

Effects of phylogeny, leaf traits, and the altitudinal distribution of host plants on herbivore assemblages on congeneric *Acer* species

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Abstract Historical, niche-based, and stochastic processes have been proposed as the mechanisms that drive community assembly. In plant–herbivore systems, these processes can correspond to phylogeny, leaf traits, and the distribution of host plants, respectively. Although patterns of herbivore assemblages among plant species have been repeatedly examined, the effects of these factors among co-occurring congeneric host plant species have rarely been studied. Our aim was to reveal the process of community assembly for herbivores by investigating the effects of phylogeny, leaf traits, and the altitudinal distribution of closely related host plants of the genus *Acer*. We sampled leaf functional traits for 30 *Acer* species in Japan. Using a newly constructed phylogeny, we determined that three of the six measured leaf traits (leaf thickness, C/N ratio, and condensed tannin content) showed a phylogenetic signal. In a field study, we sampled herbivore communities on 14 *Acer* species within an elevation gradient and examined relationships between herbivore assemblages and host plants. We found that herbivore assemblages were significantly correlated with phylogeny, leaf traits, phylogenetic signals, and the altitudinal distribution of host plants. Our results indicate that the

interaction between historical and current ecological processes shapes herbivore community assemblages.

Keywords Functional traits · Host specificity · Maple · Phylogenetic conservatism · Temperate mixed forest

Introduction

The mechanisms and processes of community assembly are a central theme in ecology (Samuels and Drake 1997; Webb et al. 2002; Chase 2003). Among the various factors proposed to affect community assembly, deterministic niche-based processes are proposed to be primary drivers (Diamond 1975; Tilman 1982; Chase and Leibold 2003). In contrast, Hubbell (2001) emphasised the importance of stochastic processes of random dispersal in community assembly through his unified neutral theory of biodiversity and biogeography. Recently, ecologists have acknowledged the importance of the synergetic effects of both deterministic niche-based and stochastic processes (Adler et al. 2007; Chase 2007; Chase et al. 2009). In addition to these contemporary processes, historical and evolutionary processes also influence community assembly (Cornell and Washburn 1979; Ricklefs 1987; Sax et al. 2002; Emerson and Gillespie 2008). The introduction of phylogenetic approaches has allowed ecologists to link short-term local processes to global ones that occur over long evolutionary time scales (Losos 1996; Ackerly 2003; Ricklefs 2004; Graham and Fine 2008; Cavender-Bares et al. 2009).

Plant–herbivore systems have been utilised to examine the effects of both contemporary and historical processes on herbivore community assemblages via evolutionary histories (i.e. phylogenies) and present statuses of host plant species (Novotny et al. 2006; Rasmann and Agrawal 2011).

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Recent applications of phylogenetic techniques at the community level have allowed researchers to investigate the effects of evolutionary processes on the herbivore assemblages of various plant species (Weiblen et al. 2006; Graham and Fine 2008; Cavender-Bares et al. 2009; Rasmann and Agrawal 2011). Specialisation on several host plant taxa is common in most lineages of herbivorous insects (Rasmann and Agrawal 2011). For example, Weiblen et al. (2006) showed that a large proportion of herbivore species found in the tropical rain forest of New Guinea are clade specialists at the genus to family level. Host plant specialisation at the same taxonomic level has also been found in herbivorous beetles in the tropical forests of Panama (Ødegaard et al. 2005). The phylogenetic relatedness of plants can constrain herbivore communities, a trend that can be partially attributed to plant trait similarities between close relatives (Pearse and Hipp 2009; Rasmann and Agrawal 2011). However, other studies have indicated that herbivores prefer less closely related plant species (phylogenetic overdispersion) in a community because of convergence in relevant plant defences (Becerra 1997; Kursar et al. 2009).

It is important to compare closely related plant species when examining effects of host plant relatedness on herbivore assemblages because patterns of similarity among assemblages are generated through phylogenetic and trait-based host constraints (Weiblen et al. 2006; Novotny et al. 2010). Although several studies have investigated herbivore community assemblages of congeneric host plant species (e.g. Becerra 2007; Kursar et al. 2009), detailed phylogenetic comparisons among insect assemblages of a large number of regionally co-occurring congeners are lacking (but see Becerra 2007). The genus *Acer* (maple species) provides an excellent opportunity to explore influences of evolutionary histories of host plants on herbivore assemblages because of its diversity in Japan, its interspecific variation in life history, morphology, and physiology, and the plasticity of these traits (Lei and Lechowicz 1990, 1997; Sipe and Bazzaz 1994, 1995; Tanaka 1995; Ackerly and Donoghue 1998). A few studies have shown the specialisation of several Lepidoptera species on *Acer* plants (Kumata et al. 2013), but only a limited number have examined the herbivore assemblage of *Acer* tree species (e.g. Murakami et al. 2007; Zehnder et al. 2009).

