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LETTER

Turnover of plant lineages shapes herbivore phylogenetic beta diversity along ecological gradients

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Abstract

Understanding drivers of biodiversity patterns is of prime importance in this era of severe environmental crisis. More diverse plant communities have been postulated to represent a larger functional trait-space, more likely to sustain a diverse assembly of herbivore species. Here, we expand this hypothesis to integrate environmental, functional and phylogenetic variation of plant communities as factors explaining the diversity of lepidopteran assemblages along elevation gradients in the Swiss Western Alps. According to expectations, we found that the association between butterflies and their host plants is highly phylogenetically structured. Multiple regression analyses showed the combined effect of climate, functional traits and phylogenetic diversity in structuring butterfly communities. Furthermore, we provide the first evidence that plant phylogenetic beta diversity is the major driver explaining butterfly phylogenetic beta diversity. Along ecological gradients, the bottom up control of herbivore diversity is thus driven by phylogenetically structured turnover of plant traits as well as environmental variables.

Keywords

Butterflies, functional diversity, plant defence, specific leaf area, leaf palatability, phylogenetic conservatism.

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INTRODUCTION

Dissecting and interpreting drivers of ecological community structure remains a major challenge in biology (Futuyma & Agrawal 2009; Ricklefs 2010; Tilman *et al.* 1997). Theory predicts that the composition of herbivores communities should be related to host plant availability in a predictable manner (Siemann *et al.* 1998; Schaffers *et al.* 2008; Basset *et al.* 2012). Particularly, more diverse plant communities have been postulated to represent a larger resource-space that is able to sustain a more diverse assembly of herbivore species (Tilman *et al.* 1997). Several examples have shown that higher levels of plant species richness are frequently associated with increases in herbivore diversity (Siemann *et al.* 1998; Scherber *et al.* 2010), but plant and herbivore richness relationships may also be weak or absent (Hawkins & Porter 2003). Such contrasting results might arise from the fact that measures of plant species richness do not always accurately reflect the functional space available to herbivores. Integrating more proximal phenotypic measures of plant functional diversity is expected to yield better predictions of herbivore diversity and composition (Dinnage *et al.* 2012). Several studies have shown that phylogenetically conserved plant traits determine herbivore colonisation, suggesting that measures of plant phylogenetic diversity may also be relevant for predicting herbivore richness (Ødegaard *et al.* 2005; Weiblen *et al.* 2006; Yguel *et al.* 2011; Whitfeld *et al.* 2012). Investigating both functional trait and diversity within a phylogenetic framework should thus allow to tease apart whether the traits of importance are phylogenetically conserved or not, and helps to narrow down the potential processes that might be shaping observed community structure.

The repeated and ongoing evolution of novel plant resistance traits and their neutralisation by herbivores (Ehrlich & Raven 1964) has been suggested as the major driver of diversification of plants and herbivores (Farrell *et al.* 1991), and resulting in lineages with phylogenetically structured defence and neutralisation pathways (Fordyce 2010). Evidence of these reciprocal evolutionary pathways (Berenbaum & Feeny 1981; Wheat *et al.* 2007; Fordyce 2010) has supported the view that phylogenetic information is imbedded in the ways plant–herbivore interactions shape community structure and evolution (Agrawal 2007). However, herbivore community mirroring plant community structure is expected to arise only if consumers have phylogenetic structure of host specialisation in their diets, which may be a characteristic shared by many herbivores food webs (Ødegaard *et al.* 2005; Weiblen *et al.* 2006; Futuyma & Agrawal 2009).

It is also increasingly recognised that how successfully a plant is defended may depend on the effects of many traits acting in concert (Agrawal 2007); hence, a phylogenetic approach has the advantage of providing a somewhat synthetic measure of many different phylogenetically conserved defence and resource traits, generally too complex to be obtained for a large number of species (Agrawal 2007). Recent findings have highlighted the role of plant phylogenetic relationship in structuring herbivore communities (Dinnage *et al.* 2012). However, no studies have simultaneously investigated how environmental variables, plant functional and phylogenetic community structures affect the composition of herbivore communities along ecological gradients.

Plant traits, including resistance strategies, co-vary with the abiotic environment (Rasmann & Agrawal 2011), and thus, investigation of their effect on herbivore community should account for environ-

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mental variation (Pellissier *et al.* 2012a). For instance, slow growing plants, which typically have evolved in low resource environments, will invest relatively more in resistance than phylogenetically related fast-growing species that evolved in high resource environments (Fine *et al.* 2004). In addition, recent evidence suggests that plants evolving in environments with low herbivore abundance, such as high elevation or latitude, typically display a lower level of resistance (Pellissier *et al.* 2012a). The abiotic environment may also influence plant phenotypic characteristics that indirectly impact herbivores, such as the evolution of trichomes or waxes that primarily evolved as an adaptation to dry climate (Agrawal 2007). Thus, herbivore community and trait variation should be investigated along broad environmental gradients to compare the effect of biotic and abiotic factors on herbivore community structure (Cavender-Bares *et al.* 2009).

