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Basic and Applied Ecology 10 (2009) 726–736

Basic and
Applied Ecology
www.elsevier.de/baae

Variability in leaf traits, insect herbivory and herbivore performance within and among individuals of four broad-leaved tree species

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Received 30 December 2008; accepted 19 June 2009

Abstract

Individual plants may vary in their suitability as hosts for insect herbivores. The adaptive deme formation hypothesis predicts that this variability will lead to the fine-scale adaptation of herbivorous insects to host individuals. We studied individual and temporal variation in the quality of leaves of the tree species ash, lime, common oak, and sycamore in the field as food for herbivores. We determined herbivore attack and leaf consumption and performance of the generalist caterpillars of *Spodoptera littoralis* in the laboratory. We further assessed the concentrations of carbon, nitrogen and water in the leaves.

All measures of leaf tissue quality varied among and within individuals for all tree species. The level of herbivory differed among the tree individuals in lime, oak and sycamore, but not in ash. Within host individuals, differences in herbivory between the upper and lower crown layer varied in direction and magnitude depending on tree species. In feeding experiments, herbivore performance also varied among and within tree individuals. However, variation in palatability was not consistently related to the leaf traits measured or to herbivory levels in the field. The ranking of individuals with respect to the quality of leaf tissue for herbivorous insects varied between years in lime and oak. Thus, trees of both species might present moving targets for herbivores which prevents fine-scale adaptations. In contrast, among individuals of ash and sycamore the pattern of insect performance remained constant over 2 years. These species may be more suitable hosts for the formation of adapted demes in herbivores.

Zusammenfassung

Die Eignung als Wirtspflanze für herbivore Insekten kann zwischen einzelnen Individuen einer Pflanzenart variieren. Die "*adaptive deme formation hypothesis*" sagt eine kleinräumige Anpassung herbivorer Insekten an diese Variabilität der Wirtsindividuen vorher. Wir untersuchten die individuelle und zeitliche Variabilität der Blattqualität von Gemeiner Esche, Winter-Linde, Stiel-Eiche und Berg-Ahorn im Feld als Nahrung für Herbivore. Wir bestimmten die Fraßintensität im Feld und die Reaktion der generalistischen Raupen von *Spodoptera littoralis* im Labor. Weiterhin bestimmten wir den Gehalt an Kohlenstoff, Stickstoff und Wasser in den Blättern.

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doi:10.1016/j.baae.2009.06.006

Alle gemessenen Blattqualitätsmerkmale variierten zwischen und inerhalb der untersuchten Individuen für alle Baumarten. Das Ausmaß des Herbivorenfraßes variierte zwischen den Individuen von Linde, Eiche und Ahorn, jedoch nicht von Esche. Innerhalb der Baumindividuen gab es Unterschiede in der Herbivorie an Blättern aus der unteren und der oberen Schicht der Baumkrone, welche jedoch bezüglich ihrer Stärke und Richtung zwischen den Baumarten variierten. Die Reaktion der Herbivoren in den Fraßexperimenten variierte ebenfalls zwischen und innerhalb der Baumindividuen. Diese Variabilität war jedoch nicht durchgängig mit im Feld erhobenen Blatt- oder Herbivoriemerkmalen erklärbar. Die Rangfolge der Individuen einer Baumart bezüglich ihrer Blatt- oder Herbivoriemerkmale war nicht gleich in den zwei Untersuchungsjahren für Linde und Eiche. Baumindividuen dieser Arten repräsentieren daher eher „*moving targets*“für Herbivore, welche eine kleinräumige Anpassung verhindern. Dagegen war das Muster der Reaktion der Herbivoren auf Blätter der jeweiligen Individuen von Esche und Ahorn in beiden Jahren konstant. Diese beiden Arten können daher als geeignete Wirte für die Herausbildung von adaptierten *demes* herbivorer Arten angesehen werden.

Keywords: Adaptive deme formation; Broad-leaved trees; Canopy; Insect herbivory; Intraspecific variability; Palatability; Resource heterogeneity

Introduction

Leaf traits such as thickness, water content, C/N-ratio and the content of secondary compounds differ among individuals but also within individuals of a given tree species (among individuals: e.g. Laitinen, Julkunen-Tiitto, & Rousi 2000 ; Osier, Hwang, & Lindroth 2000b, within: e.g. Dudt & Shure 1994; Henriksson et al. 2003). These differences affect the feeding behaviour and development of insect herbivores (e.g. Ayres, Suomela, & MacLean 1987; Fortin & Mauffette 2002; Osier & Lindroth 2001). Ultimately, such differences in leaf quality among plant individuals may lead to the evolution of genetically distinct groups (demes) within herbivorous insect species that are adapted to individual plants (adaptive deme formation hypothesis, Edmunds & Alstad 1978). Because of their longevity, trees are the most likely host candidates for the formation of adaptive demes. However, most experimental tests have not supported the adaptive deme hypothesis (see Ruhnke, Schädler, Matthies, Klotz, & Brandl 2006) and the hypothesis is still controversial (e.g. Cobb & Whitham 1998; Van Zandt & Mopper 1998).

