Predator identity and the nature and strength of food web interactions

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Summary

1. Most trophic interaction theory assumes that all predators are an abstract form of risk to which prey respond in a quantitatively similar manner. This conceptualization can be problematic because recent empirical work demonstrates that variation in the responses of prey to different predators can play a key role in structuring communities and regulating ecosystem function.

2. Predator identity – the species specific response of prey to a predator – has been proposed as an ultimate mechanism driving the relative contribution of indirect effects in food webs; however few studies have explicitly tested this hypothesis.

3. This study explores the impact of predator identity on direct consumptive (CE) and non-consumptive effects (NCEs), and on the relative contribution of indirect, density and trait-mediated effects in trophic cascades within host-parasitoid communities.

4. We systematically compared the individual, host-parasitoid-plant interactions of two actively foraging parasitoid species with disparate foraging styles, one aggressive and one furtive, a common aphid host and plant. Our results demonstrate that the degree of risk aversion by prey to each particular predator species (i.e. predator identity) is a key factor driving the nature and strength of direct and indirect transmission pathways.

5. Both parasitoid species, in general, had a negative impact on plants. The magnitude of the aphid anti-predator dispersal response was positively correlated with plant infestation and plant damage. The qualitative effect of predator-induced infestation of new plants superseded the quantitative effects of predator-mediated reductions in aphid numbers.

6. The greatest indirect impact on plants was generated by the aggressively foraging parasitoid, and the strength of the aphids anti-predator response (a NCE) antagonistically traded-off with CEs due to an increased investment in attempting to capture risk-sensitized prey. In contrast, the furtive parasitoid did not elicit a strong anti-predator response, had little indirect impact on plants, but generated very high CEs due to the advantage of ovipositing into a sedentary prey population.

7. Our data suggest the responses of prey to different predatory cues may be an important mechanism driving the relative contribution of transmission pathways in trophic cascades. We conclude that predator identity is a key factor influencing the nature and strength of food web interactions.

Key-words: adaptive behaviour, *Aphelinus abdominalis*, *Aphidius matricariae*, *Aulacorthum solani*, community dynamics, food web, host-parasitoid interactions, indirect effects

Introduction

An underlying assumption in most trophic interaction models is that all predators pose threats, to which prey respond in a quantitatively similar manner (e.g. Diehl *et al.* 2000; Krivan & Sirot 2004; Peacor 2003; Lima 2002; but see Schmitz, Krivan & Ovadia 2004). This approach necessarily abstracts the mechanistic details about the ecological inter-actions among species within food webs. However, this can be problematic because studies have shown that interspecific variation among predators, and even intraspecific variation within a predator species (Post *et al.* 2008), can have significant impacts on community structure and the regulation of ecosystem function (e.g. Carpenter *et al.* 1987; Peckarsky & McIntosh 1998; Pace *et al.* 1999; Gelwick 2000; Bernot & Turner 2001; Schmitz & Suttle 2001; Schmitz 2008). The importance of individual species interactions in food webs argues for greater effort to uncover the factors driving

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transmission pathways in order to develop a more comprehensive theory of community dynamics.

The nature of species interactions in ecological communities dictates, among other things, the direct and indirect effects that are transmitted through food webs. In classic population ecology, predators ('initiator') directly reduce prey ('transmitter') abundance thereby having a positive effect on the basal resource ('receiver') (e.g. Rosenzweig 1973; Oksanen *et al*. 1981). Thus the effects generated by direct consumption ('consumptive effects' – CEs) may indirectly influence the basal resource through the reduction of prey density thereby generating density-mediated indirect effects (DMIEs) (Abrams 1995). More recently, studies have shown that predators can also directly cause non-consumptive effects (NCEs) through modifications of prey behaviour, growth or development when in the presence of predators, which can change the way transmitting and receiving species interact (Abrams 1995; Werner & Peacor 2003; Preisser, Bolnick & Benard 2005). Thus predator-induced NCEs may also produce associated indirect effects that are transmitted through trophic levels (i.e. trait-mediated indirect effects (TMIEs), see Abrams 2007 for a discussion of terms), which can be additive (positive TMIE) or compensatory (negative TMIE) to the indirect effects generated by direct consumption. In most ecological communities CEs and NCEs operate simultaneously. Therefore, indirect effects associated with the two pathways can be difficult to assess empirically due to confounding interactions between TMIEs and DMIEs (Werner & Peacor 2003 for review). The phenomenon known as a trophic cascade – the indirect effects of carnivores on plants mediated through herbivores – can therefore be driven primarily by a single pathway or a combination of both CEs and NCEs (Okuyama & Bolker 2007); however, factors influencing the relative strengths of each pathway are poorly understood.

