Research

Herbivore damage increases avian and ant predation of caterpillars on trees along a complete elevational forest gradient in Papua New Guinea

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Signals given off by plants to alert predators to herbivore attack may provide exciting examples of coevolution among organisms from multiple trophic levels. We examined whether signals from mechanically damaged trees (simulating damage by herbivores) attract predators of insects along a complete elevational rainforest gradient in tropical region, where various predators are expected to occur at particular elevational belts. We studied predation of artificial caterpillars on trees with and without 'herbivorous' damage; as well as diversity and abundances of potential predators at eight study sites along the elevational gradient (200–3700 m a.s.l.). We focused on attacks by ants and birds, as the main predators of herbivorous insect. The predation rate decreased with elevation from 10% d^{-1} at 200 m a.s.l. to 1.8% d^{-1} at 3700 m a.s.l. Ants were relatively more important predators in the lowlands, while birds became dominant predators above 1700 m a.s.l. Caterpillars exposed on trees with herbivorous damage were attacked significantly more than caterpillars exposed on trees without damage. Results suggest that relative importance of predators varies along elevational gradient, and that observed predation rates correspond with abundances of predators. Results further show that herbivorous damage attracts both ants and birds, but its effect is stronger for ants.

Parasitic and predatory arthropods and insectivorous birds are among natural enemies of the herbivorous insect which can prevent plants from being severely damaged by herbivores. Recent studies show that plants suffering from an attack by herbivores can actively reduce the number of herbivorous insects by attracting predators; this phenomenon, known as 'plants crying for help', is due to a tritrophic interaction, in which the damaged plants are more attractive for natural enemies of herbivores. There is some evidence that predators have evolved to find their prey by detecting the herbivores or by detecting the signals that the infested plant emits.

Such a response has been documented for caterpillars and their parasitic wasps (Klemola et al. 2012). There are also reports on the increased attraction of predatory nematodes (Rasmann et al. 2005), mites (Takabayashi and Dicke 1996), flies (Hulcr et al. 2005), true bugs (Mochizuki and Yano 2007) and thrips (Shimoda et al. 1997) to plants suffering from herbivory.

Behavioural experiments showed that birds could use visible feeding marks on plant leaves and changes in reflectance as cues to find insect herbivores (Mäntylä et al. 2004, 2008a, Boege and Marquis 2006, Müller et al. 2006). More recent work then showed that birds could discriminate herbivore-rich trees also through olfactory detection of volatile organic compounds (VOCs; Amo et al. 2013). This is a relevant results since insectivorous birds are thought to be even more readily beneficial than insects to plants since avian predators can respond quickly and can considerably reduce herbivore load or damage to plants (Van Bael et al. 2003). However, birds can also play a role of possible potential intraguild predators because they can feed on various predatory arthropods and ants (Mooney 2007).

Ants are recognized as another important predator of herbivorous insect in many ecosystems, tropical forests particularly (Hölldobler and Wilson 1990). Their recruitment by herbivore damage-induced plant volatiles (HIPVs) is known to increase rapidly following an attack by herbivorous insects on a host plant (Agrawal 1998). These examples involved ants that had obligate relationships with their host plants and were thus more effective protectors than more common, opportunistic ants having facultative mutualistic relationships with plants (Heil and McKey 2003).

Both insectivorous birds and ants are important predators in most terrestrial communities, and the overall predation of caterpillars is determined to a large extent by their combined effects (Mooney 2007). Yet the strength of trophic cascades between plants, herbivores and with ants or birds

as predators can be quite variable both within (Mooney and Linhart 2006) and among communities (Shurin et al. 2002). The predation pressure experienced by a herbivore is thus a combined result of the local abundance of predators and their preferences for particular herbivores and/or their particular host plants (Perfecto and Vandermeer 1996, Richards and Coley 2007).

The elevational diversity and abundance gradient is one of the most striking biogeographic patterns on Earth (Rahbek 1995), and the role of multitrophic biotic interactions is a pervasive theme in efforts to understand these elevational gradients (Schemske et al. 2009). Elevational trends in predation pressure are poorly known in the tropics since most of the studies have focused on lowland forest (Novotny and Basset 2005), and relatively few studies have investigated biotic interactions along the entire elevational gradient (Schemske et al. 2009). Only few studies describe how predators, prey, and their interactions vary with elevation (Samson et al. 1997, Rodríguez-Castañeda 2012, Tvardikova and Novotny 2012). Therefore, parallel studies about predator diversity and abundance are needed to obtain clear evidence about the predation pressure that herbivores may experience.