Important preconditions for the evolution of adaptive demes are that differences in leaf quality exist among host individuals (host heterogeneity) and that at least the relative differences in the suitability of individual host plants (the ranking of individuals) remains the same for several generations of insects. Resource heterogeneity has been studied in several tree species (e.g. Fortin & Mauffette 2002; Marquis 1988; Reynolds & Crossley 1997; Roslin et al. 2006; Rowe & Potter, 1996; Suomela & Nilson 1994). Most of these studies relied on chemical analyses, but some also studied palatability as an integrative and functionally relevant measure of resource heterogeneity. Few studies, however, have related levels of herbivore attack in the field to the palatability estimated in laboratory trials (Rowe & Potter 1996).

We investigated the intraspecific variability in insect herbivory and in the quality of leaf tissue for herbivorous insects in four broad-leaved tree species (*Fraxinus excelsior*, *Tilia cordata*, *Quercus robur*, and *Acer pseudoplatanus*) both among and within tree individuals in two consecutive years. For this, we assessed and compared measures of leaf quality, herbivore attack in the field and palatability in the laboratory. These data allowed us to answer the following questions: (1) do leaf traits differ among individual trees and between layers within a tree? (2) Are herbivory levels in the field as well as consumption and growth of a polyphagous insect herbivore in the laboratory related to leaf traits? Specifically, we would expect positive correlations to nitrogen and water content and negative correlations to carbon content. (3) Are differences in leaf traits and palatability consistent across years?

Methods

The study area and tree species

The study was performed in Central Germany in the “Leipzig flood-plain forest” (city of Leipzig, Saxony, Germany) which covers an area of ca. 1900 ha. In spring 2001, a construction crane was set up in this forest, which made it possible to work in the canopy of an area of 1.6 ha. In the crane plot, the vegetation is classified as a typical flood-plain forest (Querco-Ulmetum) on nutrient-rich loamy flood-plain soils (Morawetz & Horchler 2004). It is rich in species and dominated by ash, sycamore, oak, lime, maple and hornbeam. In this study, four of the dominant tree species were used: Ash

(*F. excelsior* L.), lime (*T. cordata* Mill.), common oak (*Q. robur* L.), and sycamore (*A. pseudoplatanus* L.).

Levels of herbivore attack in the field

In August 2002 and 2003, herbivore attack was estimated in the field. Three mature individuals of each tree species growing less than 150 m from each other were selected. Within the trees an upper layer consisting of sun leaves and a lower layer of shade leaves were distinguished. In each layer of each tree 25 leaves from each of four branches were collected (200 leaves per tree). Total leaf area was measured using Sigma Scan Pro 5 and the leaf area consumed by herbivores estimated. The specific weight of leaf material differed between the two sampled layers and therefore the consumed leaf area in the upper tree layer was corrected by tree-specific conversion factors to take the thickness of leaves into account. These factors were estimated using 20 leaves collected from each tree individual and layer in July 2002 and July 2003.

Feeding experiments

Palatability of leaves of the same tree individuals and layers as in the field study were investigated in laboratory experiments using larvae of the African cotton leafworm (*Spodoptera littoralis* (Boisduval); Lepidoptera: Noctuidae), a polyphagous herbivore. Local genetic structure in response to host plant quality is considered to be more likely in specialist herbivores (Boecklen & Mopper 1998; Mopper 1996). However, for the assessment of differences in leaf quality, feeding by a generalist herbivore should provide a more unbiased measure which is less likely affected by possible pre-adaptations to quantitative and qualitative leaf traits of a given plant species. Furthermore, only generalist herbivores can be used for a comparative assessment of heterogeneity of leaf quality across different tree species. To avoid potential effects of induced plant defence due to previous leave sampling, four branches not used in the assessment of herbivore attack were selected. Six leaves were sampled from each of these branches. The caterpillars of *S. littoralis* used in the experiment originated from a lab stock bred on a bean-based artificial diet to avoid adaptation of the insects to specific trees. Furthermore, to control for potential differences among larvae originating from different egg batches, individuals from each egg batch were distributed across experimental treatments. The bioassays were carried out in July 2002 and 2003. The foliage was fully mature and in both years of comparable age. After sampling, the leaves were immediately sprayed with deionised water to keep them turgescent, put into plastic bags, transported in a cooled box to the laboratory and stored in a refrigerator at 4 °C. The feeding experiments started a few hours after sampling. From each leaf one disc of 23 mm diameter was punched, weighed and placed individually in a Petri dish together with a piece of moistened filter paper. Early third instar larvae of *S. littoralis* were weighed and one larva was added to each Petri dish. The dishes were placed in a climate chamber at 26 °C with 12 h of light. After 24 h the larvae were killed by freezing. The larvae and the remaining leaf material were dried at 60 °C to weight constancy. Initial larval mass in the feeding experiments was converted into dry mass by using a calibration curve obtained from 60 further larvae. Initial fresh mass of leaf discs in the feeding trial was converted into dry mass using the mean water content of leaves for every branch (see below). Palatability was estimated as mg consumed leaf dry mass during 24 h.