Schmitz, Krivan & Ovadia (2004) hypothesized that, ultimately, trophic cascades may be determined by the behavioural responses of prey to different predators. This hypothesis suggests that there is a continuum of ways that prey respond to different predator species, likely based on the costs and benefits of responding to predators with particular hunting modes (i.e. actively foraging, sit-and-wait and others) in different foraging domains (Schmitz & Suttle 2001). Under this theory the relative contribution of TMIEs and DMIEs in trophic cascades can be predictable by the hunting mode of the predator. Alternately, prey may be responding based on the amount of information they have about each predator (i.e. predator identity) and the strength of the prey's response (NCE) is based on the degree of risk aversion to each particular predator species (Bouskila & Blumstein 1992; Sih 1992).

In this study we focus on the role of predator identity in mediating the nature and strength of direct and indirect effects in host-parasitoid communities by controlling for the foraging mode of the predator. We systematically compare the interactions of two different species of actively foraging parasitoids on a common aphid host and basal resource. The foxglove aphid, *Aulacorthum solani* (Kaltenbach) (Hemiptera: Aphididae), is a phloem-feeding insect that causes extensive leaf curling, chlorosis, and impedes plant growth, and has several adaptive behavioural defences, including a predator-induced dispersal response. Both parasitoids are widely distributed, natural enemies of the foxglove aphid and are indigenous in the native range of *A. solani* (Takada 2002; Mackauer & Stary 1967). *Aphidius matricariae* (Haliday) (Hymenoptera: Aphidiidae) is an aggressive parasitoid that has a quick sting (< 1 s), can sting moving hosts, and elicits a strong anti-predator response in aphids resulting in dispersal (NCE). In contrast, *Aphelinus abdominalis* (Dalm.) (Hymenoptera: Aphelinidae) uses a furtive form of attack. It cautiously moves through aphid patches, has a much slower sting (> 1 min), requires a sedentary host in order to oviposit, and elicits a very weak anti-predator dispersal response in aphids. This model system allowed us to investigate the impact of predator identity on transmission pathways with actively foraging predators that occupy the same guild and foraging habitat, but are recognized and responded to differently by the aphid host (transmitter species). Our goals for the current study were to:

1. Quantify the variation in direct consumptive and non-consumptive effects when aphids are exposed to natural enemies with different foraging strategies.
2. Determine the influence of predator identity (i.e. predator-based cues and causes) on the nature, strength and relative contribution of indirect effect pathways in trophic cascades.

Materials and methods

EXPERIMENTAL OUTLINE

This study draws from three experiments that break down the behavioural and community level interactions causing trophic cascades in aphid-parasitoid systems. The first experiment quantifies the variation in the behavioural response of an aphid species when exposed to parasitoids with different foraging styles, one aggressive and one furtive, using small patches of aphids exposed to single parasitoid females. A second experiment explores how the differences in behavioural responses of the prey impact direct and indirect species interactions at a community level using a mesocosm experiment. A final experiment dissects the interaction between the degree of predator-induced risk-sensitization in prey (NCE) and the opportunity for predation (CEs), thus demonstrating a key factor influencing the relative contribution of trait- and density-mediated effects causing trophic cascades.

MAINTENANCE OF INSECT COLONIES

Foxglove aphids were collected from commercial pepper greenhouses (Abbotsford, British Columbia) and maintained on excised leaves of sweet pepper, *Capsicum annuum* L. (Bell Boy, Stokes Seeds St. Catherines, ON, Canada), in 500 mL plastic cups in the laboratory for 1-month prior to the start the experiment to allow the aphids to acclimate to laboratory conditions. *Aphidius matricariae* and *A. abdominalis* were reared on *Myzus persicae* (Sulzer) (Hemiptera: Aphididae) feeding on pepper. Parasitoids emerged in glass jars, and were allowed 1–4 days (depending on parasitoid species) to mate

and acclimate to laboratory conditions. All experimental insects were maintained at 20 ± 2 °C and a 16L : 8D photoperiod. Test parasitoids were always 1–2 day old *A. matricariae* and 2–4 days old *A. abdominalis* (ages based on female responsiveness to aphid hosts, D. Gillespie pers. obs.) naïve females (no contact with hosts prior to experiment) that had been given continuous access to a 10% honey solution, water and males. Sweet pepper, cv Bell Boy, was used as a basal resources in all experiments.

PREDATOR IDENTITY AND NON-CONSUMPTIVE EFFECTS – QUANTIFYING A. SOLANI DEFENSIVE BEHAVIOUR

We determined the influence of predator identity on the strength of the anti-predator dispersal response in aphids (i.e. a NCE). The response of aphids was examined at the aphid patch level. A single female parasitoid was transferred into a 50 × 9 mm Petri dish (FALCON®, Franklin Lakes, NJ, USA) containing a 2 cm diameter leaf disc with 5–10 aphids of mixed instars. Each parasitoid was allowed to oviposit in a single aphid in the patch, and the patch response was recorded. Patch-level responses were: no disruption (no aphids moved); local (only the stung aphid was disrupted and emigrated); patch (the stung aphid plus at least one neighbouring aphid dispersed); and global disruption (stung aphid produced alarm pheromone, resulting in a global aphid dispersal response). Aphids were considered dispersed if they left the leaf disc, which in most natural scenarios would result in the aphids dropping off the plant. A Chi-squared test was used to analyse differences in the proportion of aphid patch responses when exposed to each parasitoid species.