Our aim was to explore the processes generating the structure of herbivore assemblages through an understanding of the effects of phylogenetic relationships among host plant species and other factors. Other important factors potentially affecting community assemblages of herbivores include leaf traits as food resources (Pearse and Hipp 2009) and geographical position (e.g. altitude), which affects assemblages as an environmental filter and also by affecting dispersal of individuals (Beck and Khen 2007; Rominger et al. 2009). Here, the altitudinal distribution of

trees was considered a proxy for current ecological process including environmental factors and dispersal. In this study, we examined leaf trait conservatism among closely related host plant species. Defensive traits were then divided into groups by evaluating representative axes of leaf traits with and without phylogenetic signals. In the field, we collected herbivorous insects from 14 *Acer* species and examined the relationships among phylogeny, leaf traits, and the altitudinal distribution of host plants and herbivore assemblages.

Materials and methods

Study sites

Field surveys were conducted in a mosaic of primary and secondary temperate mixed forest at the University of Tokyo Chichibu Forest in central Japan (35°54'N, 138°49'E). The secondary forest was dominated by *Quercus crispula*, and the primary forest was dominated by *Fagus japonica* at lower altitudes and *Tsuga diversifolia* at higher altitudes (The University of Tokyo Chichibu Forest 2012). The average annual temperature for 1996–2010 was 11.0 °C, and the average annual rainfall was 1,514.2 mm at Tochimoto (The University of Tokyo Chichibu Forest 2012).

We examined 30 species of *Acer* distributed in Japan, including species that are not found in the study area and also some subspecies. We found 23 species during the field survey in Chichibu forest (850–2,000 m in elevation). Leaf traits were measured for each *Acer* species, but because nine species were rare, only 14 were sampled for herbivore community assembly: *Acer amoenum*, *Acer capillipes*, *Acer carpinifolium*, *Acer japonicum*, *Acer maximowiczianum*, *Acer micranthum*, *Acer palmatum*, *Acer pictum* subsp. *dissectum*, *Acer rufinerve*, *Acer shirasawanum*, *Acer sieboldianum*, *Acer tenuifolium*, *Acer tschonoskii*, and *Acer ukurunduense*. The nine rare species were *Acer argutum*, *Acer austral*, *Acer crataegifolium*, *Acer cissifolium*, *Acer diabolicum*, *Acer distylum*, *Acer nipponicum* subsp. *nipponicum*, *Acer pictum* subsp. *pictum*, and *Acer pictum* subsp. *savatieri*. In this study, we followed the taxonomic nomenclature of the YList (Yonekura and Kajita 2003).

Herbivore sampling and identification

Herbivorous insects were collected by hand (Novotny et al. 2002; Murakami et al. 2007) from the foliage of 14 *Acer* species over a period of 3 months (June–August 2011). Twelve individuals from 14 *Acer* species were chosen for sampling. Samples were taken from the foliage of a branch (3- to 5-cm diameter) on each tree during the day. For each species, we recorded maximum altitudinal distribution ranges within the study site (850–2,000 m). We chose trees for sampling

as evenly as possible across the maximum altitudinal distribution for each *Acer* species. All sampling points were recorded using a Global Positioning System, and the altitude of each point was measured. Lepidoptera, Coleoptera, and Hymenoptera larvae were reared in the laboratory on the leaves of their host plant species at a temperature of 25 °C. All insects were identified to the morphospecies.