Here we use artificial caterpillars exposed on plants across a complete tropical elevational gradient (200–3700 m a.s.l.) in Papua New Guinea to investigate predation pressure by two key predatory groups, ants and birds, and study their response to 1) simulated herbivory on tropical trees, 2) elevation, and 3) abundance of ants and birds in the studied forests.

To our knowledge, predation rate by ants or birds has not been studied along a comparable elevational gradient in the tropics. In a previous study, we observed that the frequency of bird attacks on artificial caterpillars in lowlands (200 m a.s.l.) was lower than at higher elevation (1700 m a.s.l.) while ants caused more attacks in lowlands than at higher elevations (Tvardikova and Novotny 2012). Ants are generally thermophilic, and their diversity rapidly decreases with increasing distance from the equator and with increasing elevation (Samson et al. 1997). On the other hand, homoeothermic birds do not show such rapid decrease in diversity and abundance towards higher elevations (McCain 2009). This means that they could become key predator at the higher elevations, where ants are less abundant; however, we need replicated studies of predation rates across long or preferably complete elevational tropical gradients to test this hypothesis.

Direct studies of predation rates are scarce, particularly in comparison to the studies of herbivory or parasitism, because predation is a momentary event that is particularly difficult to observe in tropical forests. Therefore, predation has also been documented by stomach content analysis, using both morphological and molecular methods of analysis (Symons and Beccaloni 1999). Alternatively, predation pressure can be estimated from attack rates on baits, such as tuna baits for ant predation, or artificial caterpillars for bird predation (Posa et al. 2007, Howe et al. 2009). However, it should be kept in mind that such methods do not measure the abundance of many predatory ant species which a) do not recruit nestmates and b) are not attracted to dead matter.

Material and methods

Our study was performed along an elevational transect on the slopes of Mt Wilhelm (4509 m a.s.l.) in the Central Range of the Papua New Guinea. The complete tropical elevational gradient spanned from the lowland floodplains of the Ramu River (200 m a.s.l., 05°44′S, 145°20′E) to the timberline (3700 m a.s.l., 05°47′S, 145°03′E). The transect comprised eight study sites, starting from 200 m a.s.l. and evenly spaced at 500 m elevational increments. The experiments were conducted throughout the wet season (20 April–20 June 2012 – successively at sites 3700, 3200, 2700, 2200, 200 m a.s.l. – and 15 September–15 October 2012 – successively at sites 1700, 1200, 700 m a.s.l.) to eliminate possible changes in rainfall regime that can affect biotic interactions (Connahs et al. 2011).

Prior to the experiment, we selected 30 experimental trees from 2–3 locally common species at each elevation (listed in Supplementary material Appendix 1, Table A1), which had at least 30 accessible saplings with leaves between 2.5 and 4 m above ground. Selected species did not produce any exudate, sugar droplets, had similar leaf size, did not have any ant nests, and were growing new leaves, largely without herbivore damage. All selected saplings had also similar size and leaf density. We used either 10 saplings from each of three species, or 15 saplings from two species at each elevation. We tried to find trees with low herbivory damage on old leaves, and avoided trees with damage on young leaves.

We used artificial caterpillars exposed on the study trees to monitor attacks by natural enemies. Caterpillars were made from natural-looking dark green colour modelling clay (Koh-I-Noor Hardtmuth, Ceske Budejovice, Czech Republic), which is malleable, oil-based and non-toxic. We modelled artificial caterpillars by pressing the plasticine through a syringe to ensure that each caterpillar had an absolutely smooth surface. Artificial caterpillars were 15 mm long and 3 mm in diameter, matching in body size locally common crambid and tortricid caterpillars, and also matching the median caterpillar size in the entire caterpillar community (Novotny and Basset 1999), as well as the size of caterpillar most commonly taken by birds (Tvardikova 2013). This method has been successfully used in previous studies (Posa et al. 2007, Richards and Coley 2007, Remmel et al. 2011, Tvardikova and Novotny 2012).