VARIATION IN PHYSIOLOGICAL ABILITY TO PARASITIZE A. SOLANI

The probability of parasitism following a single oviposition was assessed. Parasitoids were allowed to sting an aphid once, and the aphid was then transferred into a rearing cup where the outcome of the parasitism event was recorded (mummy formation, aphid deceased or aphid alive). This was repeated 49 and 52 times for *A. abdominalis* and *A. matricariae* respectively and analysed across species using a Chi-square.

INFLUENCE OF PARASITOID FORAGING BEHAVIOUR ON DIRECT AND INDIRECT EFFECTS

Experimental setup, treatment structure, and data collection

Experiments on the impacts of predator identity on species interactions and trophic cascades were performed in 0·61 m² Bugdorm® cages (BioQuip, Rancho Dominguez, CA, USA). Uniform plots of plants were established by transferring 49, 3-week old pepper plants into square (7 × 7 cell) plant trays (one plant per cell). Plots were fertilized with 1% W/V, 20–20–20 water soluble fertilizer, and allowed to grow for an additional 3 days under high pressure sodium (HPS) lights 16L : 8D photoperiod in cages, after which caged plant plots were dispersed in three experimental rooms. The rooms were maintained at a 16L : 8D photoperiod using a mix of GE® plant and aquarium fluorescent bulbs and normal fluorescent bulbs at a temperature of 20 ± 3 °C. Four cages were housed in each room. The experiment was completely randomized within room and blocked by room so that each room had an equal number of replicates of each treatment (*n* = 6) over three repeats in time.

Experimental treatments were: plants without aphids, plants with aphids only, plants with aphids and *A. matrcariae* females, and plants with aphids and *A. abdominalis* females. A cue treatment for each parasitoid species simulated parasitoid foraging, but without female parasitoids, thus controlled for CEs and DMIEs.

The cue treatments received disturbances to simulate parasitoid foraging that were based on the aphid anti-predator response laboratory assays. The presence of *A. abdominalis* was simulated by gently prodding the aphid's abdomen with the blunt end of an insect pin (5–10 aphids, each on separate leaves), twice daily, which resulted in local or patch level aphid disruption. Prodding 5–10 aphids twice daily was also used to simulate the presence of *A. matricariae*, and an additional four 2nd instar aphids (from an outside source) were pinched behind the head with a pair of fine tip forceps to release alarm pheromone, killed and placed near aggregations of aphids producing large, long lasting signals of the presence of a predator that caused most of the aphids on a plant to disperse (i.e. global disturbance). Male parasitoids of each species were added to both cue treatments to produce physical disruption through aphid contact; males forage for females on host plants in a manner similar to female parasitoids.

At the start of each trial, one hundred foxglove aphids (*c.* 50 juveniles and 50 adults) were transferred to small pepper leaves in individual cups. After all aphids had settled (*c.* 24 h) the aphids (and leaf) were carefully placed on the central plant of each plot. The following day, in the parasitoid treatments, five parasitoids were released under the central plant. To maintain foraging activity, the parasitoids were removed and five new parasitoids were released under the central plant every 3 days. This ensured parasitoids maintained consistent foraging activity, irrespective of species (parasitoids were all at their peak foraging age), over the course of the experiment. All female parasitoids were allowed to sting a single foxglove aphid prior to release to acclimate the parasitoids to the host species as well as to ensure the females were actively foraging. Male parasitoids were acclimated to foxglove aphids in a similar manner. Trials were stopped after 10 days as the plots was only large enough to maintain the rapid reproduction of aphids without saturating the plants and to control for the population of parasitoids, as after 10 days the new generation of parasitoids would emerge thus confounding predator density.

Parasitoid-aphid-plant interactions

The number of plants infested, and the number of aphids on each plant was recorded on each of the first 3 days and then every other day until the 10th day. Counts were performed before the first of the two daily disturbances was applied to the cue treatments. Aphid emigration was assessed using the numbers of plants infested over the first 3 days of the experiment. This measure of dispersal was analysed from the initial point infestation to day 3 only in order to examine aphid emigration due to the anti-predator response of aphids, while controlling for emigration due to crowding (numbers of aphids, and thus dispersal due to crowing, differed between treatments past day 3). Aphid dispersal and population size were analysed with a Generalized Linear Model using a GENMODE procedure (SAS, 1999). An additional, generalized estimating equation (GEE) was included with an AR1 correlation to account for repeated measures of experimental units over time. Variables for dispersal included the number of plants infested, time and the interaction between number of plants infested and time. Variables for aphid population size included the number of aphids per plot, time, and the interaction between population size and time. A Poisson distribution with a log link function was used for

both dispersal and population size (the scale parameter 'dscale' was estimated by the square root of 'deviance/degrees of freedom').

