



Plant quality or quantity? Host exploitation strategies in three Chrysomelidae species associated with Asteraceae host plants

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Abstract

Phytophagous insects which feed on the leaves of herbaceous host plants have to adapt their life histories to the fact that protein nitrogen is usually highest in growing tissues in spring. We monitored field populations of larvae and adults of three chrysomelid species (*Galeruca tanacetii* (L.) (main host *Achillea millefolium* (L.) Yarrow), *Cassida rubiginosa* (Mueller) (main host *Cirsium arvense* (L.) Scop.) and *Oreina luctuosa* (Suffrian) (host *Centaurea scabiosa* (L.)) together with the amount of protein nitrogen of their food resources and host plant biomass. As expected, the development of host quality, measured as concentration of protein nitrogen, and host plant biomass showed inverse trends during the season. The euryphagous *G. tanacetii* attacks *Achillea* early and profits from high nitrogen concentrations in the leaves. Occasional overexploitations of local populations of *Achillea* are compensated by the capacity to move to other host species. In *C. rubiginosa*, a species with a host range restricted to the Cardueae, the main larval feeding activity is postponed to a period when the nitrogen content of the host leaves had dropped to 50% of its initial value, but when host plant biomass had increased by 30%. In the monophagous *O. luctuosa* the larval development is synchronized with a still later phase of host phenology, at which the nitrogen content is below 50% but plant biomass has reached its maximum. There seem to be selection factors, which oppose the use of high quality food in spring and which force the latter two species to postpone their larval development to a later time in the year. This could be caused by numerous factors like, for example, mean daytime temperature. Later in the season the larvae have to cope with the low quality of their host plants. They have, however, the advantage of large quantities of food available.

A laboratory study with adults and mature larvae of *O. luctuosa* shows that this species can overcome low levels of protein nitrogen either by selecting younger leaves with higher nitrogen concentrations or by increasing the daily food consumption rate (RCR) on leaves with a low level of nitrogen and by a prolongation of the feeding period. In this way the larvae compensate the effect of lower daily growth rates (RGR) and a lower food conversion index (ECI) on poor food quality: Regardless of the level of protein nitrogen there was no statistically significant difference in total gain of weight during the third-instar feeding period and in the weight at the end of the third larval stage. The three investigated chrysomelids show that there exists a broad spectrum of adaptations to overcome the dilemma of variable food quality.

Introduction

The nutritive quality of plant tissues plays an important role in insect-plant interactions (Feeny, 1976; Mattson, 1980; Strong et al., 1984; Price, 1991). The major nutrient required by phytophagous insects is protein, which in most cases is the limiting factor

for optimal growth of insects (Bernays & Chapman, 1994). The level of protein nitrogen is therefore an adequate measure for 'plant quality'. Plant quality is variable both in time and in space (Simpson & Simpson, 1990). This is particularly important for insects feeding on leaf tissues, where highest nutrient nitrogen concentrations occur during early growth in

spring. The level gradually declines during the course of the growing season until tissue senescence (Mattson, 1980; Bernays & Chapman, 1994). A widely used feeding strategy by which herbivores overcome the problem of shortage of nutrient nitrogen (McNeill & Southwood, 1978) is a synchronization of the life cycle with time periods when nutrients in the host plant are readily available (Slansky & Rodriguez, 1987). It is most pronounced in the 'flush feeders' (White, 1993) which exploit their host plant early in the season. The obvious benefits of flush feeding involve, however, other risks, like a higher unpredictability of the food, the overexploitation of the resource particularly when flush feeders with a restricted host range use the early growth of herbaceous host plants, low spring-temperatures or unfavorable microclimatic conditions within the small plants early in the season.

Our study deals with three Chrysomelidae species associated with herbaceous host plants. We examine the feeding strategies of *Galeruca tanacetii* on *Achillea millefolium*, *Cassida rubiginosa* on *Cirsium arvense*, and *Oreina luctuosa* on *Centaurea scabiosa*. These host plants belong to the Asteraceae, a plant family where nitrogen is translocated within the plant from the roots to the rosette leaves, from there to the stem leaves, and later into the flower heads and seeds (Heilmeier, 1988). The amount of leaf biomass, which can be used as food, varies over the course of the season. Plant quality and plant quantity are inversely correlated: In spring, biomass of young growing leaves is limited, but leaves have high levels of nutrient nitrogen. In late summer when plants are fully grown and leaf-biomass has reached its maximum, nitrogen concentrations of matured leaves have declined to low concentrations. We compare this seasonal change in host quality and quantity with the seasonal phenology of the three chrysomelid species and investigate the way each species exploits its host plant under the given situation. We test the hypothesis that the life stages of the beetles which are most dependent on nutrient nitrogen should occur at a time when the nitrogen level in leaves of their host plants is high.

The second part of this study examines the influence of food quality on food selection and on larval development of *O. luctuosa*, the most specialized species whose larvae have a late phenology and are therefore confronted with low food quality. Under such conditions phytophagous insects may have evolved different types of compensatory responses (Slansky & Rodriguez, 1987; Simpson & Simpson,

1990). Therefore choice experiments with different leaf-age classes and with plants grown under different levels of nitrogen fertilization were designed to answer the following questions: (1) How does the level of nutrient nitrogen influence behaviour and development of a late-season species? (2) By which compensatory mechanisms does *O. luctuosa* overcome the handicap of low nitrogen concentrations in its host? We tested the hypotheses that larval development is delayed on nitrogen deficient plants and/or that the beetles select younger leaves which have higher nitrogen concentrations.

Materials and methods

The chrysomelid species studied. The three species are folivorous on the leaves of their host plants, which are consumed by the adults as well as by the ectophytic larvae. They differ in their degree of host specificity and their life histories. For each chrysomelid species we selected in our study that host plant species which was most heavily attacked, in our observation area in Upper Frankonia. Throughout the season these plants were monitored together with the development of field populations of the three chrysomelid species.