Each experiment was conducted along a single 2250 m long transect at each study site. Thirty sampling points, represented by individual trees, were spaced at approximately 75 m intervals along transect. This spacing ensured that the experimental trees could be considered independent, as the transmission of VOCs between two plants requires air contact of up to 60 cm in field conditions (Karban 2007). Ten artificial caterpillars were randomly (between inner and outer leaves) placed on each tree, between 2.5 and 4 m above the ground. Artificial caterpillars were pinned on the distal half of young leaves such that the head of pin was hidden in modelling clay and going through the leaf. Actual caterpillars present on sapling were removed from the trees prior to experiment so they did not bias the herbivore density. We exposed about two caterpillars per m² of leaf area at each elevation, which are natural densities of live caterpillars constant across all studied sites (Novotny unpubl.). A subset of

leaves on every second experimental tree were cut by scissors so that 5% of original leaf area was damaged and removed, simulating herbivory. The damage by scissors (simulating herbivory feeding pattern) was repeated daily (resulting in 5% of leaf area removed every 24 h from each tree) to ensure that potential attraction of leaf damage to predators remained constant for the duration of experiment. The overall experimental damage to leaves was thus increasing from 5 to 25% of leaf area in the course of the experiment. Previous studies showed that only fresh leaf damage attracted ants (Karban 2007). Leaves for experimental herbivory were randomly selected from the entire sapling.

We exposed 10 artificial caterpillars per tree, i.e. a total of 300 caterpillars along the transect at each study site, of which 150 placed on trees with artificial herbivory damage and 150 on undamaged trees. Each caterpillar was inspected (by first two authors and one field assistant who had previous experience with the method, and access to photo reference collection) at 24-h intervals for five consecutive days and carefully examined for characteristic attack marks (Supplementary material Appendix 1, Fig. A3–A4; Tvardikova and Novotny 2012, see also <tvardikova.weebly.com/downloads> for attack mark collection obtained by offering the caterpillars to potential predators captured along the gradient). Probationary blind test showed, that authors and assistants correctly identified 94% attack marks at coarse level (bird, arthropod and mammal) and 80% arthropod attack marks at more precise level (ant, wasp, other arthropod; Supplementary material Appendix 1, Fig. A5).

Caterpillars attacked by two different predators (n = 23) in same day were treated as two independently attacked caterpillars. Missing caterpillars were excluded from the analyses as their status could not be ascertained. All missing caterpillars and caterpillars with marks of attack were replaced by new ones, pinned to approximately the same locations (Posa et al. 2007, Howe et al. 2009). The caterpillars were not pinned to exactly the same place to decrease the chance that ants/birds learn to avoid an inedible larva.

Bird sampling

We surveyed bird communities by two types of census at each elevation – point counts and mist-netting. Point counts (15 min/point) were carried out at 16 points regularly spaced along the 2250 m long transect between 5:45 to 11:00 am. We conducted five point-count surveys during the duration of experiment with caterpillars. Further, we mist-netted birds into a 200 m long line of nets for 3 d (using nets 2.5 m high × 12–18 m long, mesh 16 mm) from 5:30 am to 5:30 pm daily, with regular checks every 20 min. We classified all recorded species into feeding guilds and strategies, and used the number of insectivorous species occurring in understory and mid-story at each elevation for the analysis.

We recorded 33 639 bird individuals of 238 bird species across eight elevational sites on the slope of Mt Wilhelm. Altogether, 236 species and 25 240 individuals were recorded during point-counts, 1354 individuals of 105 species were mist-netted. In total, 118 bird species was identified as insectivorous, and their species richness showed a plateau between 200 and 1700 m a.s.l. (50–46 bird species) and

then decreased to 18 insectivorous bird species at the tree line (3700 m a.s.l.). The most common insectivorous bird species were grey-streaked honeyeater *Ptiloprora perstriata* (811 records, 2200–3700 m a.s.l.), large scrubwren *Sericornis nouhuysi* (759 records, 1700–3700 m a.s.l.), friendly fantail *Rhipidura albolimbata* (716 records, 1700–3700 m a.s.l.), buff-faced scrubwren *Sericornis perspicillatus* (594 records, 1700–2700 m a.s.l.), mountain mouse-warbler *Crateroscelis robusta* (573 records, 1200–3700 m a.s.l.). See Tvardikova (2013) for further details.

Ant sampling

We surveyed ant communities occurring on experimental trees by observation and hand collection, as well as using tuna baits. Observation of ant activity was performed prior to the exposure of caterpillars. The trunk of each tree was examined for 10 min, all foraging ant individuals were counted and voucher specimens were taken for identification. Commercial canned tuna was used in baits, which is a standard method in the studies of foraging ant communities. One tea spoon of tuna was placed as bait under a stripe of gauze at breast height at each experimental tree. Baits were inspected one and three hours following their exposure. All ants present were counted and voucher specimens for each species were collected without disturbing the remaining ants. We used a combination of two methods, to eliminate for known fact, that not all ant species are attracted to bait (Véle et al. 2009). Results of another two studies conducted at the same sites were used to discuss accuracy of our results (T. Fayle – pitfall traps, tuna baits, M. Leponce – canopy baits, beating; pers. comm.).

