took place and presumably resulted in higher induction of plants in monoculture, this would have led to non-significant differences between monoculture and mixtures. Although measurements conducted in previous years have similarly found that chemical defences in *C. dodecandra* are not influenced by tree diversity (Rosado-Sánchez *et al.*, 2018), the scenario described cannot be ruled out for our study; hence the importance of measuring leaf traits prior to outbreaks to control for initial differences in plant traits. Additionally, we also note that new leaves were collected about two months after the outbreak subsided. This sampling scheme was chosen to provide a long enough time window to collect fully expanded new leaves on all or most plants, as the production of first (presumably more defended) new leaves after the outbreak and their full expansion takes between 1 and 2 months. Still, it is possible that this amount of time was too long to detect the systemic induced response, which may have occurred earlier and subsided at the time of leaf collection. A closer look at short-term temporal patterns of induction after damage is necessary to evaluate this possibility. At the same time, levels of herbivory and induced defences may vary over the growing season, such that surveying leaf damage and quantifying leaf defences at multiple time points would help to better describe and understand both short- and longer-term temporal dynamics (i.e. feedbacks) in insect outbreaks and plant-induced defences.

In contrast to leaf chemical traits, tree diversity did influence *C. dodecandra* leaf physical traits. However, neither trichome density nor SLA were significantly associated with herbivory, suggesting that these traits were not influenced by insect herbivory via induced responses to damage. This is in contrast to a study by Frederickson *et al.* (2013), who found a positive correlation between insect leaf herbivory and trichome density in *C. nodosa*. Accordingly, the effect of diversity on both traits remained unchanged after accounting for leaf damage in the statistical model (i.e. model 2), which confirms that diversity effects on these physical traits were not mediated by herbivory. The abaxial surface of *C. dodecandra* leaves has glandular trichomes which may serve functions other than herbivore resistance, such as water retention or thermal regulation. Accordingly, lower temperature and evapotranspiration due to lower light availability in polycultures (monoculture photon flux density, $2,132 \pm 14.6 \mu\text{mol s}^{-1} \text{m}^{-2}$; polycultures, $297 \pm 126.5 \mu\text{mol s}^{-1} \text{m}^{-2}$; data from this study) may be driving factors behind reduced leaf trichome densities in polycultures. Similarly, higher SLA in polycultures could have been associated with reduced light availability in order to reduce leaf

construction costs (Meziane & Shipley, 1999), suggesting that this trait was more strongly controlled by changes in the abiotic environment than by herbivory. Also, the fact that these two traits were negatively correlated themselves and neither one was associated with leaf damage suggests that an increase in SLA in polycultures could have driven a concomitant decrease in trichome density (or vice versa) via correlated trait responses.

Recent work has shown that effects of diversity on plant growth and primary productivity may be (indirectly) shaped by herbivory. For example, a study by Schuldt *et al.* (2015) reported greater insect herbivory with increasing tree diversity, which in turn was associated with (and presumably drove) reductions in tree growth in a young forest plantation. Our findings, however, do not support this dynamic. First, there was no association between herbivory and plant growth rate. Second, even if there had been an association, we found no effect of diversity on *C. dodecandra* growth rate. Although these results may change later in the development of this system, thus far there is no evidence that herbivory mediates diversity effects on the growth of this species.

Overall, this study illustrates the specificity of plant diversity effects on different species of insect herbivores. These results strengthen the current view that in order to fully comprehend the effects and underlying mechanisms of plant diversity on herbivory, separate measurements of herbivory by different species or guilds are needed. Teasing apart these individual effects on herbivores will then increase our understanding of how overall levels of damage are shaped by plant-based habitat heterogeneity. In addition, our results also indicated a weak link between plant diversity and traits through changes in herbivory, which would have presumably been mediated by differences in the strength of plant-induced responses to damage. Still, we call for future work that specifically addresses the effects of diversity via herbivory on ecologically relevant plant traits, as these trait changes can, in turn, affect ecosystem functions and interactions, including herbivory. In addressing this task, it will be important to determine the relative importance of direct versus indirect effects between plant diversity, traits and herbivory, as well as to account for additional sources of variation influencing herbivory (e.g. soil conditions, predator top-down control). Finally, whereas our study sought to evaluate whether diversity effects on herbivory resulted in changes in plant traits, patterns of herbivory could have been determined initially by differences in plant traits responding to some other factor varying between monocultures and polycultures. In order to address a more dynamic and realistic view of plant diversity–herbivory relationships, measurements of plant traits before and after herbivory events within and across growing seasons are needed, particularly in systems which regularly undergo insect outbreaks. Doing so will allow us to describe fluctuations in interaction strength (e.g. temporal stability) as well as to elucidate feedbacks between plant defences and insect herbivory, ultimately increasing our understanding of how plant diversity influences the function of plant–herbivore interactions.