Among the three species investigated *Galeruca tanacetii* (subfamily Galerucinae) has the broadest host range. This species feeds on many Asteraceae species but occasionally also on members of other families (Prevett, 1953). In our study area near Bayreuth, *Achillea millefolium* was the preferred host of *G. tanacetii*. This plant species is widely spread in Europe and is common in Upper Frankonia. Eggs hibernate and the larvae hatch early in spring. They develop from April to June. After a short pupal stage the adults appear from early June onwards. They enter a reproductive diapause during mid-summer. Mating and oviposition takes place in fall (Siew, 1966). Eggs and larvae of *G. tanacetii* contain anthrachinones which are assumed to provide protection against predators (Hilker & Schulz, 1991).

Cassida rubiginosa belongs to the subfamily Cassidinae and is restricted to the Asteraceae tribe Cardueae. For Europe, Zwölfer & Eichhorn (1966) recorded it from 19 species of the subfamily Carduinae and from a *Centaurea* species. In our study we have chosen *Cirsium arvense*, the Creeping Thistle, for the monitoring program. *C. rubiginosa* hibernates in the adult stage. Oviposition starts at mid-May, larvae can be found till late July and adults of the new generation

appear from mid-July onwards. A small percentage of the adults survives until the next flight season. The biology and ecology of this species is discussed in more detail by Steinhausen (1949) and Kosior (1975). Chemical defense is not known in *Cassida*. The adults have a cryptic colouration and shape and the larvae use fecal shields for their protection (Olmstead & Denno, 1993).

Oreina luctuosa is a highly host specific member of the subfamily Chrysomelinae. It is monophagous on a few species of the genus *Centaurea* (Knapweeds). Reitter (1912) recorded it from *Centaurea jacea* (L.). In the Frankonian Jura the species restricts its host range almost completely to *Centaurea scabiosa*. *Centaurea jacea* is only attacked in exceptional cases. In Upper Frankonia *O. luctuosa* occurs only locally, but it tends to form large populations which at certain sites persist for many years. The adults appear on their host plant towards late May. Oviposition starts in June and larvae can be found on the hosts until September. Full-grown larvae leave the host and enter the soil, where they hibernate either as larva or as pupa. Both larvae and adults of *O. luctuosa* contain autogeneously produced cardenolides which protect the species against predators (Dobler & Rowell-Rahier, 1994).

Field surveys. The field data were collected from 16 April to 18 September 1992 in Upper Frankonia in northern Bavaria, Germany. On seven sites plant parameters as well as the abundance of the beetle species were recorded at intervals of 14 days. The growth of each of the three host plant species was surveyed by measuring the height of 10 stems which were randomly chosen. As leaf biomass is a function of plant height, average plant height was used as an indicator for the development of food quantity. On 30 shoots/census we assessed the formation of flower heads. At one site plant material was harvested for the analysis of protein nitrogen. Leaf samples of ten plants per date and species (age classes mixed in the proportion of their respective occurrence) were harvested and oven dried. For *Cirsium* and *Centaurea*, the analyses could be done separately for each plant individual, but in *Achillea* the small amount of leaves per individual plant made it necessary to analyze pooled samples of the harvested plants.

During the field surveys the adult leaf beetles and, where possible, their larvae and eggs were counted in two or three of the study sites and data were pooled to obtain a larger basis for the reconstruction of the phenology of the feeding stages. The number

of *G. tanaceti* larvae feeding on different plant species was counted in 10 plots each measuring 0.25 m². The numbers of *C. rubiginosa* and *O. luctuosa* individuals were counted on samples of 25 and 50 individual host plants per site examined.

During the field survey, the age classes of all leaves occupied by adult *O. luctuosa* were registered. In addition, the relative quantity of leaf material available in the different classes was estimated from 5 plants/census. This was done by dividing the leaves into the categories 'young', 'mature', or 'senescent'. As criteria we used the position of the leaves on the individual plant and their general appearance.

Larval consumption and development on fertilized plants. At the end of May, i.e., one and a half month after sowing, *Centaurea scabiosa* plants were apportioned to four treatment groups which were then grown at different nutrient levels. The N0-group was kept on a nutrient-free peat-sand-substrate. For the N1- and the N3-group we used the same substrate but added two different levels of the long-term fertilizer 'Osmocoter (16-8-12-2)' (N1: 1 kg m⁻³; N3: 3 kg m⁻³). The fourth group (Nmax) was grown on a nutrient rich foliage-substratum and was provided with 3 kg Osmocoter m⁻³. By adding quicklime the pH was maintained at 5.6 in all treatment groups. After the last feeding experiment in August subsets of the four treatment groups were harvested for an analysis of biomass and protein nitrogen content.

For use in the laboratory experiments with larvae of *O. luctuosa* 11 pairs of field-collected adults were reared in a climatic chamber at a constant temperature of 20 °C and 16 h of light/day. Eggs were isolated and inspected daily for hatching larvae. Per treatment group 36 L1-larvae (not older than 24 h) were randomly selected. From the beginning they were provided with *C. scabiosa* leaves ad libitum and raised to the third stage. An experiment started when on the same day at least 10 larvae of a group had molted to the third stage. The surplus L3 were frozen and dried for determination of the larval dry weight at the start of the test. The L3-larvae were isolated in plastic containers and their weight was recorded daily at the same time with a micro-balance. After having reached the fourth stage, the L4 were again weighed alive and finally their dry weight was assessed.

The leaves used for the feeding experiments were always harvested at the same time of the day because of possible diurnal rhythms in the level of nitrogen. After their fresh weight had been assessed, they were

transferred into plastic boxes, where their stalks were kept in water. Of the remaining leaf material the fresh and dry weight was recorded individually per test box. In addition we used 5 boxes without larvae, per fertilizer group, to obtain a control for the amount of leaf water lost during the experiment. The amount of leaf material consumed was calculated as the difference between the fresh weight of the leaves before and after the experiment, corrected for water loss. These values were then transformed to dry weight values.

Body weight, developmental time, and the leaf mass consumed by the L3-larvae were directly recorded. From the dry weight data, growth and consumption parameters (relative growth rate (RGR), relative consumption rate (RCR), efficiency of consumption and ingestion (ECI)) were calculated after Waldbauer (1968). To assess mean larval weight, the weights of the larvae at the beginning and the end of the experiment were averaged.