Statistical analyses

The data across five experimental days were lumped together because daily number of attack was low and did not differ between days (Kruskal–Wallis, H4 = 7.05, p = 0.13, n = 150 – H_4 = 9.26, p = 0.06, n = 150). Prior to analyses, we excluded all unidentified attack attempts (1%) and lost caterpillars (2%) from the analysis. The effect of elevation and herbivory on the incidence of attacks was tested by ANOVA with nested design and two within-category effects. All 30 sampling trees were nested in each of the eight experimental sites. Percentages of caterpillars attacked at each sampling tree were arcsine transformed to meet conditions of normality. Presence or absence of herbivory was used as the first within-sampling effect and the type of predator (birds, ants, wasps or other insect) as the second within-sampling effect. Tukey post hoc tests were performed to inspect differences between elevational sites and predator taxa. Further, numbers of caterpillars attacked by various predators were regressed against the abundances of predators or their species richness. All analyzes were conducted in Statistica 9 (StatSoft 2010).

Results

We exposed a total of 2400 caterpillars for five days, resulting in 12 000 caterpillar-days of exposure, during which we

identified 1790 attack attempts. Number of attack attempts for trees with herbivory was generally higher (median = 5) than on trees without herbivory (median = 3) across the whole gradient. Mean predation along the whole gradient was $10 \pm$ SE 0.8% d^{-1}.

The percentage of attacked caterpillars was significantly higher (effect of elevation, Table 1) in the lowland forests (14.3 $\pm$ SE 5.4% d^{-1} of all attacks at 200 m a.s.l. and 16.6 $\pm$ SE 4.3% d^{-1} at 700 m a.s.l.), and decreased with elevation towards 1.8 $\pm$ SE 1.1% d^{-1} at 3700 m a.s.l.. This pattern was observed for both trees with and without herbivory (Fig. 1). In total, trees damaged by herbivory had significantly more attacked caterpillars (effect of herbivory, 11.4 $\pm$ SE 1.2% d^{-1}; Table 1) than trees without herbivory (8.9 $\pm$ SE 1.3% d^{-1}; Fig. 1). This pattern did not change across the gradient (interaction herbivory and elevation, Table 1). However, the number of attacks on trees with herbivory was significantly higher on trees without herbivory at only the five lower elevations (200, 700, 1200, 2200 m a.s.l.; Fig. 1), but not at the higher elevations.

The majority of all recorded attacks on caterpillars were made by birds (52%), and ants (38% of recorded attacks). Attacks of birds and ants showed significant results (Fig. 2), and drove observed patterns. The attacks by other predators (wasps = 4%, all other insect = 6%) were low. Neither elevation (Tukey post-hoc test results for other insect: p > 0.304, wasps: p > 0.06) nor herbivory (other insect: p > 0.51, wasp: p > 0.08) had significant effect on the number of their attacks. Both ants and birds attacked caterpillars exposed on trees with artificial herbivory damage significantly more than caterpillars exposed on trees without damage (results of Tukey post-hoc tests for birds and ants; Fig. 2). Caterpillars on trees with herbivory damage across all elevations were attacked more by ants than by birds (ANOVA; ants: SS = 175.98, F = 12.027, p < 0.001; birds: SS = 32.46, F = 7.31, p = 0.007; Fig. 2).