Aphid aggregation (*J*-index, Ives 1988) was used to measure differences in aphid movement between treatments over 10 days:

$$J = (S^2/\bar{x} - 1)/\bar{x} \qquad \text{eqn 1}$$

where $\bar{x}$ = mean aphids per plant in each sample plot and s^2 = plot variance. The *J*-index is similar to other commonly used aggregation indices, but it is less vulnerable to biases through differences in density or sample size (Rohlfs & Hoffmeister 2004). *J*-values were compared across treatments over time to determine the differences in aggregation using repeated measures MANOVA (correction for sphericity violation as in O'Brien & Kaiser 1985). *J*-values are predicted to decrease (become less aggregated) over time in all treatments as aphids reproduce and spread from a point infestation to the finite number of plants in each plot.

The influence of parasitoid species on aphid-plant interactions was investigated through the total number of plant-infested days (PID), which is the sum of the total number of plants infested on each sample day, over the course of the experiment.

$$\text{PID} = \sum_{i=1}^{10} (\# \ of \ plants \ infested)_i \qquad \text{eqn 2}$$

where *i* is the days that the number of infested plants were sampled. Total plant-infested days were analysed across treatments using ANOVA.

On day 10, all aphids from each of the plots were counted and those from the plots containing parasitoids were transferred into rearing cups. Aphids were maintained in these cups with fresh leaves supplied as needed until all of the parasitized aphids had formed mummies (parasitoid larvae pupation), thus generating a measure of direct consumption of aphids by each parasitoid species. The number of mummies per treatment was analysed with ANOVA.

Cascading trophic interactions

The indirect impact of parasitoids on plant growth, transmitted through the aphids, was compared across treatments. On day 10 the number of plants showing signs of aphid damage was recorded. Removing and photographing all the leaves from each plot then using the area function in SIGMASCAN PRO 5·0 (Systat Software Inc., Chicago, IL, USA) was used to calculate leaf area. Due to the large number of leaves, area was determined for the first true leaves only. This subsample accurately represented foxglove aphid damage as the damage from the aphid is systemic, and occurs while the plant is developing, thus damaging the entire plant. Additionally, all above ground plant parts in each plot were combined, dried and weighed to obtain total dry plant biomass. Total number of plants damaged, leaf area and plant biomass were analysed across treatments using ANOVA, blocking for room and room by treatment interactions. Two replicate plots were removed from the study due to unhealthy plants prior to the addition the aphids.

NON-CONSUMPTIVE EFFECTS AND THE OPPORTUNITY TO OVIPOSIT

The impact of predator identity on a parasitoid's opportunity to oviposit was examined as a possible mechanism influencing the relative strength of indirect effects in this system. This experiment also addressed an important antagonistic interaction between the transmission pathways observed in the mesocosm experiment: the NCE

can reduce CEs through reduced opportunity to oviposit when prey express anti-predator traits and are risk-sensitized. A patch of 10 aphids on a 2 cm diameter leaf in a Petri dish was exposed to either *A. matricariae* or *A. abdominalis* for 2 min, beginning after the first oviposition. After this first foraging period the parasitoid and any aphids that dispersed from the leaf as a result of the parasitoid attack were removed. The remainder of the patch was then exposed to a second parasitoid of the same species for a further 2 min. Loss of opportunity, and thus the potential to generate direct CEs, was measured by the reduction in the number of prey due to the adaptive anti-predator response after each 2-min period. The number of aphids receiving an oviposition was also recorded in each foraging event. The difference in the number of aphids escaped due to the anti-predator dispersal response as well as the difference in number of ovipositions from the first foraging bout to the second was analysed across parasitoids using a paired *t*-tests.

Analyses were performed using JMP 7·0·2 statistical software (SAS Institute Inc., Cary, NC, USA), except for the GEE analyses which were performed in SAS 8·2 (SAS Institute, Cary, NC, USA).

Results

INTERSPECIFIC VARIATION IN APHID-PARASITOID INTERACTIONS

Predator identity and non-consumptive effects – quantifying A. solani defensive behaviour

The anti-predator dispersal response (NCE) was greater when patches of aphids were attacked *by A. matricariae* compared to those attacked by *A. abdominalis* ($n = 89$, $\chi^2_{(3)} = 33·24$, $P < 0·0001$). When attacked by *A. matricariae*, 6% of aphids did not respond, 34·7% responded locally by moving away from the parasitoid, 30·4% of attacks resulted in the aphid patch being disrupted and 28·2% produced alarm pheromone resulting in a global dispersal. When attacked by *A. abdominalis* 44·1% of the aphids did not respond, 46·5% responded locally, 6·9% of attacks disrupted the patch and 2·3% (one attack) produced global dispersal.

Variation in physiological ability to parasitize A. solani

The outcome of a single oviposition did not differ between the two parasitoid species ($n = 101$, $\chi^2_{(2)} = 0·073$, $P = 0·96$), demonstrating there was no difference between parasitoids in the physiological ability to parasitize foxglove aphids. In 49 aphids attacked by *A. abdominalis*, 29% were parasitized, 19% were dead, and 52% were alive. In 52 aphids parasitized by *A. matricariae*, 27% were parasitized, 20% were dead and 53% were alive.