Plant leaf traits

For 29 *Acer* species, excluding *Acer amamins*, which we were not able to sample, we examined six leaf traits that are known to affect herbivory: leaf thickness (T ; μm), specific leaf area (SLA; g/m^2), water content (WC; % wet weight), C/N ratio (C/N), condensed tannin (CT), and total phenolics (TPh). For each measurement, 20 leaves from five individuals were analysed. The thickness of a fresh leaf was measured with digital callipers, avoiding major leaf veins. To measure the SLA and leaf WC, four leaf disks, 17.2 mm in diameter, were punched as soon as possible after sampling. These disks were weighed to the nearest 0.01 mg and weighed again after drying for 24 h at 60 °C. The C/N ratio was measured using a CN coder (NC-220F; Sumika, Tokyo). Concentrations of total phenolics and condensed tannins were determined colorimetrically following extraction with 50 % acetone for 16 h. The procedure for phenolic measurements followed Price and Butler (1977), and the measurement of condensed tannins followed Broadhurst and Jones (1978). Although the method for phenolic quantification suggested by Price and Butler (1977) has been criticised (Appel 1993), it is still frequently used as a classic index for phenolic content (e.g. Kurokawa et al. 2010; Jackson et al. 2013). Leaf thickness is related to structural defence, and thicker leaves tend to have greater structural resistance against leaf-chewing herbivores (Onoda et al. 2011). The C/N ratio indicates nutritional quality and may act as a defence because low nutritional quality deters herbivores (Mattson 1980; Silva and Batalha 2010). N is a limiting nutrient for many terrestrial organisms, and low levels of N increase feeding time and therefore increase exposure to natural enemies and energy expended on consuming and processing food (Lavoie and Oberhauser 2004; Craine 2009; Silva and Batalha 2010). Tannin is an organic, N-free chemical defence that binds with proteins, reducing N available to herbivores (Bergvall and Leimar 2005; Craine 2009; Silva and Batalha 2010).

Plant phylogeny

A phylogenetic analysis of 30 species of Japanese *Acer* was undertaken based on sequences of the chloroplast genes *rbcl*, *matK*, *trnL-F*, and *rpl16*. In addition, we included three sequences for three outgroups: *Dipteronia sinensis*, *Aesculus hippocastanum*, and *Koeleruteria paniculata*. Total genomic DNA was isolated from freeze-dried leaves using a DNeasy

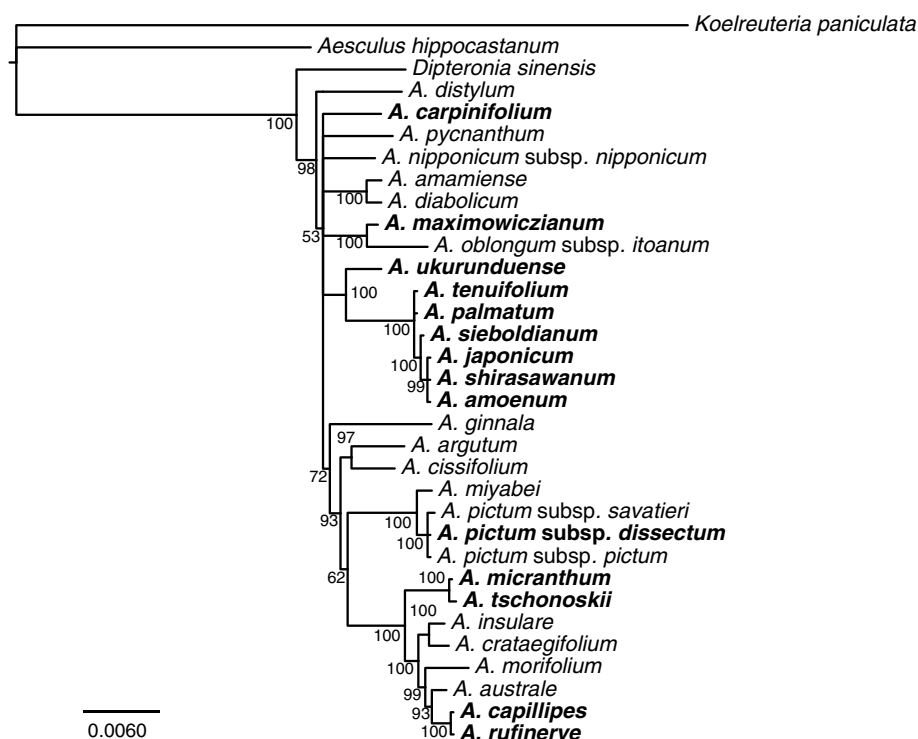
Plant Mini kit (Qiagen, Hilden, Germany). The polymerase chain reactions (PCRs) followed standard protocols. Reaction products were purified with a QIAquick PCR purification kit (Qiagen), and cycle sequencing was performed with BigDye Terminator cycle sequencing kits (Applied Biosystems, Foster City, CA). The dye terminators were removed using ethanol precipitation. Purified sequencing reactions were run on an ABI 3500 automated sequencer (Applied Biosystems). The primers used to amplify the *rbcl* gene were 1F of Fay et al. (1997) and 1460R of Olmstead et al. (1992). For cycle sequencing, they were supplemented by the internal primers 600F and 800R (Kocyan et al. 2007). For the *trnL* intron and adjacent *trnL-F* spacer, the primers c, d, e, and f of Taberlet et al. (1991) were used. For the *rpl16* intron, we used the primers 71F of Jordan et al. (1996) and 1067F of Asmussen et al. (1999), and for the *matK* gene, we used the primers 3F and 1R (K. J. Kim, unpublished data). Forward and reverse reads were obtained for all samples. Sequences were edited with Mega (version 5.05; Tamura et al. 2011) and aligned using mafft (version 6.901; Katoh and Toh 2008). Bayesian analysis was performed using MrBayes (version 3.2.1; Huelsenbeck and Ronquist 2001; Ronquist and Huelsenbeck 2003) with 500,000 generations of Markov chain Monte Carlo chains and sampling of one tree every 100 generations. Trees were rooted with *D. sinensis*, *A. hippocastanum*, and *K. paniculata* (Fig. 1). We calculated phylogenetic distances for all possible pairs among 14 targeted *Acer* species.