Here, using a comprehensive spatial dataset of plant and herbivore communities, we compare the variation in abundance, composition and phylogenetic structure within (alpha) and across (beta) 192 plant and herbivore communities (butterflies *sensu* Heikkilä *et al.* 2012) distributed along broad environmental gradients in the Swiss Alps. Butterflies interact with plants in antinomic ways (i.e. they feed on plants as larvae and act as pollinators during adulthood), and are often specialised on a restricted host plant range as larvae (Fordyce 2010; Vila *et al.* 2011). Investigating the variation across communities (beta) through pair-wise comparisons can yield more subtle information for understanding the drivers of community structure (Graham & Fine 2008). We reconstructed both a plant species and a butterfly species phylogeny, measured several commonly used plant palatability related traits [specific leaf area or (SLA), leaf dry matter content (LDMC), leaf C/N ratio, Wright *et al.* 2004)], and asked whether plant trait variation, phylogenetic diversity or climate variables better predict three facets of herbivore communities: species richness, abundance and phylogenetic diversity. Because the relationship between plant and herbivores is theorised to be phylogenetically structured (Berenbaum & Feeny 1981; Fordyce 2010), we expect a positive effect of plant phylogenetic diversity on herbivore phylogenetic diversity. However, because they directly impact plant palatability and resource availability to herbivores, leaf functional traits should correlate with herbivore community structure, regardless of the phylogenetic conservatism of those traits. Whether host plant phylogeny can predict the diversity of herbivores in communities will ultimately depend on the phylogenetic conservatism or lability of plant palatability traits (Fine *et al.* 2006).

MATERIALS AND METHODS

Field sampling and plant trait collection

The study area spans from 1000 m to 3210 m a.s.l. and is located in the Western Swiss Alps (Figure S1 in Supporting Information). Along the elevation gradient, several grasslands types are found that have site-specific climatic and land-use conditions. Site selection, only including open vegetation grasslands, was conducted following a balanced stratified random sampling design based on elevation, slope and aspect in open grasslands (Hirzel & Guisan 2002). Between May and September 2009 and 2010, 192 plots of 50 m × 50 m were sampled across the whole study area (700 km^2, Figure S1). All butterfly species belonging to the Papilionoidea super-family (*sensu* Heikkilä *et al.* 2012) were monitored. We collected adult specimens instead of caterpillars because they are more conspicuous, more reliable to survey and easier to identify. Most butterfly species occupy the same habitat throughout their entire life cycle. In addition, we excluded species known to be very mobile (10 species out of 122), as those are not necessarily related to the environments or resources where they occur and would therefore add noise to our analyses. During each visit, we also counted the number of butterfly individuals as a measure of abundance at each site.

Plant species were exhaustively inventoried in 4 m^2 subplots at the centre of each plot, making sure that the vegetation was representative of the entire 2500 m^2 area where butterfly were monitored. Species cover was estimated visually into 10 classes: <0.1%, 0.1–0.5%, 0.5–1%, 1–3%, 3–5%, 5–15%, 15–25%, 25–50%, 50–75% and >75%. Leaf traits were gathered for all the 231 most frequent and abundant vascular plant species occurring in the study area, and span major portions of the angiosperm phylogeny (Baraloto *et al.* 2010; Pellissier *et al.* 2012b). Specific leaf area was calculated as the ratio of the leaf surface to its dry mass expressed in $mm^2\ mg^{-1}$. The LDMC was measured as the ratio of the leaf dry mass to its water-saturated fresh mass (in $mg\ g^{-1}$); this measurement is related to the average density of the leaf tissue. C/N ratio was obtained from the combustion of dried leaves using an elemental analyser. Given the sampling effort of collecting traits for a large number of species, this study does not explicitly incorporate within-species trait variation and analyses may underestimate leaf trait diversity (Swenson *et al.* 2011).

For each site, we extracted climatic predictors known to influence plant and insect life-history traits (Hodkinson 2005). All predictors were calculated from meteorological stations using a Digital Elevation Model (DEM) at 100 m resolution. Values for degree-days, moisture index (computed as the difference between precipitations and evapotranspiration) and number of freezing days were interpolated following Zimmermann & Kienast (1999). Solar radiation values were calculated using the tool implemented in ArcGIS 10. For each plot location, we extracted the values of environmental variables from associated environmental layers.

Butterfly and plant phylogenetic analyses

Phylogenetic relationships of all butterfly species found in the study area were inferred using DNA sequences. Five Thyrididae and two Callidulidae species were included as outgroups. Sequences were obtained from GenBank and included two nuclear markers (EF1-alpha, Wgl) and four mitochondrial markers (16s rRNA, COI, NDH1, NDH5). Sequences were aligned in Seaview (Gouy *et al.* 2010) which provided a final concatenated matrix of 4310 base pairs. BEAST v 1.6.1 (Drummond & Rambaut 2007) was used to infer phylogenies in a Bayesian ultrametric approach. As part of the analysis, internal node ages were calibrated using fossil data. Five nodes were thus constrained based on dates estimated by previous studies or on the fossil record following Heikkilä *et al.* (2012). All priors for the fossils were drawn from the mean and standard deviation of a lognormal distribution, with hard and soft bounds chosen to reflect the 95% confidence in the fossils. The tree prior was set using Yule speciation model. The searches used an uncorrelated relaxed molecular clock, and were run for 80 000 000 generations, sampling parameters every 1000 trees. Convergence was assessed from the ESS and model parameters in Tracer 1.4. A burn-in of the

initial 40 000 trees was applied, and the final dated tree was reconstructed from the remaining 40 000 trees.

Phylogenetic relationships of the 231 most frequent and abundant plant species sampled from the vegetation plots were inferred using DNA sequences extracted from collected vegetal material (see Pellissier *et al.* 2012a for further details). Additional sequences were obtained from GenBank comprised rbcL and matK markers. We included two gymnosperm outgroups: *Abies alba* and *Picea abies*. Sequences were aligned in Seaview (Gouy *et al.* 2010). The final concatenated matrix consisted of 3092 nucleotide base pairs. A time-calibrated ultrametric tree was constructed from nine published fossils using the BEAST software (Drummond & Rambaut 2007). We used the same parameters as described above for butterfly phylogenetic reconstructions. The resulting phylogenetic trees were checked against the Angiosperm Phylogeny Group tree for accepted relationships among plant orders and families.