INFLUENCE OF PARASITOID FORAGING BEHAVIOUR ON DIRECT AND INDIRECT EFFECTS

Parasitoid-aphid-plant interactions

Aphid dispersal was estimated through the number of plants that became infested over the first 3 days of the experiment (Fig. 1). *A. matricariae* and *A. matricariae* cue lead to a rapid
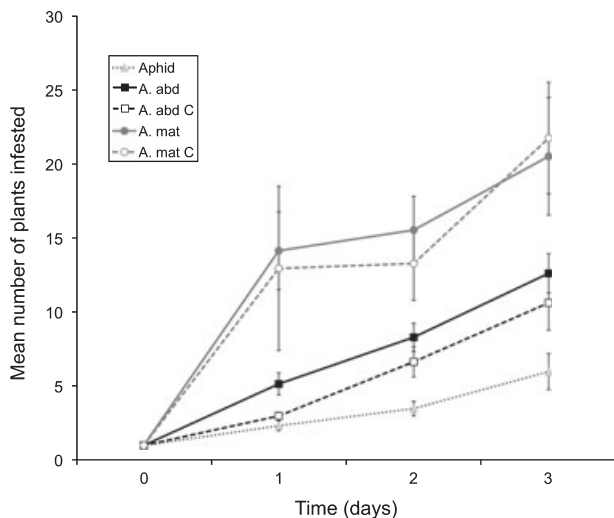
Fig. 1. Effect of parasitoid treatments on plant infestation in caged mesocosm experiments. Treatments were *A. abdominalis* (A. abd), *A. matricariae* (A. mat), aphids only (A) or cue treatments that simulate either *A. abdominalis* (A. abd C) or *A. matricariae* (A. mat C).

increase in the plant infestation rate over the first 24 h (GEE days 0–1 treatment × time $\chi^2_{(4)} = 17.50$, $P = 0.0015$) compared to *A. abdominalis* cue, *A. abdominalis* and the aphids only treatment. Over 3 days there was a significant difference in the number of plants infested between treatments ($\chi^2_{(4)} = 9.67$, $P = 0.04$) with *A. matricariae* and *A. matricariae* cue maintaining higher numbers of infested plants compared to all other treatments, however the rate of infestation was not different across treatments beyond the first day ($\chi^2_{(4)} = 3.18$, $P = 0.53$). Plant infestation did not differ between *A. matricariae* and *A. matricariae* cue (treatment $\chi^2_{(1)} = 1.30$, $P = 0.25$; treatment × day $\chi^2_{(1)} = 1.13$, $P = 0.28$) or between *A. abdominalis* and *A. abdominalis* cue (treatment: $\chi^2_{(1)} = 3.48$, $P = 0.062$; treatment × day; $\chi^2_{(1)} = 0.45$, $P = 0.50$). Thus, both cue treatments were accurately mimicking the respective, anti-predator dispersal response of the aphids. The number of infested plants was higher in the *A. matricariae* treatment compared to the *A. abdominalis* treatment ($\chi^2_{(1)} = 5.50$, $P = 0.02$), demonstrating that the aphids identified and responded to the different parasitoid species by varying the magnitude of their anti-predator dispersal response.

Aphid aggregation (*J*-index) differed among treatments over 10 days (MANOVA: treatment $F_{4,15} = 98.36$ $P < 0.0001$; time $F_{6,10} = 40.40$ $P < 0.0001$; treatment × time $F_{24,36.1} = 3.77$ $P = 0.0002$) (Fig. 2a). As predicted all treatments became less aggregated with time. The aphid-only treatment was the most aggregated, *A. abdominalis* generated an intermediate aphid aggregation and *A. matricariae's* presence resulted in little to no aphid aggregation. Cue treatments were different from corresponding parasitoid treatments. Independent contrast analysis demonstrated that aphids were more aggregated when exposed to *A. abdominalis* compared to *A. matricariae* (treatment: $F_{1,15} = 4.47$ $P = 0.016$), and that aggregation between them differed over time (time × treatment: $F_{6,10} = 4.47$, $P = 0.019$). Both parasit-
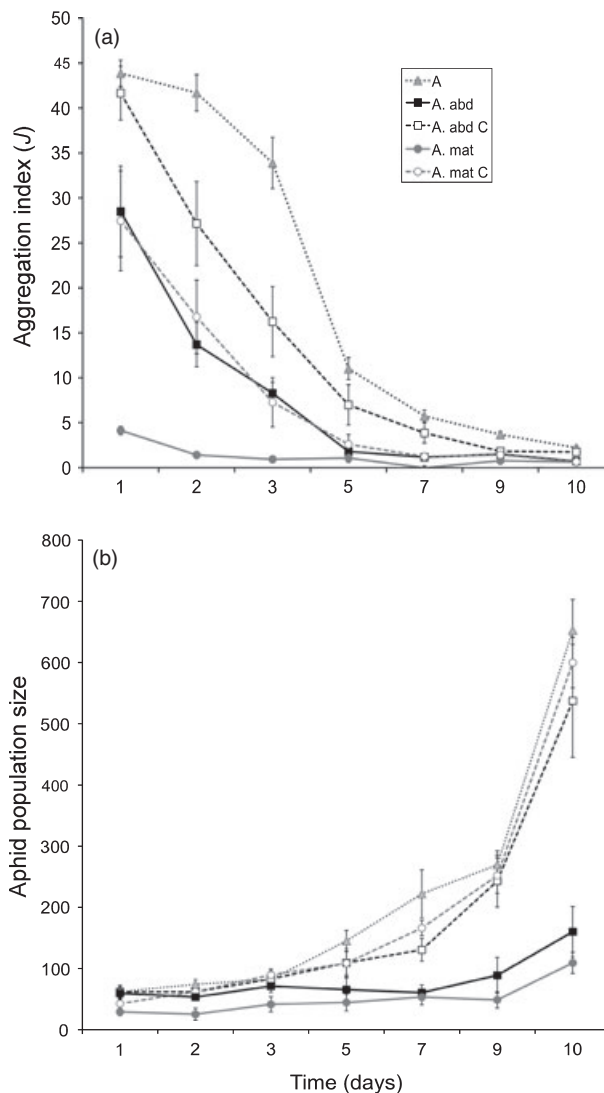