Statistical analyses

The phylogenetic signals were quantified using a generalisation of Abouheif's test (Pavoine et al. 2008) in the R package adephylo. To summarise the multi-dimensionality of leaf traits using a defensive index (DI) (Pearse and Hipp 2012), two independent PCAs were performed, one for traits carrying a phylogenetic signal, and the other for traits not showing a phylogenetic signal. We calculated distance matrices of the DIs of all possible pairs of plant species examined here. Jaccard's dissimilarity indices were calculated for all possible pairs of herbivore assemblages among plant species (Oksanen et al. 2011). Furthermore, Euclidean distance matrices for all possible pairs of plant species were calculated for the first two axes of the PCAs for leaf traits with and without phylogenetic signals. The overlap of altitudinal distribution was also measured by Jaccard's dissimilarity indices (van Jaarsveld et al. 1998). The proportions of intra- to inter-specific leaf trait variation were compared among traits with and without phylogenetic signals using *t*-tests.

Correlations between the dissimilarity of herbivore assemblages and the following variables were tested using Mantel tests (Oksanen et al. 2011): (1) host plant phylogeny (phylogeny), (2) a PCA index of leaf defences with a phylogenetic signal (pDI1 for the first axis and pDI2 for the second axis),

Fig. 1 Bayesian phylogram of 30 species of Japanese *Acer* (plus three outgroups) based on 3,694 nucleotides, excluding gaps from four chloroplast DNA loci. Values *above* the nodes indicate Bayesian posterior probabilities >50 %. The 14 species sampled in this study are shown in *bold*



(3) a PCA index of leaf defences with no phylogenetic signal (nDI1 for the first axis and nDI2 for the second axis), and (4) altitudinal distribution of host plant species (altitude). Then, correlations between the dissimilarity of herbivore assemblages and the variables that showed significant correlations were tested while controlling for the effects of other factors (i.e. plant phylogeny, leaf traits, and the altitudinal distribution of host plants) using partial Mantel tests (Tuomisto and Ruokolainen 2006; Barber and Marquis 2011; Milla and Reich 2011) (Table 3). These provide a test of significance without inflating the probability of type I error caused by the indirect effect of a third factor (Barber and Marquis 2011). The correlations between phylogeny, leaf traits (pDI1, pDI2, nDI1, nDI2), and the altitudinal distribution overlap of host plants were also analysed with a partial Mantel test. All statistical tests were performed in R (R Development Core Team 2007) using the packages adephylo (Jombart et al. 2010), vegan (Oksanen et al. 2011), and prcomp.

Results

We recorded 1,859 herbivore individuals from 279 species. Our data set included members of six major herbivore orders (1,005 individuals from 161 species of Lepidoptera, 627 individuals from 67 species of Hemiptera, 142 individuals from 42 species of Coleoptera, 55 individuals from five species of Orthoptera, 25 individuals from two species of Hymenoptera, and five individuals from two species of Phasmatodea).