Phylogenetic signal of all traits were assessed following Blomberg *et al.* (2003) using the full plant phylogeny. Bloomberg's K statistic compares the observed distribution of trait values to expectations under a Brownian motion model of trait evolution, and was calculated using Picante (Kembel *et al.* 2010) in R (R Development Core Team 2012). K values close to 0 indicate a random distribution of trait values with respect to the phylogeny, while K values close to 1 signify trait evolution consistent with a Brownian motion model of evolution (Blomberg *et al.* 2003). The significance of this test was derived by comparing the observed K values to a null distribution generated from 999 randomisations of trait values across the tips of the phylogenetic tree (Kembel *et al.* 2010).

To detect phylogenetic structure in trophic association between butterflies and host plants, we used ParaFit, a method originally developed for the coevolutionary analyses of hosts and parasites (Legendre *et al.* 2002). ParaFit, implemented in the ape R package, is a matrix permutation test of co-speciation, which aims to test the signal of a phylogenetic structure in plant–herbivore association. This approach has an advantage over tree-based methods because it can accommodate multiple butterfly species per plant lineage. Distance matrices for butterflies and plants were derived from the reference phylogenies using the 'cophenetic' function in the ape R package. Association between host plant species and butterfly were extracted from a comprehensive literature survey (Pellissier *et al.* 2012a,b).

Butterfly alpha diversity

Three facets of butterfly diversity at sites (butterfly species richness, phylogenetic diversity and total abundance) were compared with factors at each site, plant species richness, functional and phylogenetic diversities of plant communities and climate. Butterfly species richness at each site was calculated as the sum of all species inventoried, while butterfly abundance was calculated as the average number of individuals at each site per visit irrespective of species identity. The phylogenetic alpha diversity of butterfly assemblages was calculated using the standardised effect size (SES) of the mean pairwise phylogenetic distance (MPD) of all species in a community (equivalent to -NRI) as implemented in the picante R package. We considered only butterfly presence and absence, because species specific abundance measurements are less reliable (Pellissier *et al.* 2012a). To compute SES MPD we built null distributions by randomly reshuffling the tip labels on the butterfly phylogeny. This procedure of randomisation evaluates the effect of phylogeny while holding other factors constant and thus preserves important properties over the study area (e.g. species prevalence, species richness). It is equivalent to testing the null hypothesis that phylogeny is not an important component to explaining butterfly community structure when compared to baseline change in taxonomic composition.

To evaluate which, if any, factors predict butterfly phylogenetic diversity, we computed the plant species richness and measured community weighted mean leaf traits and diversity of leaf traits (for SLA, LDMC, N, C/N) using the modified version of the Rao index proposed by de Bello *et al.* (2010). We calculated the plant phylogenetic alpha diversity as the mean pairwise phylogenetic distance (MPD) and the mean nearest taxon distance (MNTD – not compared to a null-model) using the *mpd* and *mntd* function, respectively, in the picante R package (Kembel *et al.* 2010). Plant species per cent cover was considered in the diversity measures. Because we did not sample all plant taxa for trait data and not every species was included in our plant phylogeny, we only consider those communities for which at least 80% of the total vegetation cover included taxa in our phylogeny and functional dataset (excluding 32 plots). Because not all plant species interact with butterfly in the study area, we also computed plant functional and phylogenetic diversities with the subset of plants known to interact with butterfly from a literature survey (Pellissier *et al.* 2012a,b).

We related butterfly community species richness, abundance and phylogenetic diversity to abiotic variables and descriptors of the plant communities using a linear model with a Gaussian distribution. To improve normality and homoscedasticity of the residuals, we used a log transformation when appropriate. The best fit model was chosen based on the minimum AIC score. From the most parsimonious model, we calculated the total explained variance as well as the variance explained by the abiotic and biotic components.

Butterfly beta diversity

We related variation in butterfly abundance, composition and phylogenetic diversity among sites (through multiple pair-wise comparisons) to variation in the composition, functional, and phylogenetic beta diversities and climatic differences of plant communities. As for alpha diversity, three facets of butterfly diversity between sites were investigated. We measured turnover in butterfly abundance using the *dist* function in R, taxonomic composition using the Jaccard similarity index implemented in the *vegan* R library, and butterfly phylogenetic beta diversity using the *comdist* and *comdistnt* functions in the picante R package (Kembel *et al.* 2010). These functions, respectively, calculate MPD and MNTD separating taxa across communities. We computed SES of MPD and MNTD in the same way as for the NRI, by calculating a standardised effect size as described in Swenson *et al.* (2011).

For plant communities, we measured taxonomic beta diversity using the Jaccard similarity index and functional beta diversity using the modified version of the Rao quadratic entropy presented in de Bello *et al.* (2010). We measured plant phylogenetic beta diversity (MPD and MNTD – not compared to a null-model) using the *comdist* and *comdistnt* functions in the picante R package (Kembel *et al.* 2010). We related the species abundance, taxonomic turnover and SES phylogenetic beta diversity among butterfly communities to taxonomic, functional and phylogenetic beta diversity among plant communities and abiotic environmental variables. We tested the

relationship with Mantel tests as suggested in Graham & Fine (2008) with 9999 permutations. Because changes in plant functional and phylogenetic structure may explain the same variance as baseline plant taxonomic turnover, we assessed the independence of those factors using partial Mantel tests. We also ran all alpha and beta diversity analyses for the four most representative butterfly families, including Papilionidae, Pieridae, Hesperiidae and Lycaenidae (excluding Papilionidae and Riodinidae).