Osier, Hwang, and Lindroth (2000a) showed that the growth of caterpillars reared in bags on trees was positively correlated with the growth of larvae reared in the laboratory on leaves of the same tree. Further, foliage kept turgescent under insect-rearing temperature or refrigerated was shown to have stable protein and phenolic compound contents for up to 48 h following field sampling (Kleiner 1991). Hence, our results should be representative for field conditions, but without the confounding effects of microclimatic differences.

Leaf traits

Water, nitrogen and carbon content are known to influence leaf palatability (Schädler, Jung, Auge, & Brandl 2003). Thus, these traits were assessed for the foliage of every branch that was used in the feeding trials at the same time as the bioassays were performed. Leaf water content was determined by measuring the fresh mass and dry mass of five reference leaf disks for each branch. For the measurement of leaf carbon and nitrogen, a subsample of leaves from every branch was vacuum-dried for 48 h, milled and kept at −20 °C until C and N concentrations were measured using an element analyzer (Vario EL, Elementar Analysensysteme GmbH, Hanau, Germany).

Data analysis

Data were analysed separately for each tree species. For the analyses of levels of herbivory in the field, the 25 individual values of consumed leaf area per branch were averaged. To normalize the distribution of residuals, these means were square-root transformed. The effects of year, tree individual and tree layer were tested by three-factor ANOVAs (Proc. GLM, SAS Version 9.1).

For the analyses of the feeding experiments, the six values obtained for each branch were averaged and the effects of year, tree individual and tree layer on leaf consumption and larval performance were analysed by ANCOVAs. For the analyses of consumed leaf material and final larval dry mass, initial larval dry mass was used as a covariate. Thus, possibly confounding effects of initial dry mass were removed from the analysis of the effects of the factors of interest and analyses of relative consumption rate and relative growth rate of larvae were obtained (Horton & Redak 1993; Raubenheimer & Simpson 1992). Values of initial and final larval dry mass were log-transformed in the analyses of the relative growth rate. For the analyses of the biomass gain of larvae, consumed leaf material was used as covariate, giving an estimate of the efficiency of conversion of ingested food into body substance (ECI; Waldbauer 1968).

Effects of year, tree individual and tree layer on leaf C/N-ratio and water content of the leaves were analysed using three-factorial ANOVAs with arcsin-square root transformed values of the water content. The relationships between the utilization of leaf material in the bioassays (adjusted means from the ANCOVA for each layer of each individual tree), herbivory levels in the field, leaf C/N-ratio, and leaf water content were analysed by Spearman rank correlations (software "R", version 2.0.0) of mean values of each trait per tree layer, tree individual and experiment.

Results

Leaf traits

The mean C/N-ratio of the leaves differed among the individual trees in all studied species (ash 14.1–15.4, lime 13.5–14.8, oak 16.2–17.7, sycamore 16.5–18.2, Table 1). Within trees, C/N-ratio varied in ash and lime between the two tree layers, but there was no consistent pattern among the individual trees (significant interactions tree individual × tree layer in the two species, Table 1). Across the 2 years, the C/N-ratio of the leaf tissue varied significantly in all studied tree species. In lime and sycamore, the pattern of the C/N-ratio among-tree individuals changed across the 2 years. Further, the ranking of the C/N-ratio within trees between the two layers changed in lime and in oak.

The mean water content of the leaves differed significantly among individual trees in ash and sycamore (ash 63.6–65.4%, sycamore: 63.2–65.9%, Table 1). In all studied species it was higher in shade leaves than in sun leaves (ash 66.7% vs. 61.8%, lime 68.0% vs. 59.3%, oak 57.6% vs. 53.4%, sycamore 68.1% vs. 60.9%). This difference, however, varied among the tree individuals (ash, lime and sycamore) and across years (ash and sycamore).

Levels of herbivore attack in the field

Less than 5% of total leaf area of each of the four tree species was consumed by herbivores. Levels of herbivory in the field differed significantly among the individual trees of lime and, but not of ash (Fig. 1A and Table 2). In sycamore, levels of herbivory differed between tree individuals depending on year (significant interaction year × individual). Within trees, the leaf area consumed differed between the two tree layers in all four tree species (Fig. 1B). There was, however, no consistent effect of layer on herbivory levels across tree species: In ash, the upper layer of the trees suffered more leaf damage than the lower layer, whereas in lime the lower tree layer suffered more leaf damage than the upper layer, but the strength of the effect differed between years (Table 2). In sycamore and oak, there were no consistent differences between the tree layers. Across the 2 years, the consumed leaf area varied significantly in all studied tree species (means for 2002/2003: ash 56/38 $\mathrm{mm}^2\,\mathrm{leaf}^{-1}$, lime 144/109 $\mathrm{mm}^2\,\mathrm{leaf}^{-1}$, sycamore 106/183 $\mathrm{mm}^2\,\mathrm{leaf}^{-1}$, oak 127/61 $\mathrm{mm}^2\,\mathrm{leaf}^{-1}$). In oak and sycamore, the ranking of the tree individuals with respect to herbivory varied across years (Fig. 1A), and the direction and magnitude of the difference between the two tree layers varied in lime, oak and sycamore between the 2 years (Fig. 1B and Table 2).