Table 1 Phylogenetic signals (Abouheif/Moran's test) of plant leaf traits (Pavoine et al. 2008)

Leaf trait	Moran's <i>I</i>	<i>P</i> -values
Thickness	0.194	0.025
Specific leaf area	−0.002	0.364
C/N ratio	0.209	0.027
Water content	−0.161	0.898
Condensed tannin	0.339	0.002
Total phenolics	0.035	0.226

Bold letters indicate the significance in Phylogenetic signals

The analysis of phylogenetic signals for each leaf trait detected significant signals for *T*, C/N ratio, and CT (Table 1; Fig. S2). No significant phylogenetic signals were detected for any other leaf traits (SLA, WC, and TPh; Table 1; Fig. S2). The PCs of leaf traits with and without phylogenetic signals were calculated to obtain summary variables (Table 2). The proportions of intra- to inter-specific variation in leaf traits did not show any explicit trend in relation to the presence of phylogenetic signals ($P = 0.520$).

Jaccard's dissimilarity among herbivore assemblages was significantly correlated with phylogenetic distance ($r = 0.245$, $P = 0.018$; Fig. 2), distances of the PCA indices of leaf defence with phylogenetic signals (pDI1, $r = 0.205$, $P = 0.032$; pDI2, $r = -0.268$, $P = 0.024$; Fig. 3), and the overlap in altitudinal distributions ($r = -0.330$, $P = 0.003$; Fig. 4), but it was not significantly correlated with distances of the PCA indices without phylogenetic signals (nDI1,

Table 2 Loading of leaf trait variables on the first and second principal components (PCs) for each trait group

Leaf traits with phylogenetic signal	PC1	PC2	Leaf traits with no phylogenetic signal	PC1	PC2
Thickness	0.711	0.288	Specific leaf area	1.000	0.007
C/N ratio	−0.018	−0.315	Water content	0.007	−0.315
Condensed tannin	0.703	−0.904	Total phenolics	−0.005	0.949
Total variance explained (%)	46.6	37.1		75.0	17.4

Table 3 Results of partial Mantel tests of the correlations between community dissimilarity and phylogenetic distance, the distance of a PCA index of leaf defences, and altitudinal overlap, including the third variable as a covariable

	Partial Mantel <i>r</i>	<i>P</i> -values
Phylogeny Leaf traits (pDI1)	0.204	0.048
Phylogeny Leaf traits (pDI2)	0.212	0.035
Phylogeny Altitude	0.296	0.010
Leaf traits (pDI1) Phylogeny	0.152	0.103
Leaf traits (pDI1) Altitude	0.156	0.082
Leaf traits (pDI2) Phylogeny	−0.238	0.043
Leaf traits (pDI2) Altitude	−0.316	0.013
Altitude Phylogeny	−0.368	0.001
Altitude Leaf trait (pDI1)	−0.305	0.003
Altitude Leaf trait (pDI2)	−0.369	0.001

Bold letters indicate the significant effect of phylogeny or leaf trait on community dissimilarity of herbivores

For example, A|B indicates the correlation between A and community dissimilarity with the effect of B

$P = 0.363$; nDI2, $P = 0.256$, Fig. 3). Partial Mantel tests detected no significant correlation between assemblage dissimilarity and the distance of the index of leaf defences of host plants under the control of phylogenetic distance (pDI1, $r = 0.152$, $P = 0.103$) or altitudinal distribution overlap (pDI1, $r = 0.156$, $P = 0.082$), but significant correlations were observed for the other combinations (Table 3). The dissimilarity of herbivore assemblages increased as the phylogenetic distance of host plants increased and as the overlap of altitudinal distribution decreased under the control of the other factors (Table 3). The dissimilarity of herbivore assemblages increased with the distance of the PCA index of leaf defences (pDI2) under the control of the other factors (Table 3). The correlations among phylogeny, the PCA index of leaf defences (pDI1), and the overlap of altitudinal distribution of host plants were also significant (Table 4; Fig. 5).