We finally used a Constrained Analysis of Principal Coordinates (CAP) implemented in the *vegan* package, which allows relating dissimilarity matrices to environmental variables. We related butterfly and plant taxonomic (from Jaccard) and phylogenetic beta diversities (MPD and MNTD obtained from *comdist* and *comdistnt* functions) to degree-days, moisture, number of frost days and solar radiations. We tested the significance using Monte Carlo permutation tests (999 randomised runs). We plotted the CAP results and coloured the dots according to the grassland habitat categories defined using phytosociological analysis (Table S1). All analyses were performed in R (R Development Core Team).

RESULTS

Overall, our field survey yielded 122 butterfly species (14 Hesperiidae, 15 Pieridae, 4 Papilionidae, 1 Riodinidae, 59 Nymphalidae, 29 Lycaenidae), representing more than half of the species currently known to occur in Switzerland (Swiss Biological Records Center, www.cscf.ch). We found 698 plant species and the most diverse inventoried plot contained 63 species. The sampled communities spanned an elevation gradient from 1063 to 3041 m a.s.l. Because we focused on mountain fauna, it did not include the more thermophilous species known for the country. Results for the butterfly phylogenetic inferences are congruent with those obtained by Heikkilä *et al.* (2012), The exception was one single node-incongruence corresponding to the Papilionidae and Pieridae families, which are clustered together in our analysis (Figure S2). As this difference does not have a major influence on branch length, it is unlikely to affect the conclusions of the diversity analyses. The phylogenetic reconstructions for plants produced a well-supported phylogenetic tree with nodes congruent to taxonomic groups defined by the APG III (Figure S3).

Plant–butterfly association and trait conservatism

Parafit test of phylogenetic association resulted in the rejection of random associations between host plant and butterflies ($P < 0.0001$, Fig. 1). This pattern of phylogenetic matching is consistent with our expectation that closely related butterflies prefer to feed on closely related plants and suggests that butterflies are responding to phylogenetically conserved plant traits. Leaf traits were not randomly distributed with respect to the plant phylogeny. Although these traits exhibited an overall low phylogenetic signal, their K values remained higher than expected when the traits values were reshuffled across the tips of the phylogeny. LDMC had the highest signal (K = 0.33, $P = 0.001$), followed by C/N (K = 0.14, $P = 0.001$), SLA (K = 0.12, $P = 0.001$) and N (K = 0.09, $P = 0.08$).

Alpha diversity

Explanatory variables included in the most parsimonious models differed between facets of butterfly communities. For the total abundance of butterfly individuals, we found a positive effect of degree-days, solar radiation and plant richness, but a negative effect of leaf nitrogen variation within the community of plants (Table 1, Fig. 2). Butterflies were less abundant in climatically severe environments and where the nitrogen content of plant leaves was more variable between plant species. For butterfly species richness, we found a positive effect of degree-days, solar radiation and plant species richness, but a negative effect of LDMC diversity (Table 1, Fig. 2). For the SES MPD (phylogenetic alpha diversity) of butterfly communities, we found a positive effect of degree-days, SLA diver-

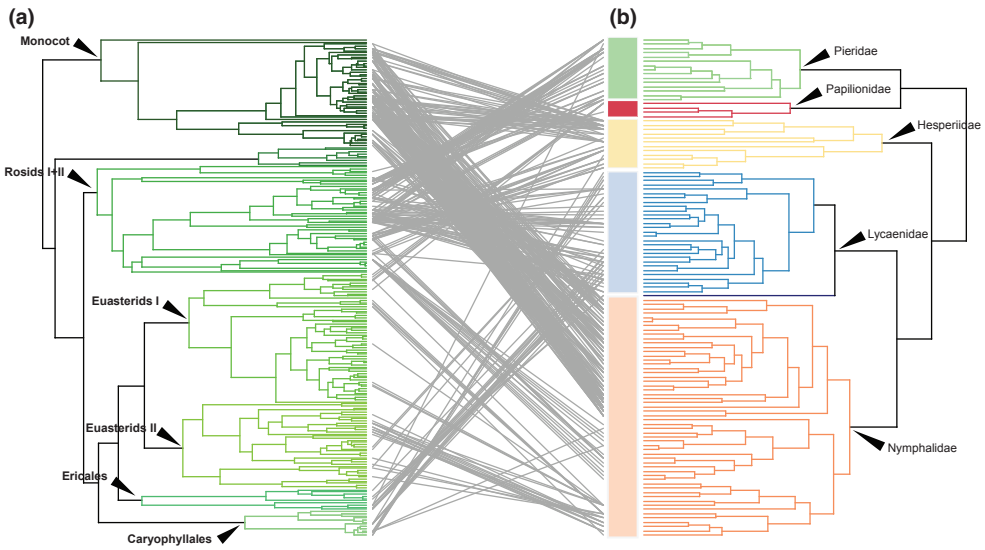

Figure 1 Chronograms of 231 most abundant angiosperm species (a) and of all butterfly species (b) found in the study area. Colours indicate different plant major diversification events and different butterfly families. Bipartite network was inferred from the literature (Pellissier *et al.* 2012a,b). The trophic network of butterflies on angiosperms is highly structured (Parafit test: $P < 0.0001$).