Feeding experiments

In the bioassays with *S. littoralis*, the relative consumption rate of the larvae differed among the tree individuals in ash, sycamore and oak (Fig. 2 and Table 3), with similar patterns in both years. Within trees, it varied between the two tree layers in lime, oak and marginally significantly ($p<0.1$) also in sycamore. The larvae consumed more leaf material from the upper layer in lime and oak (Fig. 2). In sycamore, however, the data showed no consistent pattern.

The relative growth rate of the larvae differed among the tree individuals in all four investigated tree species (Fig. 2 and Table 3). However, the pattern among individuals of oak changed between years. The relative growth rate differed also within tree individuals between the two layers in lime and oak, but showed no consistent pattern across years (Fig. 2 and Table 3). Nevertheless, in lime palatability and insect performance varied more clearly within trees than among-tree individuals (see mean squares in Table 3).

The mean efficiency of conversion of ingested food into body substance by the larvae differed among individuals of lime, oak and sycamore (adjusted means: lime 0.698–0.867, sycamore 0.546–0.947, oak −0.008 to −0.243, Table 3), but not in ash (overall mean 0.786). Across years, the pattern of efficiency of conversion

Table 1. The effect of year, tree individual, and tree layer on the carbon/nitrogen-ratio and water content of the leaves of four tree species.

Source	d.f.	*F*-values			
		Ash	Lime	Sycamore	Oak
C/N-ratio					
Year	1	92.17***	12.04**	235.26***	106.33***
Tree individual	2	7.12**	11.67***	17.00***	6.43**
Tree layer	1	66.22***	21.11***	2.86$^{(*)}$	0.88
Year × tree individual	2	0.61	9.40***	6.97**	1.88
Year × tree layer	1	3.70$^{(*)}$	23.82***	0.50	4.97*
Tree individual × tree layer	2	9.70***	10.60***	0.95	1.81
Year × tree individual × tree layer	2	1.07	0.41	8.38**	0.39
Residual	36	[MS = 1.002]	[MS = 0.716]	[MS = 0.908]	[MS = 1.506]
Water content					
Year	1	23.87***	36.90***	1.07	2.04
Tree individual	2	8.20**	2.00	19.83***	2.62$^{(*)}$
Tree layer	1	160.96***	589.87***	411.99***	85.12***
Year × tree individual	2	0.21	2.24	1.72	2.95$^{(*)}$
Year × tree layer	1	5.48*	2.15	19.85***	0.19
Tree individual × tree layer	2	12.99***	25.07***	3.93*	0.34
Year × tree individual × tree layer	2	1.73	1.19	1.00	0.40
Residual	36	[MS < 0.001]	[MS < 0.001]	[MS < 0.001]	[MS < 0.001]

Results of analyses of variance. $^{(*)}P<0.10$; $^{*}P<0.05$; $^{**}P<0.01$; $^{***}P<0.001$. MS = mean square.

varied among the host individuals only in oak. Within trees, the conversion efficiency of the larvae varied between the tree layers in all four tree species, but it did not show a consistent pattern among the tree individuals (ash and oak) and across the 2 years (lime, sycamore, and oak; Table 3).

Correlations between leaf traits, herbivore attack and laboratory performance of herbivores

The analyses did not reveal any consistent pattern of correlations between the traits. In oak, the level of herbivore attack in the field was negatively related to the C/N-ratio (Spearman's rank correlation: $r = -0.64$; $P = 0.028$). In lime, the herbivore attack was positively related to the water content of the leaves ($r = 0.73$; $P = 0.009$) and negatively to the various measures of performance of *S. littoralis* in the bioassays ($r = -0.67$ to -0.83; all $P<0.03$). In contrast, there was no significant correlation between levels of herbivory in the field and leaf traits or the results of the bioassays in ash and sycamore.