Fig. 2. Effects of parasitoid treatment on aphid aggregation and aphid population size in caged mesocosm experiments. (a) Mean index of aphid aggregation over the full 10-day experiment. (b) Mean aphid population size over the full 10-day experiment. Treatments were *A. abdominalis* (A. abd), *A. matricariae* (A. mat), aphids only (A) or cue treatments that simulate either *A. abdominalis* (A. abd C) or *A. matricariae* (A. mat C).

oid treatments differed from the aphid-only treatment (*A. abdominalis*: $F_{6,10} = 10.29$, $P = 0.0009$; *A. matricariae* $F_{6,10} = 25.07$, $P < 0.0001$).

Aphid population size differed among treatments over time (treatment: $\chi^2_{(4)} = 10.34$ $P = 0.035$; treatment × day; $\chi^2_{(4)} = 13.42$, $P = 0.0094$). There were two distinct groups: the two cue treatments and the aphid-only treatment formed one group; and the *A. abdominalis* and *A. matricariae* treatments formed a second (Fig. 2b). In the former, aphid numbers increased exponentially, whereas in the latter, numbers remained approximately the same as the starting numbers.

The cumulative PID differed between treatments (ANOVA: treatment $F_{4,23} = 5.12$, $P = 0.004$). Tukey's HSD indicated that *A. matricariae* and *A. matricariae* cue generated significantly greater plant-infested days (159.83 ± 15.1 and

173·83 ± 12·6, respectively) than the aphid-only treatment (99·83 ± 9·5), whereas *A. abdominalis* and *A. abdominalis* cue did not (132·22 ± 14·4 and 127·5 ± 12·4, respectively).

Direct consumption of aphids, as indicated by the number of parasitoid mummies, was much greater in *A. abdominalis* (77·3 ± 11·9) than *A. matricariae* (26·57 ± 12·7) ($F_{1,13} = 8·47 P = 0·012$).

Cascading trophic interactions

The number of plants showing aphid damage at the end of the experiment differed among treatments (ANOVA: $F_{4,23} = 37·63 P < 0·0001$). The *A. matricariae* and *A. matricariae* cue treatments indirectly caused the greatest number of plants to be damaged, *A. abdominalis* and *A. abdominalis* cue had moderate levels of plants damaged and the aphid treatment had the lowest plants damaged (Table 1). There was no room effect on the number of plants damaged (ANOVA: $F_{2,23} = 3·11 P = 0·10$).

Leaf area and plant biomass were significantly reduced in several treatments (ANOVA: area $F_{5,16} = 3·22 P = 0·03$; biomass $F_{5,16} = 3·32 P = 0·03$). However, both proxies were also influenced by room (area $F_{2,16} = 9·88 P = 0·002$; biomass $F_{2,16} = 17·04 P = 0·0001$) and plants in one room were consistently smaller than in the other two. Due to the large effect of room on leaf area and biomass, and because there was not a significant interaction between room and treatment (area $F_{10,16} = 1·58 P = 0·2$, biomass $F_{10,16} = 1·4 P = 0·26$), all replicates from the room that had reduced growth were removed and the analysis was repeated on a subset of the data. Exclusion of the one room removed the room effect (area $F_{1,10} = 0·04 P = 0·83$, biomass $F_{1,10} = 1·54 P = 0·14$) and there was still no room by treatment interaction (area $F_{5,10} = 1·27 P = 0·34$, biomass $F_{5,10} = 1·54 P = 0·10$). Treatment influenced leaf area ($F_{5,10} = 3·72 P = 0·04$) and plant biomass ($F_{5,10} = 3·07 P = 0·03$). Leaf area and plant biomass were lowest in the *A. matricariae* and *A. matricariae* cue treatments, followed by *A. abdominalis* and *A. abdominalis* cue, the aphid-only treatment, and greatest in the plants only treatment (Table 1). Only the *A. matricariae* and *A. matricariae* cue treatments differed in leaf area from the aphid-only and plants without aphids treatments.