Table 1 Alpha diversity – results of best fit multiple regressions for a) butterfly abundance, b) butterfly phylogenetic diversity of communities measured using the standardised effect size of the mean phylogenetic distance (MPD) and c) butterfly species richness against environmental variables, plant functional traits and plant diversity measures. Estimates for moisture, degree-days and solar radiations are low because of the large values of these variables. Best fitting models were chosen based on minimum AIC score. R^2 of abiotic, biotic and of the total model are provided

Parameter	Estimate	t		
a) Total abundance				
Degree-days	0.002	1.80	*	
Solar radiations	0.001	4.04	*	$R^2 = 0.15$
N diversity	−0.73	−2.26	*	
Plant richness	0.01	2.58	*	$R^2 = 0.14$
	AIC = 267.34			$R^2 = 0.28$
b) Phylogenetic diversity (SES MPD)				
Degree-days	0.001	5.48	*	$R^2 = 0.16$
SLA mean	−3.5	−2.65	*	
SLA mean, 2	2.00	2.28	*	
SLA diversity	0.10	2.50	*	
N mean	0.56	2.11	*	$R^2 = 0.10$
	AIC = 383.4			$R^2 = 0.24$
c) Species richness				
Degree-days	0.01	4.59	*	
Solar radiations	0.001	3.20	*	$R^2 = 0.30$
LDMC diversity	−0.05	−2.11	*	
Plant richness	0.22	4.43	*	$R^2 = 0.20$
	AIC = 799.5			$R^2 = 0.39$

sity and nitrogen community weighted mean and a nonlinear effect of mean SLA with maxima in low and high mean SLA values. This means that the most phylogenetically dispersed butterfly communities occurred in conditions of warm temperature, where SLA values strongly vary among plant species, potentially providing more feeding niches for distinct specialised butterfly lineages. Finally, and contrary to expectation, we found no significant effect of plant phylogenetic diversity (either MPD or MNTD) as a factor structuring herbivore phylogenetic alpha diversity (Table 1, Fig. 2). We found similar results when only plants known to interact with butterfly species were considered in the analysis. Plant phylogenetic diversity (MNTD) was retained in the lowest AIC models of butterfly phylogenetic alpha diversity (SES MPD) and species richness but was statistically not significant (Table S2). We found distinct effect of biotic and abiotic variables on the species richness and phylogenetic alpha diversity (SES MPD) of the four butterfly families: Hesperiidae and Lycaenidae species richness and phylogenetic diversity were correlated to SLA community weighted mean and plant species richness in addition to abiotic variables, while Nymphalidae species richness and phylogenetic diversity were only related to climate (moisture index, Table S3).

Beta diversity

Turnover in abundance of butterfly in communities was mainly driven by climate (degree-days) and plant traits (N and SLA), while plant phylogenetic alpha diversity showed no effect (Table 2). Butterfly taxonomic beta diversity was related to all investigated variables, but overall, plant phylogenetic beta diversity (MPD and MNTD), SLA and climatic variable showed the strongest effects. Butterfly phylogenetic beta diversity measured with the SES MPD was most strongly correlated to plant phylogenetic beta diversity and especially to plant MNTD. Butterfly phylogenetic beta diversity measured with the SES MNTD was also correlated to plant phylogenetic beta diversity but plant traits (C/N, LDMC, N) and climate variables (degree-days and number of freezing days) also showed significant effects. For the three investigated response variables (butterfly total abundance, taxonomic and phylogenetic diversity), partial Mantel tests indicated that the effects of leaf trait and plant phylogenetic turnover were independent of the baseline plant taxonomic turnover. Results were comparable when only plants known to interact with butterfly species were considered in the analysis (Table S4). Finally, family analyses revealed that Lycaenidae and Hesperiidae showed stronger correlation to biotic variables (including plant phylogenetic beta diversity – MPD and MNTD) compared to Nymphalidae and Pieridae (Table S5). The CAP showed a significant effect of the four abiotic variables (degree-days, moisture, solar radiation and number of frost days) on plant and butterflies beta diversities measured both with MPD and MNTD (Fig. 3).

DISCUSSION

There is growing appreciation that long-term co-evolutionary interactions across trophic levels plays a major role in driving the composition and structure of communities (Futuyma & Agrawal 2009; Dinnage *et al.* 2012). Our study provides the first evidence in a natural system that plant phylogenetic beta diversity is correlated with butterfly phylogenetic beta diversity, suggesting that the turnover of host-plant lineages structures herbivore assembly in space.

The importance of abiotic and biotic variables for predicting herbivore community structure varied across measures of herbivore diversity, but also whether diversity was measured within sites (alpha diversity) or between sites (beta diversity). Surprisingly, plant phylogenetic alpha diversity in natural communities was a poor predictor of butterfly phylogenetic alpha diversity, which contrasts with recent experimental results (Dinnage *et al.* 2012). This suggests that in natural communities, a global measure of diversity of plant lineages does not necessarily reflect the total functional space available to herbivores. In natural systems, beta diversity analyses or analyses of more direct functional traits may allow for the detection of more subtle shared structures of communities across trophic levels.