Birds attacked the highest number of caterpillars at 700 m a.s.l. when predation on both the control trees and herbivory trees was summed, and also only when herbivory trees were considered. The predation rate of birds was dependent on number of insectivorous bird species ($R^2 = 0.82$, $F_{1,6} = 28.62$, p = 0.002, n = 8) and abundances of insectivorous birds recorded ($R^2 = 0.78$, $F_{1,6} = 21.36$, p = 0.003, n = 8; Supplementary material Appendix 1, Fig. A2). The number of attacks on caterpillars exposed on leaves with herbivory was significantly dependent on the number of bird species ($R^2 = 0.80$, $F_{1,6} = 24.39$, p = 0.002, n = 8) and bird
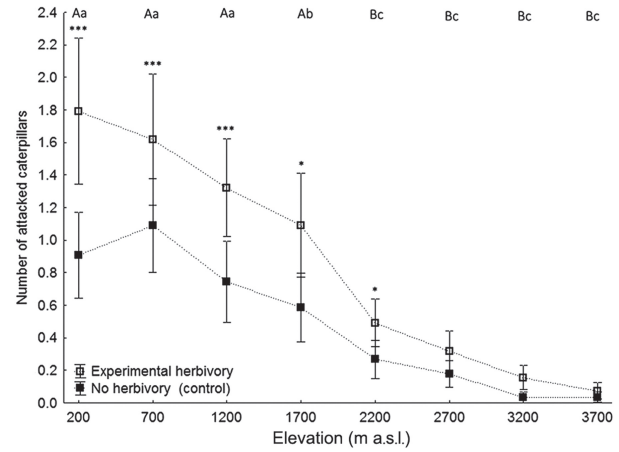

Figure 1. Mean percentage ($\pm$ 0.95 CI) of caterpillar attacks by all predators during 5 d of exposition on one tree with (n = 75) or without (n = 75) simulated herbivory at each elevational site. Sites with significantly different rates of attacks between trees with and without herbivory are marked by asterisks (***p < 0.001, *p < 0.05; Tukey post-hoc test). Elevations with significantly different incidence of attack (p < 0.05) are denoted by different letters; capital letters = trees with herbivory, small letters = trees without herbivory (Tukey post-hoc test).

individuals recorded at each site ($R^2 = 0.78$, $F_{1,6} = 21.63$, p = 0.003, n = 8; Fig. 3). Number of attacks on caterpillars on control trees was dependent on the number of bird species ($R^2 = 0.63$, $F_{1,6} = 10.14$, p = 0.01, n = 8) but not with the number of bird individuals ($R^2 = 0.47$ $F_{1,6} = 5.41$, p = 0.06, n = 8). Birds attacked relatively more caterpillars compared to ants only at elevations above 1700 m
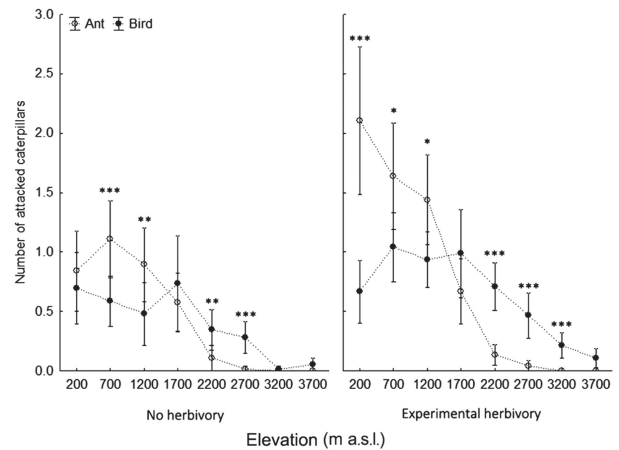

Figure 2. Mean percentage ($\pm$ 0.95 CI) of caterpillar attacks by ants and birds recorded during 5 d of exposition on one tree with (n = 75) or without (n = 75) simulated herbivory. Sites with significantly different rates of attacks by ants and birds are marked by asterisks (***p < 0.001, **p < 0.01, *p < 0.05). Differences in the attack rates on caterpillars between trees with and without herbivory for birds: 200 m – p = 0.43, 700 m – 0.01, 1200 m – 0.03, 1700 m – 0.1, 2200 m – 0.001, 2700 m – 0.15, 3200 m – 0.23, 3700 m – 0.78, and ants: 200 m – 0.008, 700 m – 0.008, 1200 m – 0.01, 1700 m – 0.2, 2200 m – 0.02, 2700 m – 0.15, 3200 m – no variance, 3700 m – no variance. Results of Tukey post-hoc test from repeated measures ANOVA are presented.

Table 1. Effects of elevation (8 sites, 200–3700 m a.s.l., 500 m elevational increment), and herbivory (present or absent), and predator (bird, ant, wasp, other insect) on the incidences of attack on caterpillars. Repeated-measures ANOVA with two within effects.