Choice tests between leaf age classes. The choice tests were conducted with L4-larvae and adult females of *O. luctuosa*, because the latter consume more food than the males and select the oviposition sites. In Petri dishes one adult or three larvae were given the choice between four leaf disks (9 mm diameter) with 2 disks of 2 different age classes. Each experiment comprised ten dishes. Only dishes where feeding occurred were evaluated. The leaf material consumed was assessed as the amount of leaf area (mm²) consumed.

Nitrogen analysis. Food quality was measured as protein nitrogen content of leaves from which the mid rib had been previously removed. The leaves were dried at 70 °C for 48 h, ground in a mill and analyzed with the Mikrokjeldahl-method (Strauch, 1965). This process assesses only the reduced nitrogen, i.e., the form of nitrogen (proteins, amino acids) which can be used by insects (Dadd, 1985). The leaf water content of the four fertilizer treatments was determined as the difference between fresh and dry weight.

Statistical analysis. The seasonally changing relationship between plant height and the level of leaf nitrogen was examined with Spearman's rank correlation. The relations between leaf nitrogen content, larval consumption, and larval development in the feeding experiments were analyzed with oneway ANOVA or the Kruskal–Wallis test, if variances were not homogenous. Differences between means were compared with the Scheffé multiple range test. The

choice tests were analyzed with the Wilcoxon matched pairs signed-rank test. All the tests were performed with the SPSS-statistical software package.

Results

Seasonal development of the leaf beetle populations and of host plant quality and quantity

Galeruca tanacetii. Figure 1a shows the seasonal development of *Achillea* and its herbivore *G. tanacetii*. In the upper graph host plant biomass, represented by plant height, and host plant quality, represented by the content of protein nitrogen in leaves, follow slightly opposed trends with a negative correlation ($r_s = -0.80$, $P < 0.05$). If compared with *Cirsium* and *Centaurea* (Figures 1b and 1c), the initial nitrogen concentration of *Achillea* leaves was relatively low and the seasonal decline less marked. Elevated nitrogen-concentrations (28.45 mg N g⁻¹ DW) originating from winter storage were found only in early May. From late May to mid-September the nitrogen level fluctuated between 22 and 18.8 mg N g⁻¹ DW (Figure 1a). At the two study sites the *Achillea*-plants remained small and did not develop flower heads, which was probably due to poor soil conditions and the exceptionally dry weather conditions.

The lower graph of Figure 1a shows the average density of *G. tanacetii* larvae and adults pooled from two sites. The larvae appeared from mid-April onwards and reached their maximum density in mid-May, i.e., they were active at a period when the host plant was still small but when they could profit from the higher levels of leaf nitrogen available. In plots with high larval densities complete local defoliations of the first young plants of *Achillea* and other food plant species by larvae of *G. tanacetii* could be observed. The adult beetles reached their first maximum of abundance at the end of June. A second maximum was found after the end of their reproductive diapause in August.

Cassida rubiginosa. At the time when its rosettes started shooting the young leaves of *Cirsium arvense* had very high concentrations of nitrogen (mid-April: 59 mg N g⁻¹ DW) (Figure 1b). Accompanied by a marked increase in shoot height there was a pronounced decrease to 24.58 mg N g⁻¹ DW in mid-July. Minimum average concentrations of leaf nitrogen (22.43 mg N g⁻¹ DW) in August/September coincided