In lime, the relative consumption rate of the larvae was negatively related to the water content of the leaves but this relation was only marginally significant ($r = -0.53$; $P = 0.084$). Again there were no correlations with the C/N-ratio and also no relations between relative growth rate or conversion efficiency and leaf traits. In sycamore, the relative consumption rate ($r = -0.58$; $P = 0.052$), the relative growth rate ($r = -0.68$; $P = 0.019$) and the conversion efficiency ($r = -0.81$; $P = 0.003$) of larvae of *S. littoralis* were negatively correlated with the C/N-ratio, but not with the water content. In oak, the relative consumption rate of the larvae was negatively related ($r = -0.64$; $P = 0.028$) to the water content of leaves, while the relation was marginally significant positive for relative growth rate ($r = 0.51$; $P = 0.091$) and conversion efficiency ($r = 0.57$; $P = 0.054$). In addition, the relative growth rate of larvae was marginally negative related to the C/N-ratio of leaf tissue in oak ($r = -0.56$; $P = 0.067$).

Discussion

Leaf traits and their effects on herbivore attack and insect performance

Carbon, nitrogen and water content of leaf tissue are important determinants of leaf herbivory (e.g. Mattson 1980). The variability of these traits may therefore determine herbivore activity and distribution. In the present study, the C/N-ratio and water content differed among host individuals. Furthermore, water content differed consistently between sun and shade leaves.

The importance of leaf chemical traits for the intensity of herbivory (Haukioja, Niemelä, & Sirén 1985; Schädler et al. 2003) and insect performance (Fortin & Mauffette 2002; Osier et al. 2000a; Schädler, Roeder, Brandl, & Matthies 2007) is well documented for trees and herbaceous plants. In our study, however, the level of herbivory in the field and the insect performance in feeding experiments was not consistently related to the measured leaf traits (see also Cipollini, Paulk, & Cipollini 2002 Cipollini et al. 2002; Dudt & Shure 1994; Rowe & Potter 1996). Variables like water content and C/N-ratio may sometimes fail to predict consumption by herbivores because they are correlated with a large number of morphological and chemical leaf traits (e.g. quantity of secondary compounds, structural carbohydrates and nutrients), which may have non-additive effects (reviewed by Haukioja 2005). For quaking aspen; Osier and Lindroth (2006) showed that the direction of changes in the content of carbon-based defence compounds is influenced by the availability of resources (light and nutrients) in accordance with the carbon/nutrient-balance hypothesis, but this effect was under strong genetic control in the investigated genotypes. Therefore, environment and plant genotype interact in their effects on plant suitability for herbivores. For specialist herbivores, which are often adapted to secondary compounds, availability of nitrogen in plant tissue may be more important.

Herbivore attack in the field

Herbivory levels differed among individuals in three of the four investigated tree species (lime, sycamore and oak). Although there were considerable differences between tree layers in the leaf area consumed, the magnitude and the direction of these effects varied among-tree species, tree individuals and across years.

Published studies on oak (Roslin et al. 2006), maple (Marquis 1988; Reynolds & Crossley 1997) and lime (Rowe & Potter 1996) found similar levels of variation in herbivory as in the present study. In these studies levels of herbivory mostly differed between the upper and the lower layer of the canopy. However, the patterns in our study are not always consistent with the findings of other authors for closely related (Reynolds & Crossley 1997) or the same tree species (Rowe & Potter, 1996). Our results add to the evidence that tree populations in a given ecosystem may be heterogeneous resources for herbivorous insects due to variability both within and among individuals.

Insect performance in feeding experiments

The bioassays in the laboratory revealed differences in leaf palatability and herbivore performance among and within tree individuals in all four tree species. However, there was no consistent pattern among the investigated tree species. For example, between the two tree layers the relative growth rate of the larvae varied only in lime and oak. It was higher on leaves from the upper layer in lime, but lower in oak. Again, the results of our study are in contrast to findings from other experiments on the same or closely related tree species (Fortin & Mauffette 2002; Rowe & Potter 1996), suggesting that there is no general pattern for differences in leaf palatability from the different layers.

Only for lime, feeding levels in the laboratory were significantly related to levels of herbivore attack in the field, but in contrast to our expectations negatively. In ash, sycamore and oak there was no correlation between the level of herbivore attack in the field and insect performance in feeding experiments with *S. littoralis* (see also Rowe & Potter 1996 for lime). A number of factors may confound the expected positive correlation between insect performance in feeding trials and herbivory in the field: microclimate, predation, parasites (Dicke & Grostal 2001; Stamp & Bowers 1988) and migration of the herbivores (Batzer, Martin, Mattson, & Miller 1995; Magalhães, Janssen, Hanna, & Sabelis 2002). Therefore, results from feeding trials in the laboratory might reflect the actual palatability of leaf tissue more precisely than