Butterflies are recognised to have radiated through the colonisation of novel host plant clades followed by bursts of diversification (Ehrlich & Raven 1964; Fordyce 2010), and indeed, most butterfly taxa are often associated with particular plant clades (Menken *et al.* 2010; Janz 2011). This is valid for the butterfly families considered in this study, as demonstrated by the strong phylogenetic structure highlighted in the Parafit test (Fig. 1). As a consequence, variation in the availability of plant lineages in space implies a shift in trophic niches available for specialised butterflies and therefore a change in phylogenetic structure as indicated by our results for beta diversity. Because the leaf traits measured showed low phylogenetic signal along our phylogeny, other phylogenetically conserved plant traits, such as plant secondary metabolites structured along the angiosperm phylogeny, are likely to be better predictors of herbivore assembly in space. Plant secondary metabolism is well-established to impact herbivore fitness (Fraenkel 1959). In addition, classes of secondary metabolites are generally conserved along the angiosperm

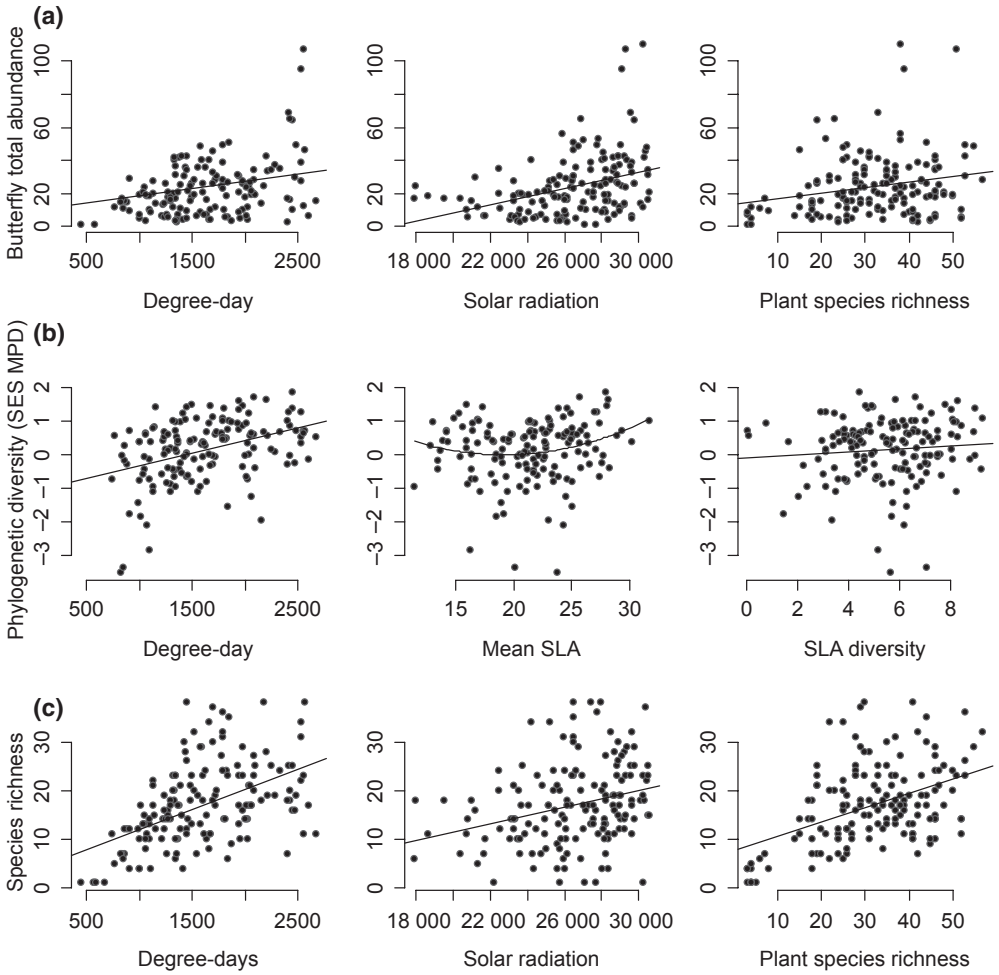

Figure 2 Relationships between (a) total butterfly abundance, (b) phylogenetic diversity of butterfly communities measured using the standardised effect size of the mean phylogenetic distance (SES MPD) and (c) butterfly species richness and environmental variables retained from the best fit models. Each dot represents a community sampled in the study area of the Western Swiss Alps.

phylogeny (Wink 2003). Secondary metabolites are thus a likely good candidate trait to investigate when looking for traits that might predict herbivore structure (Becerra 2007; Agrawal 2007).

All plant lineages did not show the same degree of colonisation by butterflies and several plant clades were even almost fully avoided (e.g. Asteraceae, Fig. 1). Levels and composition of chemical defences that evolved in those lineages may efficiently deter herbivores in this system. However, while the reasons why these phylogenetically diverse lineages are not similarly consumed by herbivores remain speculative, a global measure of plant phylogenetic diversity that accounts for those lineages is likely to overestimate the potential diversity of niches available to herbivores. Indeed, plant phylogenetic alpha diversity was retained in the model when only plants known to interact with butterfly species were considered in its computation. As stressed by Whitfeld *et al.* (2012), our results highlight the limitations of using a phylogenetic approach to study alpha diversity across trophic levels without deeper knowledge of plant traits such as secondary metabolites. Indeed, the importance of particular plant lineages for herbivore assembly may vary across the angiosperm phylogeny (i.e. some clades tend to structure herbivore communities more than others) and this should be taken into account when designing future community phylogenetic analyses. In contrast, investigating the turnover of diversity in space using beta diversity measures is likely to provide a more detailed picture of how plant phylogenetic lineages change in space and, ultimately, structure assemblages at higher trophic level (Graham & Fine 2008).