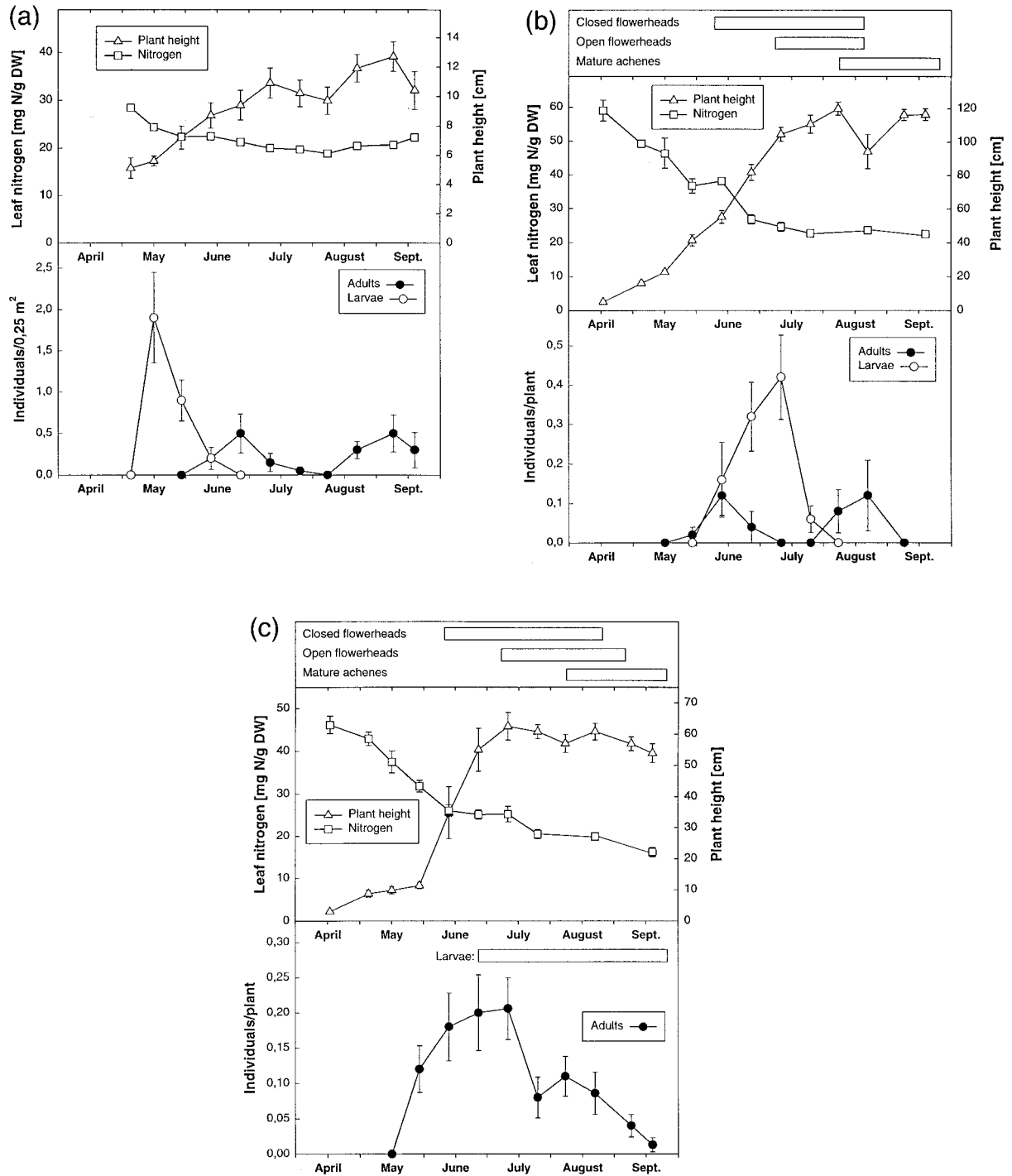


Figure 1. Seasonal phenology of three leaf beetle species and their host plants. (a) The life cycles of *Galeruca tanacetii* and *Achillea millefolium*, (b) The life cycles of *Cassida rubiginosa* and *Cirsium arvense*, (c) The life cycles of *Oreina luctuosa* and *Centaurea scabiosa*. Host plant parameters are represented by leaf-nitrogen concentration (squares) and the plant-height (triangles) in the upper graph. The life cycle of the leaf beetle species is documented by the number of larvae (open circles) and adults (closed circles) in the lower graph. Time of occurrence of larvae of *O. luctuosa* is shown as a bar. Means and standard errors.

with the phases of maximal shoot height, flowering, and maturation of the achenes. As in *Achillea*, the seasonal trends of plant height and N-concentration of leaves were negatively correlated ($r_S = -0.97$, $P < 0.01$).

Immediately after resuming their activity after hibernation in the second half of May the adults of *C. rubiginosa* (Figure 1b, lower graph) started oviposition. Adult numbers on the host plants reached a peak in mid-June, when the level of leaf nitrogen had declined to two thirds of its original value ($38.10 \text{ mg N g}^{-1} \text{ DW}$). At that time *Cirsium* was still growing and had attained only half of its final height. The maximum abundance of the larvae occurred in mid-July, i.e., one month later. By that time the level of leaf nitrogen had already dropped to $24.58 \text{ mg N g}^{-1} \text{ DW}$, whereas the leaf mass had almost reached its maximum. The development of the larvae continued until the end of August, when the adults of the new generation of *C. rubiginosa* attained their peak. These adults left the host plants at the beginning of September.

Oreina luctuosa. *Centaurea scabiosa* also showed an inverse correlation between plant height and leaf N-concentration (Figure 1c) ($r_S = -0.90$, $P < 0.01$). Nitrogen levels declined steadily from $46.00 \text{ mg N g}^{-1} \text{ DW}$ in mid-April to $26.05 \text{ mg N g}^{-1} \text{ DW}$ in mid-June. At that time the plants had attained about half of their final shoot length. From mid July ($25.24 \text{ mg N g}^{-1} \text{ DW}$) to the end of the growth period in August/September the reduction of the nitrogen level was less marked. This phase coincided with the flowering period, the maturation of the achenes and a loss in leaf mass. With $16.23 \text{ mg N g}^{-1} \text{ DW}$ the level of leaf nitrogen was at a minimum in mid-September.

O. luctuosa adults started feeding on *Centaurea* in late May (Figure 1c). In June their increase in numbers followed closely the marked growth flush of *Centaurea*. Adults of *O. luctuosa* had their maximum of abundance in mid-July and could be observed on all sites till mid-September. This peak of activity on the host coincided with the maximum of leaf mass, but occurred after the strong decline in the nitrogen concentration of the leaves.

Because of their cryptic behaviour it was not possible to count exactly the numbers of *O. luctuosa* larvae per plant. Their phenology is therefore only inferred from non-quantitative field observations and from data obtained by laboratory rearings. Under laboratory conditions the first larvae hatched on 10th of June and in the field the first larva was found on 26 June. The last

field record of a larva was made at the end of September. The lower graph of Figure 1c shows that the feeding period of *O. luctuosa*-larvae coincided with the period of high leaf mass and low nitrogen levels (between $26.05 \text{ mg N g}^{-1} \text{ DW}$ in mid-June and $16.23 \text{ mg N g}^{-1} \text{ DW}$ in mid-September).

Larval development and food consumption of third-instar O. luctuosa on leaves of different quality

Chemical analyses of fertilized plants. Leaf area and leaf biomass (E. Obermaier, unpubl. data) of all four treatment groups of *Centaurea scabiosa* differed and there was a general trend that in mature leaves the content of protein nitrogen increased with increasing fertilizer doses. The leaf water content increased with soil quality. The only consistent and statistically significant difference occurred between the non-fertilized plants (N0) and all fertilized groups (Table 1). An increase in the addition of fertilizer (N1, N3, Nmax) did not significantly change the level of leaf nitrogen. The non-fertilized plants (N0) and those with intermediate doses of fertilizer (N1 and N3) showed no significant differences in leaf water content.

Weight and development of the larvae. We found no significant effect of nitrogen treatment on weight gain or final weight of third-instar larvae of *O. luctuosa* between the treatment groups (Table 2). The N0-group, fed with nitrogen deficient plants, needed significantly longer for development (7.4 days) than the other treatment groups. The Nmax group, provided with leaves of both high nitrogen and high water content, showed the fastest development (5.2 days). The N1- and N3-group required an intermediate time for development (6 days) and did not differ significantly from each other.