Discussion

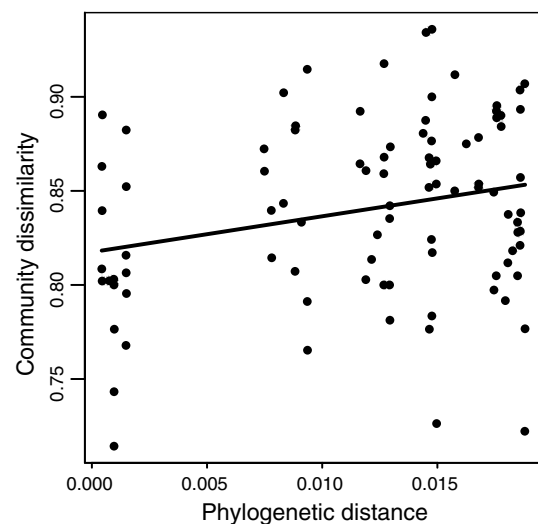
The effect of host plant phylogeny on the herbivore assemblages of congeneric *Acer* species was clearly demonstrated in this study. Weiblen et al. (2006) reported similar effects

Table 4 Results of partial Mantel tests of the correlations among phylogenetic distance, leaf trait distance, and altitudinal distribution overlap

	Partial Mantel <i>r</i>	<i>P</i> -values
Phylogeny-Leaf traits (pDI1) Altitude	0.280	0.029
Phylogeny-Leaf traits (pDI2) Altitude	−0.156	0.103
Altitude-Leaf traits (pDI1) Phylogeny	−0.215	0.034
Altitude-Leaf traits (pDI2) Phylogeny	−0.074	0.255

Bold letters indicate the significant effect of factors examined

For example, A-B|C indicates the portion of B that is explained by A with the effect of C as a co-variate

**Fig. 2** The relationship between phylogenetic distance and community dissimilarity (Pearson $r = 0.245$, $P = 0.018$; Mantel test)

of host plant phylogeny at the genus to family level. However, our results reveal finer level segregation of herbivore assemblages among congeneric, closely related host plant species as a result of historical processes. The decline in similarity of herbivore assemblages with increasing phylogenetic distance among host plant species reflects the phylogenetic conservatism of host plant selection: herbivores tend to feed on the same host lineages as their ancestors (Weiblen et al. 2006).

Host plant phylogeny both directly and indirectly affected the structure of herbivore assemblages.

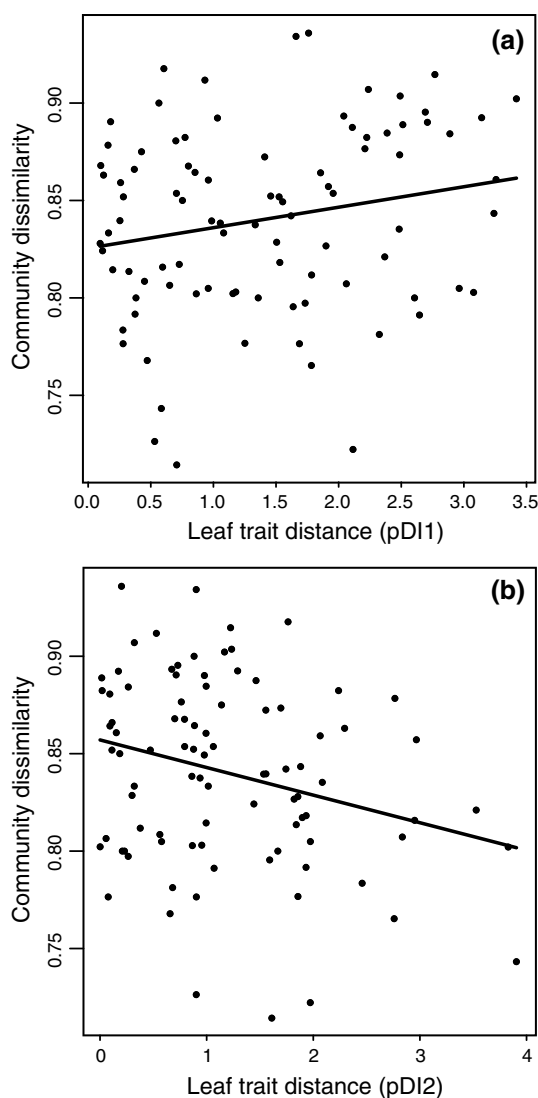


Fig. 3 Relationships between community dissimilarity and the distances of the indices of leaf defences with a phylogenetic signal for **a** the first principal component (PC) axis, pDI1, and **b** the second PC axis, pDI2 (**a** Pearson $r = 0.205$, $P = 0.032$; **b** Pearson $r = -0.268$, $P = 0.024$, Mantel tests)