We found a stronger correlation between butterfly phylogenetic diversity and plant phylogenetic diversity for Lycaenidae and Hesperiidae families. Species of the Lycaenidae and Hesperiidae families are specialised on diverse plant clades (Megens *et al.* 2005; Vila *et al.* 2011; Pellissier *et al.* 2012a,b). Because of the variation in host plant specialisation, shift in the phylogenetic structure of plant communities will affect the composition in those families. In contrast, Nymphalidae family is either composed of highly polyphagous species (e.g. Nymphalinae) or clade with low variability in host plant association (e.g. Satyrinae subfamily feeding on the grass family, Poaceae and Cyperaceae). Hence, the composition of species from those families will be less affected by shift in the composition of plant lineages.

Turnover in total butterfly abundance in space was primarily predicted by leaf trait turnover (N, SLA), while plant phylogenetic turnover showed not effect. Our results agree with Whitfeld *et al.* (2012),

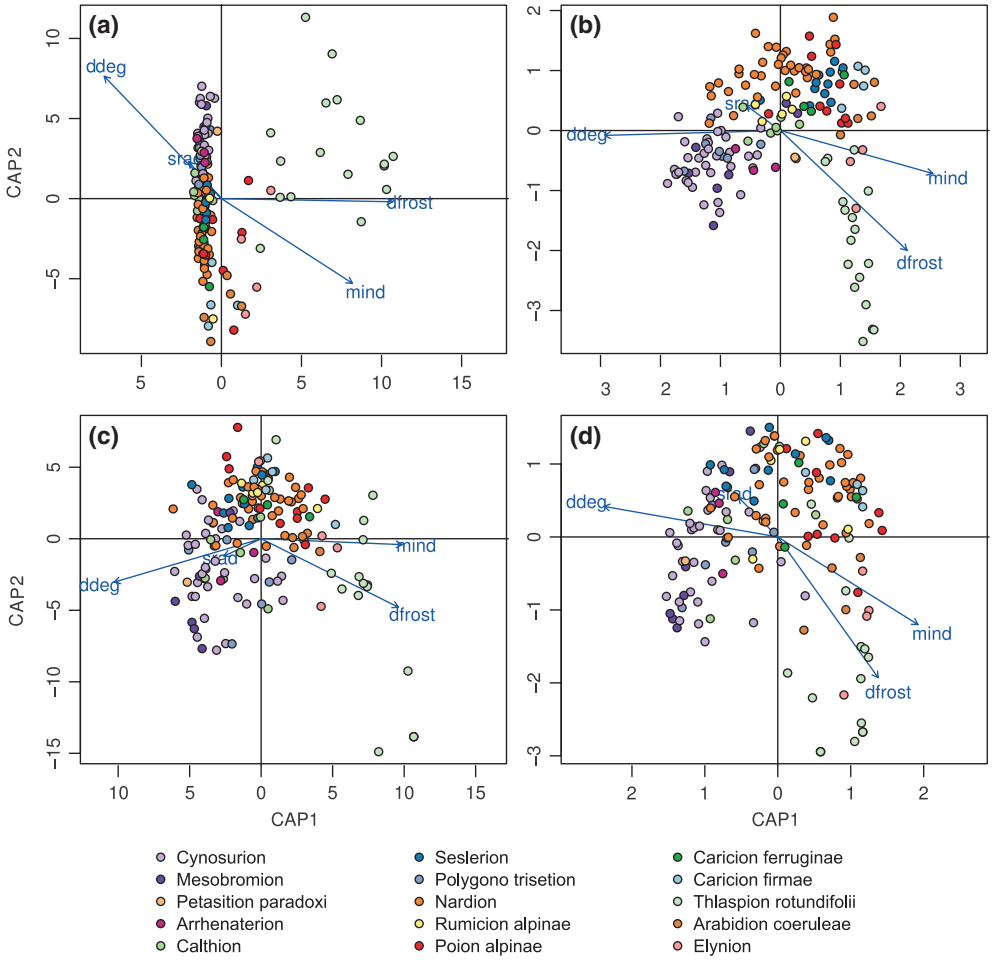

Figure 3 Constrained Analysis of Principal Coordinate analysis for (a) plant phylogenetic beta diversity measured with the mean nearest taxon distance (MNTD, CAP1 = 90.1%, CAP2 = 6.8, 38% total constrained) (b) plant taxonomic beta diversity measured with the Jaccard index (CAP1 = 55.5%, CAP2 = 27.9, 12% total constrained) (c) butterfly phylogenetic beta diversity mean phylogenetic distance (MPD, CAP1 = 48.6%, CAP2 = 22.5, 11% constrained) and (d) butterfly taxonomic beta diversity with the Jaccard index (CAP1 = 62.1%, CAP2 = 24.4, 17% total constrained). ddeg = degree-days; dfrost = frost days; mind = moisture index; and srad = solar radiation.

who found little evidence of phylogenetic signal explaining the overall abundance of leaf chewers in tropical trees, while leaf N content showed a major effect. We hypothesise that phylogenetically conserved metabolites may act as a barrier in determining whether herbivore lineages are able to colonise host plants, due to phylogenetically conserved detoxification mechanisms. In contrast, the abundance of herbivores having neutralised plant chemical defences may rather depend on leaf toughness and leaf resources such as nitrogen. This hypothesis is supported by the meta-analysis of Carmona *et al.* (2011), in which plant morphological and physical traits were shown to be more frequently associated with plant 'susceptibility to herbivores' than secondary metabolites, a measure that do not account for herbivore composition. Hence, different facets of herbivore communities (e.g. total abundance, phylogenetic diversity) are likely to be driven by distinct phenotypic aspects of plant in communities such as plant secondary metabolites or overall plant nutritive value.