Acknowledgements

We thank B. Pérez-Niño, T. Quijano-Medina, E. barbosa and L. A. Fernández-Martínez for assistance in the field and laboratory.

We also thank two anonymous reviewers for comments on a previous version of this manuscript. This research was supported by CONACyT grant 250925 awarded to VPT and LAR, as well as a grant from the i-COOP+ CSIC program (COOPB20158) to XM and LAR. INIFAP provided logistic accommodations and infrastructure for the establishment of the experiment, subsequent sampling and maintenance. LAR was supported by a CONACyT repatriation grant (250934) and XM was supported by the Ramón y Cajal Research Programme (RYC-2013-13230). The authors have no conflict of interest to declare.

Supporting Information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Figure S1. Map showing layout of experimental plots in the tree diversity experiment.

References

- Abdala-Roberts, L., Mooney, K.A., Quijano-Medina, T., Campos-Navarrete, M., González-Moreno, A. & Parra-Tabla, V. (2015) Comparison of tree genotypic diversity and species diversity effects on different guilds of insect herbivores. *Oikos*, **124**, 1527–1535.
- Abdala-Roberts, L., Hernández-Cumplido, J., Chel-Guerrero, L., Betancur-Ancona, D., Benrey, B. & Moreira, X. (2016) Effects of plant intra-specific genetic diversity across three trophic levels: underlying mechanisms and plant traits. *American Journal of Botany*, **103**, 1810–1818.
- Agrawal, A.A., Lau, J.A. & Hambäck, P.A. (2006) Community heterogeneity and the evolution of interactions between plants and insect herbivores. *The Quarterly Review of Biology*, **81**, 349–376.
- Barbosa, P., Hines, J., Kaplan, I., Martinson, H., Szczepaniak, A. & Szendrei, Z. (2009) Associational resistance and associational susceptibility: having right or wrong neighbors. *Annual Review of Ecology, Evolution, and Systematics*, **40**, 1–20.
- Bommarco, R. & Banks, J.E. (2003) Scale as modifier in vegetation diversity experiments: effects on herbivores and predators. *Oikos*, **102**, 440–448.
- Campos-Navarrete, M.J., Munguía-Rosas, M., Abdala-Roberts, L., Quinto-Cánovas, J. & Parra-Tabla, V. (2015) Effects of tree genotypic and species diversity on the arthropod community associated with big-leaf mahogany. *Biotropica*, **47**, 579–587.
- Cardinale, B.J., Matulich, K.L., Hooper, D.U., Byrnes, J.E., Duffy, E., Gamfeldt, L. *et al.* (2011) The functional role of producer diversity in ecosystems. *American Journal of Botany*, **98**, 572–592.
- Castagnyrol, B., Giffard, B., Péré, C. & Jactel, H. (2013) Plant apparency, an overlooked driver of associational resistance to insect herbivory. *Journal of Ecology*, **101**, 418–429.
- Dalin, P., Ågren, J., Björkman, C., Huttunen, P. & Kärkkäinen, K. (2008) Leaf trichome formation and plant resistance to herbivory. *Induced Plant Resistance to Herbivory* (ed. by A. Schaller), pp. 89–115. Springer, Dordrecht, The Netherlands.
- Frederickson, M.E., Ravenscraft, A., Arcila-Hernández, L.M., Booth, G., Astudillo, V. & Miller, G.A. (2013) What happens when ants fail at plant defence? *Cordia nodosa* dynamically adjusts its investment in both direct and indirect resistance traits in response to herbivore damage. *Journal of Ecology*, **101**, 400–409.