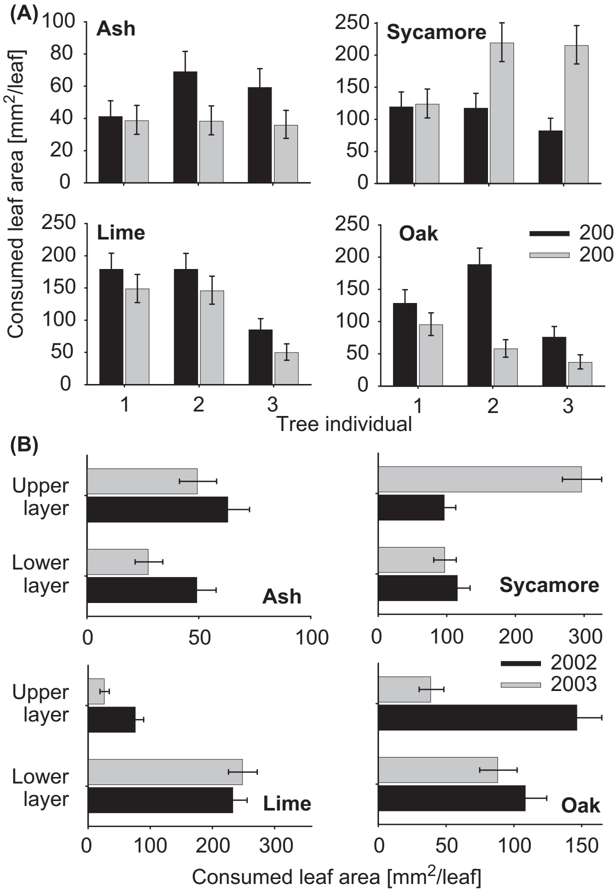

Fig. 1. The consumed leaf area by herbivores in the field. Effects of tree individual (A) and tree layer (B) in ash, lime, sycamore and oak (back-transformed least-squares means from ANOVAs ±1 back-transformed SE).

Table 2. The effect of year, tree individual, and tree layer on the amount of leaf material of four tree species consumed by herbivores in the field.

Source	d.f.	*F*-values			
		Ash	Lime	Sycamore	Oak
Year	1	5.19*	4.51*	15.37***	22.12***
Tree individual	2	0.84	17.65***	1.56	8.52***
Tree layer	1	5.31*	137.63***	15.08***	1.01
Year × tree individual	2	0.98	0.17	3.73*	3.53*
Year × tree layer	1	0.53	7.90**	25.10***	10.92**
Tree individual × tree layer	2	0.52	$2.90^{(*)}$	4.82*	2.33
Year × tree individual × tree layer	2	1.15	7.58**	1.89	0.40
Residual	36	[MS = 4.18]	[MS = 6.44]	[MS = 8.26]	[MS = 6.48]

Results of analyses of variance. $^{(*)}P<0.10$;$^{*}P<0.05$;$^{**}P<0.01$,$^{***}P<0.001$. MS = mean square.