Finally, all facets of butterfly community structure were associated with climatic conditions (Fig. 3). Severe climatic conditions impose constraints on herbivore species richness, abundance and phylogenetic diversity. The association between butterfly phylogenetic diversity and degree-days suggests that tolerance to distinct climatic conditions (e.g. colder with more risks of freezing vs. warmer and more stable conditions) is phylogenetically conserved in butterflies (Hawkins & Porter 2003). One caveat of our study is that with our correlative framework, we cannot establish a causal link between plant community metrics and herbivore assembly. However, we assumed that plant community structure is independent of herbivore community structure. Hence, in our view, deterministic conditions (e.g. abiotic environment, human land use or within trophic level interactions such as competition) structure plant communities in this mountain region, which in turn drive herbivore community assembly. However, recent studies suggest that herbivore pressure may also play a role in plant community assembly (Becerra 2007; Kursar *et al.* 2009), and this interpretation cannot be ruled out here. Plant–insect community assembly may therefore be seen as emerging through feedback processes, such as for plant–fungi communities (Bever 1994), and that some of the co-variation found in this study may have emanated from the impact of herbivore on plant phylogenetic structure.

Table 2 Beta diversity – results of the Mantel tests correlating a) turnover in total abundance, b) turnover in taxonomic composition and c) turnover in phylogenetic diversity of butterfly communities measured using the standardised effect size of the mean phylogenetic distance (SES MPD) d) turnover in phylogenetic diversity of butterfly communities measured using the standardised effect size of the mean nearest taxon distance (SES MNTD) changes in climatic as well as plant functional traits across the elevation gradient. When appropriate, partial Mantels tests with the plant taxonomic beta diversity as co-variable is provided

	Mantel r		Partial r	
a) Total abundance				
β plant taxonomic	−0.02	NS		
β plant C/N	0.05	NS	0.05	NS
β plant N	0.11	*	0.11	*
β plant LDMC	0.01	NS	0.01	NS
β plant SLA	0.10	*	0.10	*
β plant phylogenetic (MPD)	0.04	NS	0.01	NS
β plant phylogenetic (MNTD)	−0.02	NS	−0.03	NS
Δ degree-days	0.14	*		
Δ frost days	0.03	NS		
Δ moisture	0.06	NS		
Δ solar radiations	0.03	NS		
b) Taxonomic diversity				
β plant taxonomic	0.30	*		
β plant C/N	0.14	*	0.05	NS
β plant N	0.14	*	0.06	NS
β plant LDMC	0.15	*	0.05	NS
β plant SLA	0.22	*	0.12	*
β plant phylogenetic MPD	0.38	*	0.30	*
β plant phylogenetic MNTD	0.45	*	0.37	*
Δ degree-days	0.56	*		
Δ frost days	0.46	*		
Δ moisture	0.45	*		
Δ solar radiations	0.06	*		
c) Phylogenetic diversity (SES MPD)				
β plant taxonomic	0.15	*		
β plant C/N	0.05	NS	0.01	NS
β plant N	0.01	NS	−0.01	NS
β plant LDMC	0.08	*	0.01	NS
β plant SLA	0.01	NS	−0.001	NS
β plant phylogenetic (MPD)	0.22	*	0.15	*
β plant phylogenetic (MNTD)	0.33	*	0.27	*
Δ degree-days	0.10	*		
Δ frost days	0.30	*		
Δ moisture	0.16	*		
Δ solar radiations	0.001	NS		
d) Phylogenetic diversity (SES MNTD)				
β plant taxonomic	0.07	NS		
β plant C/N	0.11	*	0.09	*
β plant N	0.12	*	0.10	*
β plant LDMC	0.14	*	0.13	*
β plant SLA	0.06	NS	0.03	NS
β plant phylogenetic (MPD)	0.10	*	0.09	*
β plant phylogenetic (MNTD)	0.16	*	0.14	*
Δ degree-days	0.13	*		
Δ frost days	0.12	*		
Δ moisture	0.02	NS		
Δ solar radiations	−0.05	NS		

CONCLUSION

We found that the ability of plant phylogenetic diversity to explain variation in the phylogenetic diversity of specialist herbivores in space is related to a strong association between plant and herbivore lineages (Fig. 1). Dinnage *et al.* (2012) proposed that correlation between the phylogenetic structures of multiple trophic levels is likely to arise whenever consumers have phylogenetic structure in their diets, which may be a characteristic shared by many food webs (Ødegaard *et al.* 2005; Weiblen *et al.* 2006; Futuyma & Agrawal 2009). Our study is in accordance with this view and suggests that phylogenies provide a significant contribution to the understanding of interactions across trophic levels. However, as illustrated with our absence of signal with alpha diversity, the importance of plant phylogenetic community composition in structuring herbivores assemblages may vary across the angiosperm phylogeny, highlighting the need for more subtle metrics of community phylogenetic structure. In contrast, the prevalence of relationships between herbivore alpha and beta diversity and leaf traits suggests that proximal measures of leaf resources and toughness represent good predictors of herbivore diversity and abundance. These findings have important conservation implications, by suggesting that adequate butterfly diversity maintenance will depend on plant diversity protection.

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AUTHORSHIP

LP and AG designed the research; LP, CN, AD and JNP collected the data; LP, AD and CN conducted the analyses; LP, NS, AG and SR wrote the manuscript.

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