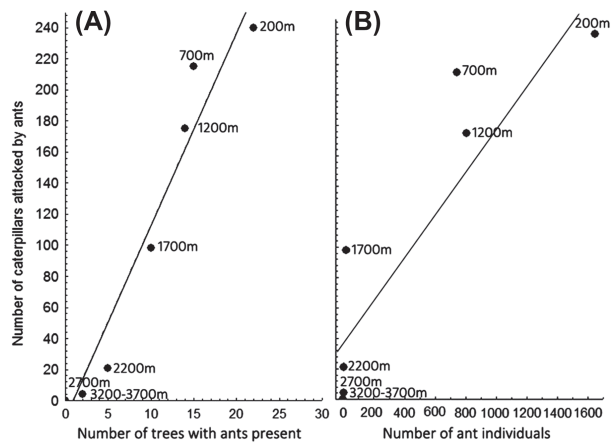|	SS	DF	MS	F	p
Elevation	5.06	7	0.72	100.36	< 0.001
Herbivory	0.41	1	0.41	61.11	< 0.001
Herbivory × Elevation	0.11	7	0.01	1.39	0.085
Predator	5.45	3	1.81	284.55	< 0.001
Predator × Elevation	3.58	21	0.17	26.68	< 0.001
Herbivory × Predator	0.06	3	0.02	3.87	0.009
Herbivory × Predator × Elevation	0.28	21	0.01	2.32	< 0.001

Figure 3. The number of trees with ants present influences significantly the number of caterpillars attacked by ants during whole experiment (A). The abundance of ants on experimental trees measured at tuna baits on trees with and without experimental herbivory together did influence the number of attacks less significantly (B).
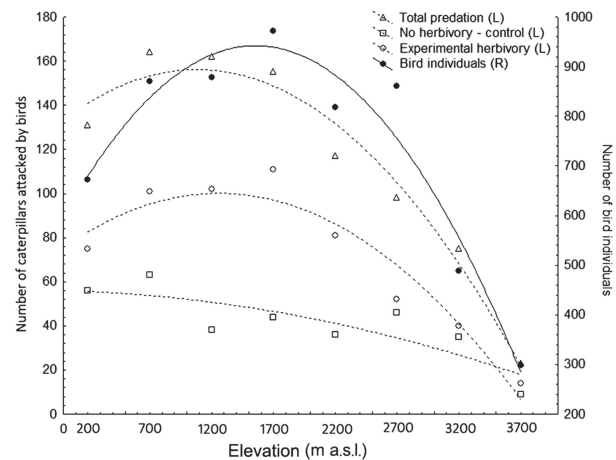

Figure 4. Number of caterpillars attacked by birds during the whole experiment correlated with number of bird individuals present at site. Total predation by birds $= 133.2721 + 0.0423 \times x - 1.9595E\text{-}5 \times x^2$, on control trees $= 56.0379 - 0.0016 \times x - 2.3571E\text{-}6 \times x^2$, and on experimental herbivory trees $= 75.3329 + 0.0394 \times x - 1.5667E\text{-}5 \times x^2$ (left y-axis). Individuals of insectivorous birds observed $= 596.5814 + 0.4444 \times x - 0.0001 \times x^2$ (right y-axis).

a.s.l. (Fig. 2). In an alternative test, the missing caterpillars were treated as if predated by birds, but it had no significant influence on the results ($F_{1,23998} = 0.56$, $p = 0.45$).

Ants attacked the largest number of caterpillars at lower elevations whilst only 4 caterpillars (0.3%) were attacked by ants at 2700 m a.s.l. during whole experiment. No ant attacks were observed at 3200 and 3700 m a.s.l. The number of caterpillars attacked by ants was significantly dependent on number of trees infested by ants at each elevation (direct observations $R^2 = 0.91$, $F_{1,6} = 64.95$, $p < 0.001$, n = 8; exposition of tuna baits for three hours $R^2 = 0.94$, $F_{1,6} = 113.68$, $p < 0.001$, n = 8; Fig. 4).

The caterpillar attacks by ants was also significantly dependent on the number of ant individuals sampled from those trees (observed or present on tuna baits) but explained low proportion of variability in caterpillar attacks (observations $R^2 = 0.63$, $F_{1,6} = 10.34$, $p = 0.038$, n = 8; tuna baits $R^2 = 0.80$, $F_{1,6} = 25.37$, $p = 0.002$, n = 8; Fig. 4). Ant abundance decreased with elevation from 200 to 2700 m a.s.l. (tuna baits: number of trees with ants $= -0.0077 \times$ elevation $+ 22.436$, n = 8; $R^2 = 0.97$; number of individuals $= -954.3\ln(\text{elevation}) + 1584.5$, $R^2 = 0.91$; n = 8).