- Grossman, J.J., Vanhellefont, M., Verheyen, K. *et al.* (2018) Synthesis and future research directions linking tree diversity to growth, survival, and damage in a global network of tree diversity experiments. *Environmental and Experimental Botany*, **152**, 68–89. <https://doi.org/10.1016/j.envexpbot.2017.12.015>.
- Haddad, N.M., Crutsinger, G.M., Gross, K., Haarstad, J., Knops, J.M. & Tilman, D. (2009) Plant species loss decreases arthropod diversity and shifts trophic structure. *Ecology Letters*, **12**, 1029–1039.
- Jactel, H. & Brockerhoff, E.G. (2007) Tree diversity reduces herbivory by forest insects. *Ecology Letters*, **10**, 835–848.
- Janzen, D.H. (1981) Patterns of herbivory in a tropical deciduous forest. *Biotropica*, **13**, 271–282.
- Kessler, A., Poveda, K. & Poelman, E.H. (2012) Plant-induced responses and herbivore population dynamics. *Insect Outbreaks Revisited* (ed. by P. Barbosa, D. K. Letourneau and A. A. Agrawal), pp. 91–112. Wiley-Blackwell, West Sussex, U.K.
- Kim, T.N. & Underwood, N. (2015) Plant neighborhood effects on herbivory: damage is both density and frequency dependent. *Ecology*, **96**, 1431–1437.
- Kostenko, O., Mulder, P.P.J., Courbois, M. & Bezemer, T.M. (2017) Effects of plant diversity on the concentration of secondary plant metabolites and the density of arthropods on focal plants in the field. *Journal of Ecology*, **105**, 647–660.
- Koricheva, J., Mulder, C.P.H., Schmid, B., Joshi, J. & Huss-Danell, K. (2000) Numerical responses of different trophic groups of invertebrates to manipulations of plant diversity in grasslands. *Oecologia*, **125**, 271–282.
- Langellotto, G.A. & Denno, R.F. (2004) Responses of invertebrate natural enemies to complex-structured habitats: a meta-analytical synthesis. *Oecologia*, **139**, 1–10.
- Littell, R.C., Milliken, G.A., Stroup, W.W., Wolfinger, R.D. & Schabenberger, O. (2006) *SAS for Mixed Models*, 2nd edn. SAS Institute Inc., Cary, North Carolina.
- Makkar, H.P.S. (2003) *Quantification of Tannins in Tree and Shrub Foliage*. Kluwer Academic Publisher, Dordrecht, The Netherlands.
- Marquis, R.J., Ricklefs, R.E. & Abdala-Roberts, L. (2012) Testing the low latitude/high defense hypothesis for broad-leaved tree species. *Oecologia*, **169**, 811–820.
- McArt, S.H. & Thaler, J.S. (2013) Plant genotypic diversity reduces the rate of consumer resource utilization. *Proceedings of the Royal Society B*, **280**, 20130639.
- Meziane, D. & Shipley, B. (1999) Interacting determinants of specific leaf area in 22 herbaceous species: effects of irradiance and nutrient availability. *Plant, Cell and Environment*, **22**, 447–459.
- Mithöfer, A. & Boland, W. (2012) Plant defense against herbivores: chemical aspects. *Annual Review of Plant Biology*, **63**, 431–450.
- Moreira, X., Abdala-Roberts, L., Parra-Tabla, V. & Mooney, K.A. (2014) Positive effects of plant genotypic and species diversity on anti-herbivore defenses in a tropical tree species. *PLoS One*, **9**, e105438. <https://doi.org/10.1371/journal.pone.0105438>.
- Moreira, X., Abdala-Roberts, L., Rasmann, S., Castagnyrol, B. & Mooney, K.A. (2016) Plant diversity effects on insect herbivores and their natural enemies: current thinking, recent findings, and future directions. *Current Opinion in Insect Science*, **14**, 1–7.
- Moreira, X., Glauser, G. & Abdala-Roberts, L. (2017) Interactive effects of plant neighbourhood and ontogeny on insect herbivory and plant defensive traits. *Scientific Reports*, **7**, 4047. <https://doi.org/10.1038/s41598-017-04314-3>.
- Moreira, X., Castagnyrol, B., Abdala-Roberts, L., Berny-Mier y Terán, J.C., Timmermans, B.G., Bruun, H.H. *et al.* (2018) Latitudinal variation in plant chemical defences drives latitudinal patterns of leaf herbivory. *Ecography*. <https://doi.org/10.1111/ecog.03326>.