Food consumption and utilization. The leaf quantity consumed by the nitrogen deficient N0-larvae was nearly twice as high as in the other groups ($P < 0.001$). The standardized consumption and growth indices (per mg body weight and per day) show always significant differences between the N0-treatment and the Nmax-treatment (Table 2) The ECI, a measurement for the efficiency of conversion and ingestion of food, had the lowest value in the larvae of the N0-group and attained a maximum in the larvae fed on Nmax-plants. The same situation is found in the relative growth rate (RGR), the gain in body weight per unit average body weight and day. On the other hand,

Table 1. Organic nitrogen and water content of mature leaves of *C. scabiosa* of different treatment groups. Different letters indicate a significant difference at $P < 0.05$

Group	N0		N1		N3		Nmax		P
	x	SE	x	SE	x	SE	x	SE	
Nitrogen [mg N g ⁻¹ DW]	16.63 ± 1.67		28.18 ± 1.29		27.57 ± 1.07		28.79 ± 1.06		0.001
	a		b		b		b		
Water [g H ₂ O/g DW]	2.43 ± 0.09		3.39 ± 0.21		3.36 ± 0.16		5.39 ± 0.39		0.001
	a		a		a		a		

Oneway ANOVA; Scheffé; $n = 10$.

Table 2. Development and conversion of food of L3-larvae of *Oreina luctuosa* on different fertilizer groups of *C. scabiosa*. Different letters indicate a significant difference at $P < 0.05$

Group	N0		N1		N3		Nmax		P
	x	SE	x	SE	x	SE	x	SE	
Increase in weight (mg)	5.28 ± 0.60		5.96 ± 0.48		5.52 ± 0.51		5.62 ± 0.39		n.s.
Final-weight (mg)	8.74 ± 0.60		8.54 ± 0.48		8.15 ± 0.51		8.07 ± 0.38		n.s.
Devel. time (days)	7.40 ± 0.31		6.00 ± 0.21		6.10 ± 0.15		5.20 ± 0.15		0.05
	a		b		b		c		
Leaf mass cons. (mg)	34.04 ± 2.41		17.69 ± 1.58		20.33 ± 1.59		14.04 ± 1.73		0.001
	a		b		b		b		
ECI* [%]	16.06 ± 2.10		36.01 ± 4.44		27.53 ± 2.04		47.53 ± 6.31		0.05
	a		b		ac		b		
RCR* (mg/mg/day)	0.76 ± 0.04		0.54 ± 0.05		0.65 ± 0.05		0.52 ± 0.08		0.05
	a		b		ab		b		
RGR (mg/mg/day)	0.12 ± 0.01		0.18 ± 0.01		0.18 ± 0.01		0.20 ± 0.01		0.05
	a		b		b		b		

Oneway ANOVA; Scheffé; $n = 10$ bzw. 15.

*Kruskal–Wallis.

the relative consumption rate (RCR), i.e., the relative food uptake per mg body weight and per day, gave significantly higher values in the N0-group than in the other three groups.

To demonstrate the trends in the rates of consumption and development of *O. luctuosa* L3 the mean values of some of the parameters given in Tables 1 and 2 have been plotted in Figure 2. (Figure 2A–D independent variable = feeding period of the L3, Figure 2E independent variable = total leaf mass consumed, Figure 2F–H independent variable = nitrogen content). The total leaf mass consumed and the RCR show positive and the ECI and RGR negative relationships with an increasing duration of the feeding period of the L3. The RGR and the total leaf mass consumed are inversely correlated. With an increasing nitrogen content the ECI values rise whereas the duration of the L3 feeding period or the amount of leaf mass consumed drop.

The effect of leaf-age in choice tests and under field conditions

Young and mature leaves had a significantly higher nitrogen content than senescent leaves (Table 3) (Kruskal–Wallis, $P = 0.001$). Statistically, young leaves could not be separated from mature leaves. When adult females and L4-larvae of *O. luctuosa* were allowed to choose between young and mature or between mature and senescent leaves (Figure 3), the adult females always showed a strong discriminating behavior and preferred young over mature and over senescent as well as mature over senescent leaves. The L4-larvae also preferred the younger age classes, but discrimination was only significant between mature and senescent leaves. These responses are consistent with field observations made in July on the position of adult *O. luctuosa* on *Centaurea* plants. The distribution of adults over young, mature and senescent leaves of *C. scabiosa* deviated significantly from the propor-