Table 1. Mean number of plants damaged, leaf area and plant biomass in 49 pepper plants in plots following exposure to treatments with and without aphids, parasitoids

	Mean number of plants damaged	Mean leaf area	Mean plant biomass
Plants without aphids	0	1427·7 ± 151·6[a]	4·19 ± 0·29[a]
Aphids	12·5 ± 1·55[a]	1355·3 ± 43·2[a]	3·83 ± 0·09[a]
A. abd	30·3 ± 1·7[b]	1051·2 ± 89·0[a,b]	3·32 ± 0·18[a]
A. abd C	28·0 ± 2·9[b]	1201·8 ± 77·9[a,b]	3·32 ± 0·25[a]
A. mat	45·0 ± 1·7[c]	1004·4 ± 34·5[b]	2·86 ± 0·37[a]
A. mat C	42·5 ± 2·1[c]	1010·9 ± 61·2[b]	2·94 ± 0·44[a]

Superscript letters indicate significant differences between treatments

NON-CONSUMPTIVE EFFECTS AND THE OPPORTUNITY TO OVIPOSIT

Parasitoid species differed in the reduction of available prey due to the anti-predator dispersal response of aphids over consecutive foraging events (paired *t*-test: $|t|_{0·05(1),9} = 6·21 P = 0·0001$). Exposure to the first *A. matricariae* resulted in an average of 97% ± 2% aphids escaping (1·9 ± 0·27 aphids stung), reducing to 99% ± 1% (0·3 ± 0·15 aphids stung) after the second foraging event. In contrast, only 22% ± 6% escaped from the first *A. abdominalis* foraging bout (1·5 ± 0·22 aphids stung), reducing to 43% ± 9% escaped on the second foraging bout (1·9 ± 0·27 aphids stung). The cumulative number of ovipositions across foraging bouts also differed between parasitoid species ($|t|_{0·05(1),9} = 2·32 P = 0·032$), with *A. abdominalis* ovipositing in a greater proportion of the aphid patch over successive bouts (0·34 ± 0·03) compared to *A. matricariae* (0·22 ± 0·03). This demonstrates that there is a substantial decrease in oviposition opportunity for subsequent *A. matricariae* females due to the displacement of aphid hosts caused by the initial female's induction of a strong NCE. In contrast, there is little difference in opportunity to oviposit in consecutive foraging bouts by *A. abdominalis*, and thus greater cumulative oviposits.

Discussion

Our results demonstrate that the response of prey to different predators is a critical interaction governing the nature and strength of food web transmission pathways. The magnitude of the aphid's anti-predator response (NCE) differed substantially in the presence of the two parasitoid species at both the plot level (Fig. 1) and the patch level. Parasitoid species directly influenced aphid aggregation (Fig. 2a), and indirectly influenced the number of plants infested by inducing aphid dispersal, resulting in a positive correlation between the magnitude of the NCE and resource exploitation by the herbivore. Subsequently, plant fitness (Table 1) was reduced by the cumulative number of plants infested over the course of the experiment. The strength of the NCE caused an equivalent negative cascading effect. This suggests a tight correlation between the magnitude of the prey's adaptive anti-predator response (a NCE) and the reduction in plant fitness (a TMIE) in this system. This result demonstrates that the magnitude of a prey's response to different predator species can play a critical role in determining the consequences of species interactions.

Plant damage did not differ between cue and non-cue treatments, which controlled for CEs (Table 1). Aphid numbers were reduced in the presence of female parasitoids (Fig. 2b), demonstrating that CEs were occurring due to the reduction in aphid reproduction when parasitized. However, there was no correlation between aphid numbers and plant damage even though aphid densities were much higher in treatments lacking female parasitoids (Fig. 2b). The strong correlation between the number of plants infested over time