- Mraja, A., Unsicker, S., Reichelt, M., Gershenzon, J. & Roscher, C. (2011) Plant community diversity influences allocation to direct chemical defence in *Plantago lanceolata*. *PLoS One*, **6**, e28055. <https://doi.org/10.1371/journal.pone.0028055>.
- Onoda, Y., Westoby, M., Adler, P.B., Choong, A.M., Clissold, F.J., Cornelissen, J.H. *et al.* (2011) Global patterns of leaf mechanical properties. *Ecology Letters*, **14**, 310–312.
- Orellana, R., Balam, M., Bañuelos, I., García, E., González, J.A., Herrera, F. *et al.* (1999) *Evaluación climática. Atlas de Procesos Territoriales de Yucatán* (ed. by A. García de Fuentes, J. Córdoba y Ordóñez and P. Chico Ponce de León), pp. 163–182. UADY, Mérida, México.
- Otway, S.J., Hector, A. & Lawton, J.H. (2005) Resource dilution effects on specialist insect herbivores in a grassland biodiversity experiment. *Journal of Animal Ecology*, **74**, 234–240.
- Pennington, T.D. & Sarukhán, J. (2005) *Árboles Tropicales de México. Manual para la identificación de las principales especies*. UNAM, FCE, México.
- Phillips-Rodríguez, E., Powell, J.A., Hallwachs, W. & Janzen, D.H. (2014) A synopsis of the genus *Ethmia* Hübner in Costa Rica: biology, distribution, and description of 22 new species (Lepidoptera, Gelechioidea, Depressariidae, Ethmiinae), with emphasis on the 42 species known from Área de Conservación Guanacaste. *Zookeys*, **461**, 1–86.
- Root, R.B. (1973) Organization of a plant-arthropod association in simple and diverse habitats: the fauna of collards (*Brassica oleracea*). *Ecological Monographs*, **43**, 95–124.
- Rosado-Sánchez, S.I., Parra-Tabla, V., Betancur-Ancona, D., Moreira, X. & Abdala-Roberts, L. (2018) Tree species diversity alters plant defense investment in an experimental forest plantation in southern Mexico. *Biotropica*, **50**, 246–253.
- Roscher, C., Kutsch, W.L. & Schulze, E.D. (2011) Light and nitrogen competition limit *Lolium perenne* in experimental grasslands of increasing plant diversity. *Plant Biology*, **13**, 134–144.
- Salazar, D., Kelm, D.H. & Marquis, R.J. (2013) Directed seed dispersal of piper by *Carollia perspicillata* and its effect on understory plant diversity and folivory. *Ecology*, **94**, 2444–2453.
- Salminen, J.-P. & Karonen, M. (2011) Chemical ecology of tannins and other phenolics: we need a change in approach. *Functional Ecology*, **25**, 325–338.
- SAS (2008) *SAS/STAT 9.2 User's Guide*. SAS Institute Inc., Cary, North Carolina.
- Scherer-Lorenzen, M., Potvin, C., Koricheva, J., Schmid, B., Hector, A., Bornik, Z. *et al.* (2005) The design of experimental tree plantations for functional biodiversity research. *Forest Diversity and Function: Temperate and Boreal Systems*. Ecological Studies, Vol. **176** (ed. by M. Scherer-Lorenzen, C. H. Körner and E. D. Schulze), pp. 347–376. Springer-Verlag, Berlin.
- Schuldt, A., Bruelheide, H., Härdtle, W., Assmann, T., Li, Y., Ma, K. *et al.* (2015) Early positive effects of tree species richness on herbivory in a large-scale forest biodiversity experiment influence tree growth. *The Journal of Ecology*, **103**, 563–571.
- Sinimbu, G., Coley, P.D., Lemes, M.R., Lokvam, J. & Kursar, T.A. (2012) Do the antiherbivore traits of developing leaves in the Neotropical tree *Inga paraensis* (Fabaceae) vary with light availability? *Oecologia*, **170**, 669–676.
- Tack, A.J., Ovaskainen, O., Pulkkinen, P. & Roslin, T. (2010) Spatial location dominates over host plant genotype in structuring an herbivore community. *Ecology*, **91**, 2660–2672.

Accepted 29 May 2018

Associate Editor: Simon Leather