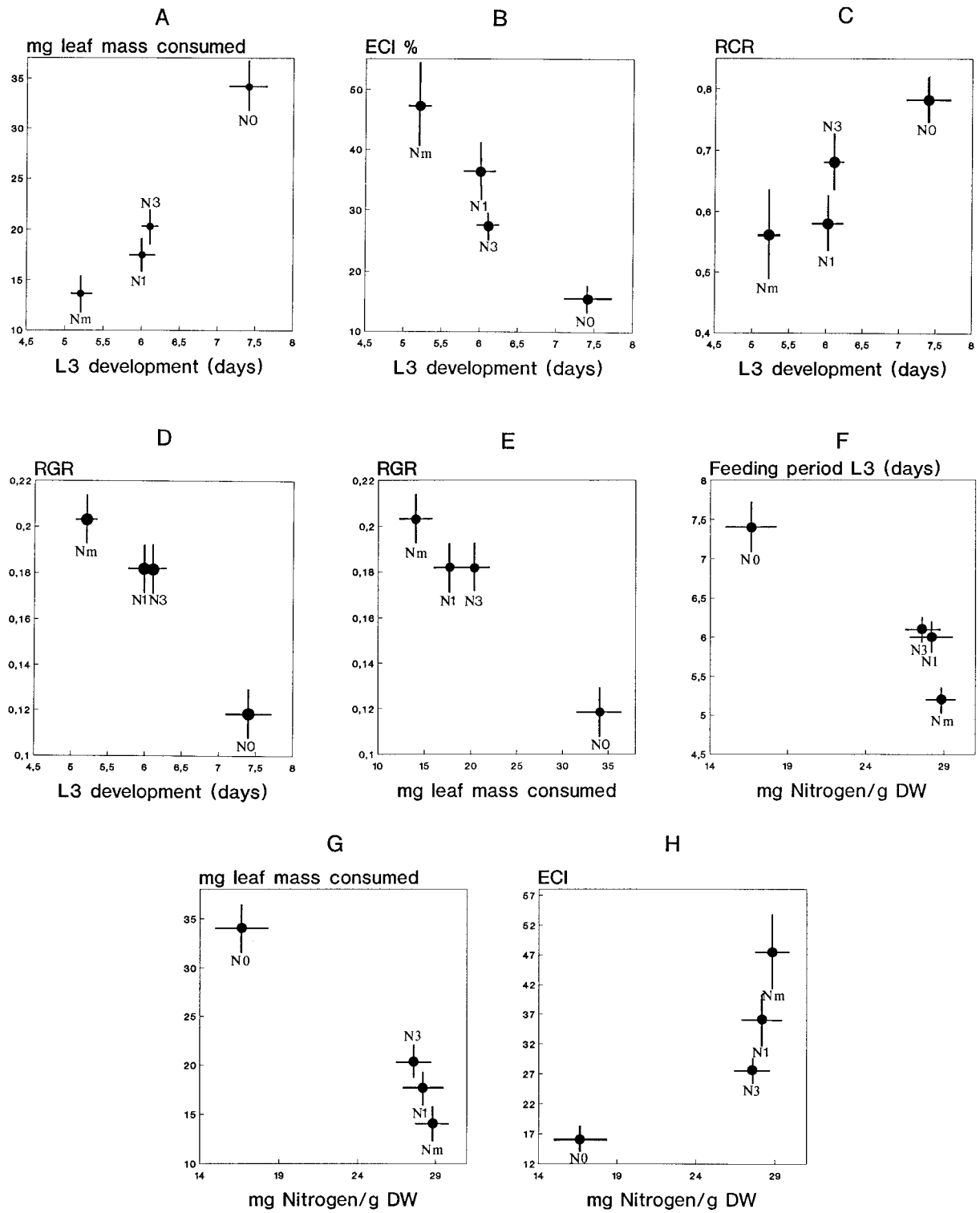


Figure 2. Plots of growth parameters of third-instar larvae of *Oreina luctuosa* on *Centaurea scabiosa* plants grown under different fertilizer regimes. Points represent mean values, bars standard errors. (A) Total leaf mass consumed and the duration of the feeding period. (B) ECI and the duration of the feeding period. (C) RCR and the duration of the feeding period. (D) RGR and the duration of the feeding period. (E) RGR and the total leaf mass consumed. (Graphs A–E, treatment groups, from left to right: Nmax, N1, N3, N0). (F) Duration of the feeding period and nitrogen content of the leaves. (G) Total leaf mass consumed and nitrogen content of the leaves. (H) ECI and nitrogen content of the leaves. (Graphs F–H, treatment groups, from left to right: N0, N3, N1, Nmax).

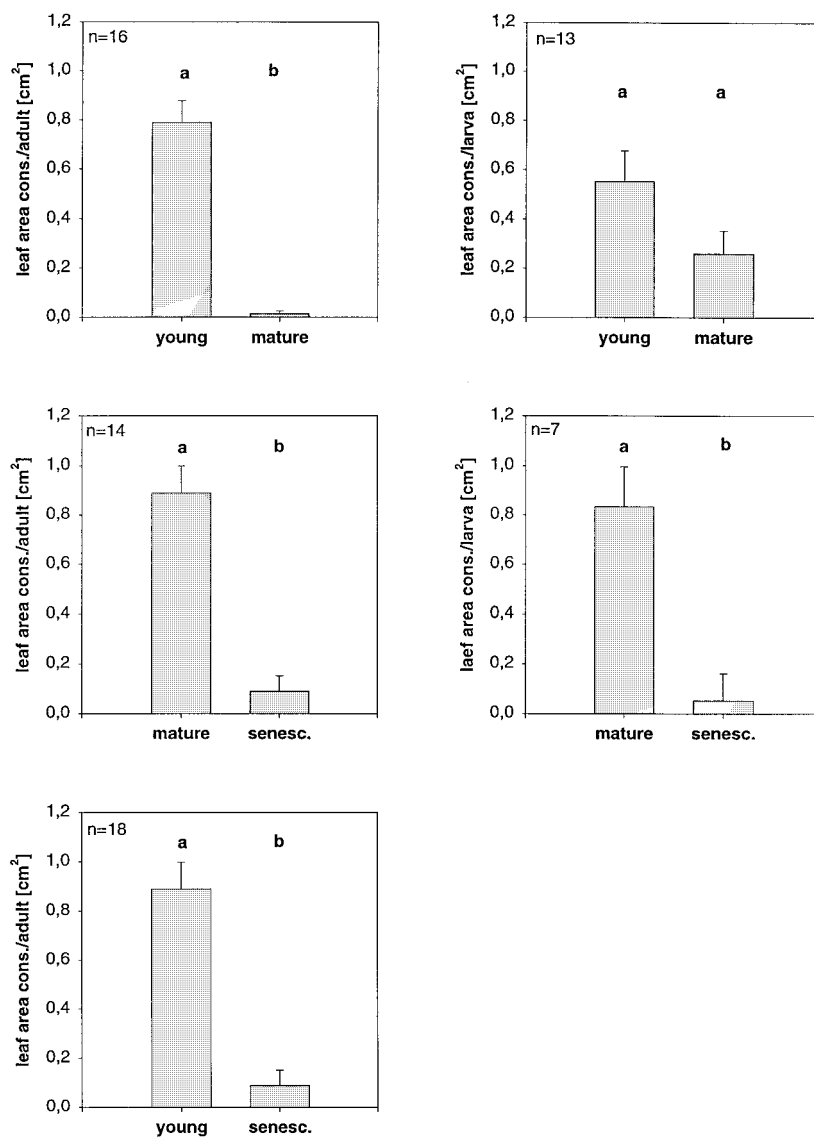


Figure 3. Choice tests between three leaf age classes (young, mature, senescent) of *Centaurea scabiosa*. The tests were conducted with adults (left side) and larvae (right side) of *Oreina luctuosa*. Means and standard errors; values with same letters do not differ significantly at $P < 0.05$ (Wilcoxon).