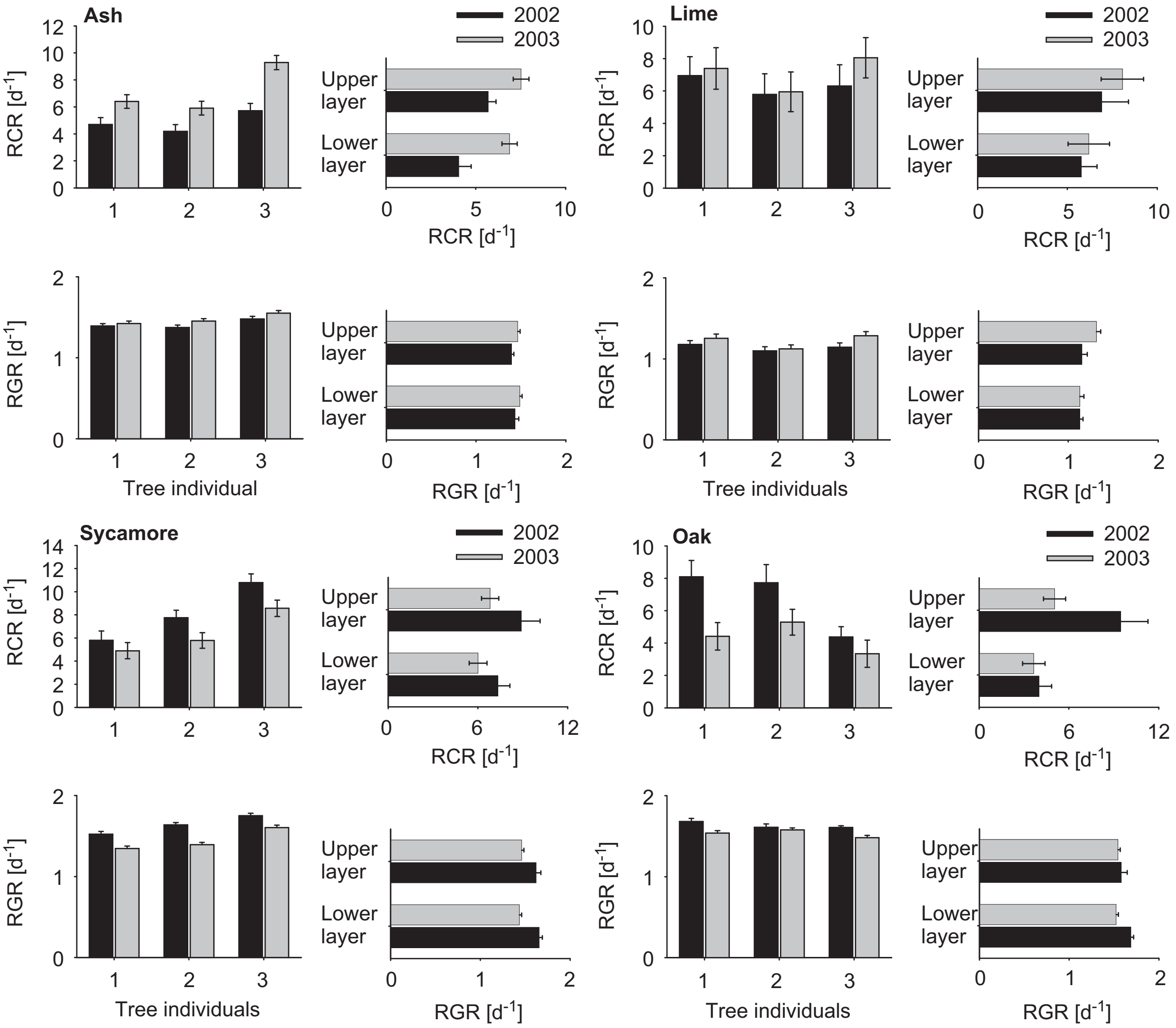

Fig. 2. Relative consumption rate (RCR) and relative growth rate (RGR) of larvae of *S. littoralis* feeding on ash, lime, sycamore or oak measured across 24 h (least-squares means from ANCOVAs ± 1 SE).

levels of herbivory in the field. Moreover, leaf damage in the field is usually caused by a multitude of herbivores, whereas in bioassays the palatability and usability of leaf tissue is tested in most cases with single herbivore species. Further, palatability trials are usually performed with generalist herbivores whereas feeding

Table 3. The effect of year, tree individual, and tree layer on consumed leaf material, final larval dry mass and biomass gain of the larvae of *S. littoralis*.

Source	d.f.	*F*-values			
		Ash	Lime	Sycamore	Oak
Consumed leaf material					
Initial larval dry mass	1	35.93***	38.23***	52.16***	68.16***
Year	1	17.01***	1.35	6.65*	0.02
Tree individual	2	15.37***	1.95	23.94***	3.61*
Tree layer	1	2.27	4.98*	3.02$^{(*)}$	5.42*
Year × tree individual	2	2.10	0.58	0.62	0.75
Year × tree layer	1	0.39	0.37	0.88	2.21
Tree individual × tree layer	2	1.33	1.02	0.76	0.66
Year × tree individual × tree layer	2	0.58	< 0.01	2.58$^{(*)}$	1.52
Residual	35	[MS = 1.836]	[MS = 4.843]	[MS = 3.272]	[MS = 3.100]
Final larval dry mass					
Initial larval dry mass	1	155.17***	271.25***	204.23***	1001.76***
Year	1	7.60**	1.06	76.03***	47.72***
Tree individual	2	9.24***	8.62***	41.98***	9.51***
Tree layer	1	1.38	22.91***	0.93	0.12
Year × tree individual	2	0.44	1.90	1.19	3.61*
Year × tree layer	1	0.19	8.53**	0.17	4.67*
Tree individual × tree layer	2	1.57	0.73	1.65	3.25$^{(*)}$
Year × tree individual × tree layer	2	0.04	0.69	3.48*	0.28
Residual	35	[MS = 0.006]	[MS = 0.007]	[MS = 0.006]	[MS = 0.003]
Biomass gain of larvae					
Consumed leaf material	1	101.66***	396.93***	424.69***	20.95***
Year	1	0.16	11.40**	52.07***	32.85***
Tree individual	2	1.97	6.77**	15.55***	6.33**
Tree layer	1	5.58*	11.85**	3.48$^{(*)}$	6.33*
Year × tree individual	2	0.77	0.42	1.35	4.62*
Year × tree layer	1	< 0.01	16.07***	6.59*	13.23***
Tree individual × tree layer	2	2.94$^{(*)}$	0.13	1.19	3.30*
Year × tree individual × tree layer	2	0.37	1.81	0.98	0.26
Residual	35	[MS = 0.066]	[MS = 0.016]	[MS = 0.058]	[MS = 0.070]

Results of analyses of covariance. $^{(*)}P<0.10$; *$P<0.05$; **$P<0.01$; ***$P<0.001$. MS = mean square.

damage in the field is often caused by specialists. However, even polyphagous insect herbivores differ in their response to variation in leaf tissue quality (e.g. Cipollini et al. 2002; Schädler, Roeder, Brandl, & Matthies 2005). This may also contribute to the missing relationship between herbivore attack in the field and palatability estimates derived from laboratory experiments.