Phylogeny is an integrated measure of species traits (Pearse and Hipp 2009; Rasmann and Agrawal 2011); thus, the variation in herbivore assemblages not explained by the leaf traits examined here (i.e. the direct effect of plant phylogeny) might be caused by plant traits that were not measurable in this study (Pearse and Hipp 2009). For example, Webster et al. (2010) showed that several synthetic chemicals (i.e. characteristic blends of volatile compounds) within plants are utilised by herbivores to detect host foliage. In another study, Agrawal (2011) showed that the density of leaf trichomes also had an effect on herbivorous insects. These variations in plant

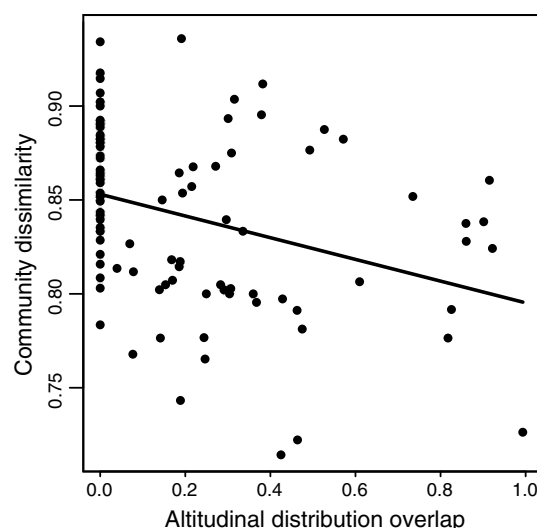


Fig. 4 Relationships between the altitudinal distribution overlap and community dissimilarity (Pearson $r = -0.330$, $P = 0.003$; Mantel test)

traits likely affect the host plant utilisation of herbivorous insects synergistically.

In this study, only the leaf traits showing a phylogenetic signal affected the structure of herbivore assemblages (Table 1; Fig. 3; supplementary Fig. S1). Defensively effective traits, i.e. leaf thickness, C/N ratio and tannin contents, were better conserved than non-effective traits, i.e. SLA, water content and phenolic content. Agrawal et al. (2009) also showed stronger phylogenetic signals in defensive traits (cardenolides, latex, and trichomes) than in the other ones for the milkweed (*Asclepias*) species. Recently, phylogenetic signals of leaf traits were reported in a variety of plant lineages (Ackerly 2009; Rasmann and Agrawal 2011; Pearse and Hipp 2012). Agrawal and Fishbein (2006) also mentioned that defence traits appear to be convergent at lower phylogenetic levels, whereas they appear to be more conserved at higher levels. They showed that Defence traits are not congruent with phylogeny, indicating phylogenetic overdispersion, in genus *Asclepias*. However, in the present study, Defence traits were conserved among congeneric *Acer* species, which showed slower trait changes in these. Thus, we have to seek an explanation for the different pace in trait change among plant taxa in relation to their differences in Defence strategy and their life history.

Recently, many studies have reported a significant covariation among plant defensive traits (Agrawal and Fishbein 2006; Pearse and Hipp 2012): the “plant defence syndrome” (Kursar and Coley 2003; Agrawal and Fishbein 2006). Dimensional reduction approaches (e.g. PCA) have typically been used to examine the plant defence syndrome as a defensive strategy against herbivores (Fine et al. 2006;