Table 3. Organic nitrogen content of different leaf age classes of *C. scabiosa*. Different letters indicate a significant difference at $P < 0.05$

Leaf age	Young		Mature		Senescent		P
	<i>x</i>	SE	<i>x</i>	SE	<i>x</i>	SE	
Nitrogen (mg N g^{-1} DW)	27.40 ± 1.47		24.79 ± 1.11		18.47 ± 0.76		0.001
	a		a		b		

Kruskal–Wallis. $n = 35$, all fertilizer treatments pooled.

tions of the different age-classes available on 26 June ($\chi^2 = 19.8$, $P < 0.0001$) and 24 July ($\chi^2 = 86.5$, $P < 0.0001$). There was always an over-proportional preference for young leaves and a strong tendency to avoid senescent leaves.

Discussion

The main feeding periods of folivorous chrysomelid species vary broadly, as can be shown from data of 59 European and North American species compiled from Heikertinger et al. (1954). The data suggest that the majority of chrysomelid species associated with woody plants attack their hosts early, in April and May, whereas the main activity periods of species exploiting herbaceous host plants have a maximum in June and range from April–May to August–September (Figure 4). The multitude of life-cycles in the leaf-feeding chrysomelids suggests different types of adaptations to the seasonal development of their host plants. This hypothesis is tested by a comparison of the life histories of three species on herbaceous plants and their adaptations to the seasonal changes in host quality and quantity.

As expected all three host plants showed inverse relationships between the development of the level of leaf nitrogen, which declined from April–May to September, and the plant biomass above ground, which increased till July or August–September, respectively. *Achillea millefolium* (Figure 1a), a hemicryptophyte growing on soils moderately rich in nutrients, starts its development with a relatively low level of leaf nitrogen; during the season its concentration declines by about 25%. Plant growth was relatively low, as from May to August–September plant height increased by a factor of only 2.0. *Cirsium arvense* (Figure 1b) is a geophyte and an indicator of soils rich in nitrogen. Its young rosettes begin their growth with a very high level of nitrogen which by flowering and seed production in August–September is reduced by 60%. The steep decline in the concentration of leaf nitrogen is accompanied by a strong increase in plant biomass, which shows the augmentation of plant height from early May to August–September by a factor of 7.3. *Centaurea scabiosa* (Figure 1c) is a hemicryptophyte occurring usually on calcareous soils with a low content of nutrients. At the beginning of the vegetation period its leaves have an intermediate level of nitrogen which by August–September has declined by about 64%. A steep increase in plant biomass takes place

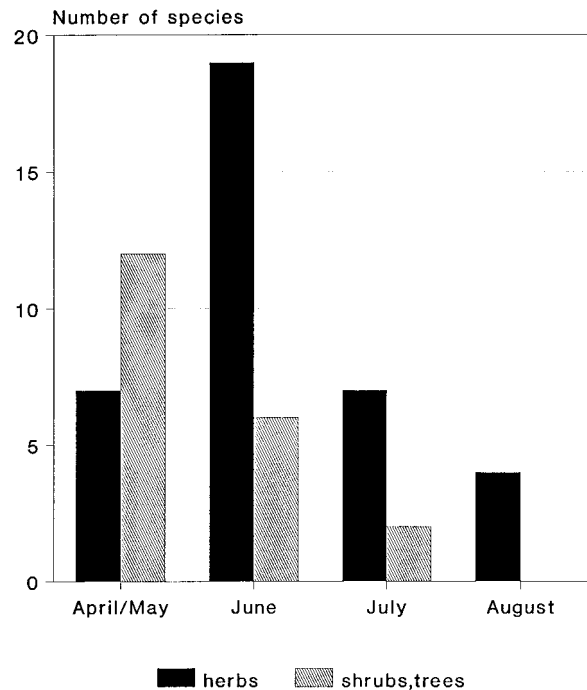


Figure 4. The main larval feeding periods of folivorous chrysomelid species on herbs as compared to species associated with shrubs and trees. Data of 59 European and North American species compiled from Heikertinger et al. (1954).

from early June to mid-July (increase in plant height by a factor of 7.2). Thus, during their seasonal history the three host plants show pronounced differences which must influence their value as a food resource.

As the young tissues of leaves are particularly rich in nutrients (Slansky & Rodriguez, 1987), it could be expected that the larval phenology of the investigated chrysomelid species coincides with the early growing period of their host plants. This is indeed the case with the majority of chrysomelids living on trees and shrubs (Figure 4) and with many other folivorous forest insects, the so called 'early season feeders' (Mattson, 1980) or 'flush feeders' (White, 1993). Among species feeding on herbaceous plants, particularly on hemicryptophytes and geophytes, 'early season feeders' are not a dominant group (Figure 4). In our case studies only *Galeruca tanacetii* belongs to this type, as its larvae started their feeding on the young growth and profited from the nitrogen peak in the leaves of *A. millefolium*. Our field observations show that during the larval feeding period of *G. tanacetii* there exists the risk of overexploitation of individual patches of *A. millefolium*. By switching to other host species *G. tanacetii* can make up this disadvantage.

Thus, larvae of this euryphagous chrysomelid species have been observed on ten different food plant species (Prevett, 1953; Obermaier, unpubl.).