Temporal predictability of resource quality

The leaves of the lower layer (shade leaves) had a higher water content than those in the upper layer in both study years. Senn, Hanhimäki, and Haukioja (1992) found that in mountain birch (*Betula pubescens* ssp. *tortuosa*) the water content of leaves of individuals was positively correlated between two successive years. This was also the case in ash and sycamore. In contrast, the C/N-ratio of the leaf tissue showed no consistent patterns across years. In contrast, Osier et al. (2000b) found for quaking aspen (*Populus tremuloides*) that concentrations of phytochemicals were highly correlated between two consecutive years, suggesting that phytochemical profiles are more specific for individuals than general nutrient levels (Haukioja et al. 1985; Laitinen et al. 2000; Riipi et al. 2004). This might not be true for all phytochemicals, proteins and sugars (Riipi et al. 2004). Hence, the ranking of palatability of plants for herbivores within a host population is not necessarily constant over time.

Levels of herbivore attack in the field and the performance of larvae of *S. littoralis* grown on leaves varied across the two investigated years. However, the effect of tree individuals on the performance of *S. littoralis* did not change across years for ash and sycamore and might therefore be predictable. Similarly, the relative ranking of individual trees of mountain birch in terms of leaf quality for herbivores remained constant between years, in spite of overall differences (Hanimäki, Senn, & Haukioja 1995; Senn et al. 1992).

Implications for local adaptation and formation of adaptive demes in insect herbivores

Resource heterogeneity among host individuals is an essential precondition for the formation of adaptive demes (Edmunds & Alstad 1978) but has usually not been tested in studies on local adaptation of phytophagous insects (Boecklen & Mopper 1998). Differences in the palatability and usability of leaves within tree individuals may reduce the likelihood of adapted deme formation by reducing the amount of leaf tissue of similar quality. This might not be crucial if the variability within host individuals is smaller than among them, so that tree individuals remain distinct from each other. However, variation within individual trees can be larger than among them (Roslin et al. 2006). Together with changes of leaf palatability within and across years, such variation precludes fine-scale adaptations to certain host individuals. Suomela and Nilson (1994) further found that the importance of within- and among-tree variability may depend on the developmental stage of the herbivore. Heterogeneity in resource quality can be also caused by variability in host plant phenology. This variability can be under genetic control (individual heterogeneity) or may even differ between years (temporal variability; see Mopper 2005). For instance, inter-tree variability of bud phenology (Masaka & Hara 2000; Tikkanen & Julkunen-Tiitto 2003) and intra-annual variation of leaf quality (Ruusila et al. 2005) affect susceptibility to herbivores. In the study reported here, the pattern of insect performance within trees varied across years. This might constrain adaptation further. Patterns of resource heterogeneity have to be further consistent over several generations of the herbivore (Mopper, Stiling, Landau, Simberloff, & van Zandt 2000). However, our study shows that in lime and oak the pattern of variation in the nutritional value of leaf tissue for herbivorous insects varied even between 2 successive years. We showed further that individual trees at least of these species might present moving targets for herbivores, which prevents fine-scale adaptations.

This study investigated one basic condition for the formation of local adaptations in phytophagous insects: the variability of host quality and its temporal predictability. However, local adaptation is driven by both plant and herbivore traits. The formation of adaptive demes in insect herbivores is expected to be more common in specialized insects (Boecklen & Mopper 1998; Mopper 1996). Van Zandt and Mopper found tentative support for a more likely adaptation in endophagous and parthenogenetic insects, but, contrary to predictions, not for more sessile herbivores. In their meta-analysis, Hoeksema and Forde (2006) concluded that significant herbivore local adaptation requires that herbivores have greater gene flow than their host plants to provide the necessary genetic variation on which selection may act. Future studies on local adaptations and the formation of adaptive demes may provide further insights by a simultaneous consideration of biological traits of both host and herbivore species.

Conclusions

Leaf tissue quality, levels of herbivory in the field, palatability and insect performance varied in ash, lime, oak and sycamore. However, the pattern of herbivory and insect performance was not always related to the measured leaf traits. Overall resource heterogeneity among-tree individuals appears to be common. However, the temporal predictability of this heterogeneity is often quite low and may differ among plant species. Therefore, the probability that herbivorous insect species adapt to individual hosts is generally low and may depend on the specific plant–insect system.

Acknowledgements

This study was done within the *Leipzig Canopy Crane Project*. We thank our colleagues of the Department of Community Ecology at the UFZ (Halle, Germany) for assistance, especially Daniel Prati, Hannelore Jany, Antje Thondorf and Ines Stolpe. We are grateful to Peter Proksch and Gero Eck (University of Düsseldorf, Germany) for providing *Spodoptera littoralis*. Thanks to Peter Horchler and Martin Unterseher, the coordinators for the *Leipzig Canopy Crane Project* (Leipzig, Germany). We thank two anonymous referees for helpful comments on a former version of the manuscript.

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