Discussion

Our results demonstrate the importance of leaf damage as a signal of herbivore presence for ants and birds, the two most important groups of predators in tropical forests. The effect of herbivory is probably important along the entire elevational gradient, although the number of attacks observed above 2200 m a.s.l. was too low for rigorous tests. We reported approximately two times higher daily predation on trees with damaged leaves than on control trees, which corresponds to other studies reporting on the effect of herbivory on predation or attractiveness of predators to herbivore infested plant (Kessler and Baldwin 2001). Two times higher

daily predation rate makes caterpillars present on trees with herbivory much more susceptible to predation during their life-time. The trend is significant both for ants and birds at many elevations, but the response to herbivore is particularly strong by ants.

The phenomenon of induced attraction of carnivorous arthropods by plants in response to herbivory is now well accepted (Takabayashi and Dicke 1996). Leaf damage may be a stronger inducer of ant activity than sole presence of herbivore (Agrawal 1998): study reports as much as a five-fold increase in ants on damaged leaves 12 min after herbivorous damage, and twofold increase in ants on damaged leaves after 24 h, compared to simple undamaged control leaves. Similarly, Romero and Izzo (2004) reported three times more ants on young leaves with damage compare to undamaged leaves.

Decreasing predation rate with elevation leads to the prediction of a higher incidence of anti-predatory defences, such as chemical or behavioural, in the lowlands, particularly against ants (Schmidt 1990). The linear relationship between the abundance of predators and the attack rate on caterpillars suggests that predator/prey ratio remains approximately constant along the elevational gradient. Šipoš and Kindlmann (2012) conclude that even with a constant predator-to-prey ratio, increasing vegetation complexity lower attack rate on prey. We agree that vegetation complexity could lower attack rate. However, we think that it will affect birds (as large, flying animals) very differently from ants (as small, flightless animals). Also our result implies that the efficiency of predators in finding their prey does not change with elevation, despite observed decrease in vegetation complexity with elevation (Tvardikova 2013), or to predators that are chemically oriented to prey on short distances, despite they can be visually oriented to prey on larger distances.

The results of experiments with artificial caterpillars have to be interpreted with caution, as clay caterpillars

provide only visual cues to their natural enemies; hence it does not test for predators react to defensive or deterrent behaviours (Gentry and Dyer 2002). Ants often attack anything unusual or artificial in their territory (e.g. clay caterpillars), and the method doesn't allow us to distinguish the attempted predation from other attacks. It is therefore possible that number of attacks by ants is mainly index of their abundance. Further, insect feeding on plants under natural conditions leaves other traces that could reveal them to natural enemies (Gentry and Dyer 2002). Such traces could be visual (e.g. excrements, leaf damage) or chemical (i.e. plant volatiles, which could be both general and species specific; Turlings et al. 1995). Specific clues could be very different for individual natural enemies and, and likely different enemy taxa handle live insect prey in different ways (Hölldobler and Wilson 1990, Dyer 2002). The design of our study is undoubtedly biased towards generalist predators that queue out on visual and mechanical plant damage. The method we used could therefore lead to the exclusion of important specialist predators.

Overall, this method provides results that are probably conservative and interpretation of the results is not without problems. Authors of previous studies employing the same method stated that the number of predation incidents are relative (Brodie 1993) and thereby comparable among habitats, provided experimental set-ups are similar (Howe et al. 2009). However, we should keep in mind that not only the interaction (bird–caterpillars, ant–caterpillar) varies along an elevational gradient, but also the identity of plants, birds and ant all change. And of course, if these were real caterpillars, they would change too.

On the other hand, several assessments have found the method to be a valid surrogate for real prey (Gonzalez-Gomez et al. 2006, Low et al. pers. comm.). Incidence of attack on our artificial caterpillars (exposed on leaves with and without herbivory) per 24 h was similar to incidence of attack measured on genuine exposed caterpillars in exclosure experiments ($7.5 \pm 6.7\%$, median = 5.8%, nine studies from both tropical and temperate habitats – Remmel et al. 2011; and consistent with the results from our previous study which used the same artificial caterpillars – Tvardikova and Novotny 2012). Manipulative studies using artificial caterpillars recorded similar daily attack rates of 11.1% (Richards and Coley 2007) in a lowland seasonal forest in Panama, 13.7% in semi evergreen lowland dipterocarp forest in the Philippines (Posa et al. 2007), and 5.8–52.4% in three studies from various tropical areas (Remmel et al. 2011).