In view of its initially high level of nutrient nitrogen, its vigorous growth and its common occurrence it is understandable that the nitrophilous *C. arvensis* is a larval host plant of stenophagous chrysomelid species belonging to three different taxonomic groups. Only one of these, *Lema cyanella*, can be considered as an 'early season feeder', as its larvae have been found from late April onwards (Zwölfer & Patullo, 1970). Observations on the monophagous *Altica carduorum* by Zwölfer (1965) and on the oligophagous *Cassida rubiginosa*, *C. vibex* and *C. deflorata* (Zwölfer & Eichhorn, 1967) show that these chrysomelids exploit more advanced developmental stages of *C. arvensis*. *Cassida rubiginosa* can develop on many *Cirsium* and *Carduus* species, but at our observation sites only *C. arvensis* was available as a host. Air temperature determines the date when the adults leave their hibernation sites in spring, the start of oviposition (Kosior, 1975), and the start of their larval feeding and pupation period (Ward & Pienkowski, 1978). In our study the first adults on *C. arvensis* were observed when temperatures had reached a daily average of 13 °C and in fall the last adults disappeared at temperatures below 15 °C. One may assume that the synchronization of *C. rubiginosa* with *C. arvensis* is mainly a result of temperature thresholds. Such a function of temperature as a signal for the availability of sufficient food resources has been discussed by Slansky & Rodriguez (1987). Low temperature may by itself be a selection pressure opposing early emergence by slowing down growth or limiting food conversion. Stamp & Bowers (1990) have shown that at 20 °C food quality had no influence on larval growth rate whereas at 30 °C larval growth was significantly faster on young than on mature leaves. Larval feeding starts when the level of leaf nitrogen is already reduced but it profits probably from higher temperatures and a strongly increasing number and size of leaves available for food. *C. rubiginosa* does not overexploit its host. In Central Europe this common chrysomelid with a marked preference for *C. arvensis* (Zwölfer & Eichhorn, 1967) uses on average less than 1% of the available leaf mass of *C. arvensis* (Zwölfer & Brandl, 1989).

Oreina luctuosa with a larval feeding period from July to September belongs to the relatively small group of 'late season feeders'. Its phenology, where the larvae have to feed on low quality food, cannot be explained by a susceptibility to low spring temperatures

but might be connected with the fact that the genus *Oreina* is primarily a taxon distributed in mountain areas (Rowell-Rahier & Pasteels, 1990). Further the mating on flower buds of the host plant (Obermaier, pers. observation) might be a phylogenetic constraint that prevents shifting the activity period. Finally the more humid microclimate in the center of grown up plants might be more favourable for the larvae than in small, young plants. The larvae are able to compensate for the low quality of the food in different ways.

Food is available at low quality but at high quantity at that time. This can be seen as advantageous because of the fact that it is a strictly monophagous species which in the case of an overexploitation of its host cannot escape to other plant species. This dependence on a specific herbaceous host is aggravated by the fact that adults and larvae of *O. luctuosa* have a low mobility and high site fidelity, so that in many populations the access to new patches of *C. scabiosa* would be difficult.

Deficiencies in the quality of a food resource can be balanced by various mechanisms of 'nutritional compensation', which have been reviewed by Slansky & Rodriguez (1987) and Simpson & Simpson (1990). In *O. luctuosa* one adaptation to poor nitrogen levels in its food consists of a tendency to select younger and to avoid older leaves, which in *C. scabiosa* contain significantly less protein nitrogen. Our laboratory tests showed that feeding adults of *O. luctuosa* are able to discriminate strongly between young, mature and senescent leaves of *C. scabiosa* and in choice tests always prefer the younger ones. Under field conditions adult *O. luctuosa* also had a distinct tendency to stay on younger leaves and to avoid senescent leaves of *C. scabiosa*. The discriminatory power of the larvae seems somewhat less developed, as we observed only a clear avoidance of senescent leaves but no significant differences in the responses towards young and mature leaves. This is, however, plausible in view of the slight and statistically not significant differences of the nitrogen level of young and mature leaves. The capacity to feed selectively on younger leaves of *C. scabiosa* with their relatively higher nitrogen concentration improves the nutritional situation of *O. luctuosa* during July to September. A comparison of the data in Table 3 and Figure 1c shows, however, that late in the season even young leaves cannot provide the nutritional quality from which an 'early season feeder' would benefit.

A second response to overcome poor food quality is a flexible adaptation of the food consumption rate during the larval period and a variable duration of the

larval period. When confronted with poor food quality the third-instar larvae increased their daily consumption rate and the total amount of leaf mass consumed was doubled. In spite of this increased feeding activity the daily growth rate declined on poor food quality because reduced levels of protein nitrogen and water caused a reduction of the efficiency of food conversion from 47.5% to 16.1%. Similar effects of poor food quality have been reported in other studies (Mattson, 1980; Tabashnik & Slansky, 1987; Simpson & Simpson, 1990). It is noteworthy that regardless of the quality of the food and its physiological consequences the third-instar larvae reached the necessary weight threshold for molting (approximately 41 mg fresh weight) in all our feeding experiments. As a consequence there is little variation in the body size of adults. Poor food quality, however, leads to an increased feeding period and delayed the molting point. Hence the development of *O. luctuosa* allows a trade-off between food quality and the duration of the feeding period and avoids the disadvantages of a reduced body size. The costs of a slower developmental rate are usually a longer exposure to enemies and other mortality factors (Price et al., 1980, Loader & Damman, 1991). As *O. luctuosa* larvae contain cardenolides as a chemical defence (Dobler & Rowell-Rahier, 1994), it can be assumed that the costs of a prolonged development time are lower than those caused by a reduced body size.

Summarizing the effects of nutrient leaf nitrogen we find different exploitation strategies in the three chrysomelid species investigated as case studies of folivorous insects on herbaceous host plants. *Galeruca tanacetii* is an 'early season feeder' which profits from the high nitrogen levels of the young leaves and compensates the risk of an occasional overexploitation by its euryphagy. *Cassida rubiginosa* attacks *C. arvensis*, a nitrophilous geophyte, in a more advanced developmental phase. The fact that here the initially high concentration of protein nitrogen has already declined, is eventually compensated by higher daytime temperatures which enhance conversion of high quality food. *Oreina luctuosa*, a strictly monophagous chrysomelid, attacks *C. scabiosa* late in the season. It improves the seasonally reduced quality of its diet by selecting younger leaves where the level of nitrogen is relatively high. Moreover the larvae of *O. luctuosa* can adapt the consumption rate and feeding time to offset the effect of poor food quality. It can use, at that time, an ample supply of foliage.

The hypothesis made in the beginning, that the life stages of the beetles which are most dependent on nitrogen, should occur at a time when the nitrogen level in the leaves of the host plant is high, has to be rejected. Larval development of two of three beetle species investigated, does not occur in that time of the season. Diverse species specific selection factors can be made responsible for this and constitute trade-offs with plant quality. Beetle larvae have developed several means to compensate for low food quality and they can profit from a high quantity of food available late in the season.

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