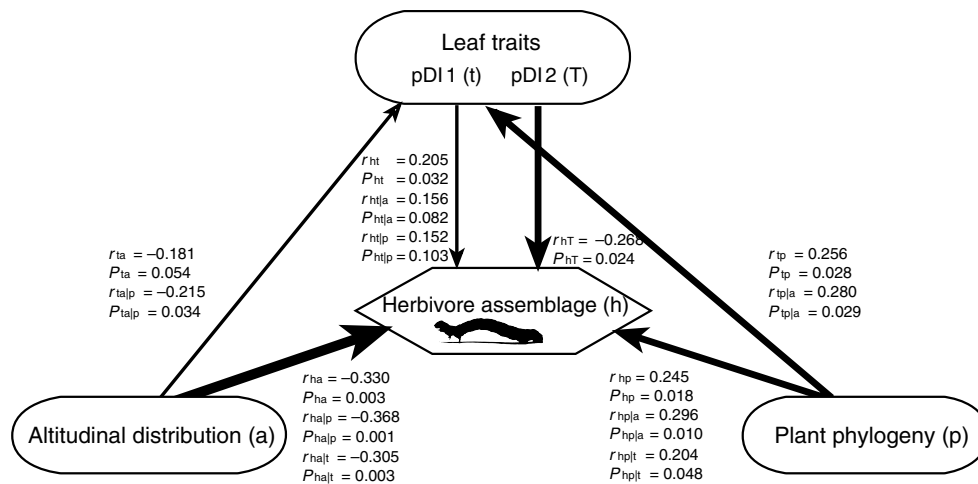


Fig. 5 A scheme to explain the community assembly of herbivores on congeneric tree species. Relative importance of plant phylogeny, altitudinal distribution, and leaf traits are shown. The simple correlations (r_{yx} , P_{yx} where y is the response and x is the predictor) and partial correlations ($r_{yx|w}$, $P_{yx|w}$ where y is the response, x is the pre-

dictor, and w is a potential covariate whose effect on both y and x is accounted for before assessing the correlation) of each path, were estimated. The r - and P -values of the correlations shown are averaged over bootstrap replicates. The width of each path arrow is proportional to the strength of the relationship

Barber and Marquis 2011; Pearse and Hipp 2012). In this study, we confirmed the effect of the *Acer* phylogeny on the set of defensive leaf traits identified by PCA (Table 4). Although both the first and second PCA indices of leaf defence traits with phylogenetic signals (pDI1 and pDI2) showed significant correlations with the herbivore assemblage structure, the effect of the first index (pDI1) was cancelled when we included the effects of phylogeny and the altitudinal distribution of host plants (Table 3). This suggests evolutionary conservation for these leaf traits. On the other hand, the effect of the second PCA index of leaf defences (pDI2) was independent of phylogeny and the altitudinal distribution of host plants (Figs. 3, 4), indicating an independent effect of these leaf traits (pDI2) on herbivore assemblages. However, the distance of pDI2 showed a negative correlation with the dissimilarity of the herbivore assemblages (Table 3), which is more complicated to interpret. One possible explanation for this phenomenon is competitive exclusion among ecologically similar herbivore species (Cavender-Bares et al. 2009). If closely related herbivore species select similar leaf traits when selecting food resources, they may not be able to coexist due to competition (Cavender-Bares et al. 2009).

An altitudinal gradient in the composition of herbivore assemblages (Fig. 4) may be caused by an environmental gradient following altitude. The montane zone contains gradients of a variety of factors, e.g. temperature, soil fertility, risk of photodamage from ultraviolet-B radiation, and precipitation (Preszler and Boecklen 1996; Darrow and Bowers 1997). Simultaneously, the difference in the altitudinal distribution of individual trees implies a spatial gap among herbivore populations, resulting in dispersal limitations.

Because host plants are distributed patchily within a forest, the limited dispersal ability of herbivores might also affect the magnitude of the beta diversity of herbivore assemblages (Hanski 1999; Novotny and Weiblen 2005).

In this study, we investigated the complicated effects of phylogeny, leaf traits, and the altitudinal distribution of host plants on the structure of herbivore assemblages among congeneric maple tree species (Fig. 5). Both historical (phylogeny) and current (e.g. the spatial distribution or environmental filters) ecological processes may have created the existing herbivore assemblage structure. Pearse and Hipp (2012) examined the influence of phylogeny and geographical distribution on leaf traits, and demonstrated that plant defences track the abiotic environment slowly over macroevolutionary time. In our study, we found evidence that these variations in leaf traits following plant phylogeny leave a signature on the herbivore assemblages that feed on them. Specifically, leaf traits with a phylogenetic signal affected herbivore assemblages. However, this was only identified in local processes (i.e. with narrow environmental gradients and geographical ranges). We must further examine the interactions among herbivorous insects, leaf traits, phylogeny, and the geographical distribution of host plants in a larger-scale study and over evolutionary time to understand how herbivore assemblages are constructed, and why herbivorous insects have become one of the most diverse groups in nature.

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