Experimental damage on leaves in our study attracted ants as well as birds. Recruitment of arthropods by HIPVs (Agrawal 1998) as well as mechanical damage (Turlings et al. 1990) is well known for long time. Recent study (Amo et al. 2013) revealed that also birds use the volatiles to locate their prey. In previous experiments, birds were attracted to infested trees despite they could not see the herbivore-damaged leaves (Mäntylä et al. 2004, 2008b, Amo et al. 2013). In these studies trees emitted also systematic volatiles induced by caterpillar herbivory. In contrast, in our study, birds were attracted to trees with damaged leaves obtained by a mechanical damage which alone does not have to lead to emission of some specific chemicals (Turlings et al. 1995).

We used mechanical damage to simulate herbivory on foliage and this may be a less efficient cue to predators than the damage done by herbivores feeding (Turlings et al. 1990), therefore underestimating the significant effect of predators observed in this study. From the literature it is unclear for how long would a tree with high leaf damage remain attractive to predators. Bolter et al. (1997) reported that wounds healed rapidly after cutting, and their attractiveness to herbivores disappeared shortly after cutting the leaves. In other experiments, emissions of compounds that attracted predators and parasitoids waned within minutes after mechanical clipping, but remained 1–3 d after actual chewing damage or application of insect regurgitant (Turlings et al. 1995). In contrast, Karban (2007) reported that mechanically clipped shoots attracted predators for up to 6 d following clipping. In order to compensate for lower efficiency of mechanical damage, we decided to repeat it daily during our experiments. We expected that response to damage is systemic, and undamaged leaves of injured plants also emit terpenoids based signals (Turlings and Tumlinson 1992). Future studies of how ants respond to VOCs released by damaged plants across elevation are needed to understand what influences predation by ants across elevation.

It is worth noting that a daily mortality rate of 1, 5 and 20% over the 3-weeks caterpillar lifespan would produce overall mortality of respectively 19, 66 and 99%. However, such extrapolations and conclusions should be taken discretely, as a natural caterpillar would probably have chemical or mechanical (hairs) defence, it could hide or change its movement behaviour. We assume that the predation rate observed in this study is plausible since it corresponds to what is available from the literature in both temperate climate in Kentucky, USA (78%; Choate and Rieske 2005) and in tropics in Campinas, Sao Paulo, Brazil (68%; Gomes-Filho 2003). Further, even if our experiments using models of caterpillars did not provide an estimate of natural predation rates, the relative number of predation incidents should be comparable among habitats (Howe et al. 2009) for individual species of generalist predators.

Schwenk et al. (2010) did not find any effect of elevation on bird predation of arthropods between 290 and 780 m a.s.l. in White Mountains of New Hampshire, while we found higher predation rate at 1700 m than at 200 m a.s.l. in Papua New Guinea (Tvardikova and Novotny 2012). This was consistent with the pattern observed in the present study, where the highest predation by birds was in mid-elevations (700–1700 m a.s.l.). The higher abundances of insectivorous birds in mid-elevations, and the relatively gradual decrease of their abundances have been observed in many other studies (McCain 2009). These suggest that birds drive the predation rates of caterpillars at elevations, where ants are relatively rare (i.e. 1700 m a.s.l.; Fig. 2). Thus, relative importance of predation of ants is more important towards the lowland forests (200–1700 m a.s.l.) and it decreases with elevation; whereas bird predation becomes more important at mid-elevations (1700–2700 m a.s.l.).

We observed a dramatic drop in abundance of ants at high elevations which correlated with the predation rate observed. This result substantiated previous studies which observed markedly reduced species richness and abundance of ants at

higher elevations (Samson et al. 1997), most likely resulting in a decrease in predation pressure on herbivorous insect. The number of trees and baits occupied by ants proved to be better correlated with predation rates than the total abundance of ants in the samples. It appeared that abundance of ant individuals influences only the number of bites into one caterpillar, and not the number of predated caterpillars.

In conclusion, our study demonstrates 1) the large importance of plant damage as a cue of herbivore presence for predators (particularly true for ants but also observed in birds) 2) decreasing attack rate of predators with increasing elevations in tropical forests, and 3) a transition in predator dominance from ants in the lowland forests to birds at the mid to high elevations. Further, the change in dominant predator group with elevation could lead to dramatic changes in anti-predation strategies of herbivores, and the structure of local food webs, along elevational gradient.

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