and plant damage suggests that the magnitude of the anti-predator response of the aphids dictated the number of plants infested, and was the primary determinant of the observed cascading effect. Therefore, the indirect effect of parasitoids on plants was primarily a TMIE not a DMIE. This result does not preclude the importance of DMIEs within aphid-parasitoid system, as DMIEs typically manifest over longer time frames as individuals are removed from the population by predation (Werner & Peacor 2003). However, due to the explicit sequence of events of NCEs (i.e. dispersal and plant infestation) followed by CEs we would predict that relatively large NCEs in aphids, such as when dispersing from *A. matricariae* attacks, would always coincide with greater degrees of TMIEs due to a spatially greater amount of plants infested by the aphids dispersing from a focal point. Although, DMIEs did not contribute much to the net indirect effects of parasitoids on plants under our experimental time frame, *A. abdominalis*, on average, parasitized 47·9% and *A. matricariae* 24·0% of the aphid population. It is possible that over longer time periods *A. abdominalis* × *A. solani* interactions could result in a shift in the relative contribution of indirect transmission pathways that may even reverse the sign of the cascading interaction from the short term weak negative effect (primarily a TMIE) to a positive effect through herbivore removal (DMIE) (Werner & Peacor 2003). The reason a sign shift may occur in this type of predator-prey interaction is due to a reduced anti-predator response (NCE) in the aphids when attacked by *A. abdominalis* allowing for a significant removal of the non-risk sensitized aphids by predation while minimizing plant infestation, and subsequent plant damage (TMIE), caused by predator-induced aphid dispersal. These results suggest that DMIEs caused by CEs may be more prevalent when there is minimal amounts of predator-induced risk-sensitization in prey (e.g. *A. abdominalis* – *A. solani*), where as increasing predator-induced disturbance and prey risk-sensitivity is likely to coincide with a greater relative contribution of TMIEs (e.g. *A. matricariae* – *A. solani*). The relative contribution of trait- and density-mediated effects causing trophic cascades may therefore be governed primarily by the interplay between prey risk-assessment and predator foraging style (i.e. predator identity) as opposed to specific foraging modes of predators as previously suggested (Schmitz & Suttle 2001).

The difference in direct CEs from parasitism between the two parasitoids was surprising, given that there was no difference in the probability of parasitism between the two parasitoid species when ovipositing in a single aphid. This discrepancy suggested that a mechanism exists that affords *A. abdominalis* a greater capability to impose CEs through parasitism than *A. matricariae*, and therefore this type of predator-prey interaction also has the potential to generate greater DMIEs. Our experiment exploring opportunity costs demonstrates that the strength of the anti-predator response can dramatically impact future opportunity for predation and that a strong NCE displaces prey through dispersal or hiding thereby removing opportunity for subsequent predators. Parasitoids in particular could suffer substantial time

and energy costs from predator-sensitized prey due to their reliance on herbivore-induced plant volatiles to locate hosts, and is the likely reason *A. matricariae* has a relatively low parasitism rate in the mesocosm experiment. Predators that generate a strong NCE, like *A. matricariae,* must invest more time in locating and/or capturing sensitized prey, which reduces the potential to impose CEs and any indirect effects associated with directly reducing the prey population through parasitism or predation. In contrast, predators with a furtive foraging style, such as *A. abdominalis*, that do not induce a strong anti-predator response gain a relative advantage in exploiting non-sensitized prey as a resource. Thus an antagonistic interaction exists between the strength of a NCE and the potential to generate CEs (and DMIEs) that is directly related to the way in which prey identify and respond to different predator species. Previous studies have suggested that a significant portion of the net effects of a predator may be attributed to an interaction between predator and prey densities and NCEs (reviewed in Werner & Peacor 2003). Our data demonstrates that NCEs may displace CEs, and subsequently influence the relative contribution of indirect effects in trophic cascades. This result has substantial implications for the relative contribution of transmission pathways in ecological communities as prey species reduce exposure through predation-related risk-aversion in many terrestrial (e.g. Messina 1981; Beckerman, Uriarte & Schmitz 1997) and aquatic systems (e.g. McIntosh & Townsend 1996; Peckarsky & McIntosh 1998; Gelwick 2000).

Indirect effects are thought to play a large role in structuring aphid-parasitoid communities (Muller & Godfray 1999). Our data suggest that the qualitative effects of predator-induced plant infestation actually supersede the quantitative effects of a reduction in aphid density. In addition, aphids are known vectors of many plant viruses so the qualitative effects of aphid-natural enemies interactions may indirectly impact plants by both mediating plant infestation rates by aphids and subsequently disease transmission in plants. Studies have shown that the dispersal response of aphids exposed to natural enemies that elicit strong antipredator behaviour, such as ladybird beetles (*Coccinella califonica*), is correlated with an increased spread of Bean Yellow Mosaic Virus (Roitberg & Myers 1978) and Barley Yellow Dwarf Virus (Smyrnioudis *et al.* 2001). Adaptive movement of prey in response to predation is also thought to be an important factor in metapopulation dynamics (Abrams 2008). The adaptive response of pests to different predator species is therefore an important factor dictating rates of infestation and spatial impact of economically important pest species. These results add to the growing body of literature that suggests predator identity as an important factor in prey suppression (Schmitz & Suttle 2001; Chalcraft & Resetarits 2003) especially in biological control (Denoth, Frid & Myers 2002; Straub & Snyder 2006).

Studies involving multiple predators and the same prey in a single system have provided insight into the role different predators play in resource consumption and in the structuring of ecological communities (Peckarsky & McIntosh 1998; Sih, Englund & Wooster 1998; Gelwick 2000; Bernot &

Turner 2001), however these generally do not address the relative contribution of effect pathways. In a study of species interactions among hunting spiders, a single grasshopper species and two plant species in an old-field food web Schmitz & Suttle (2001) demonstrated that spiders with different hunting modes elicited very different direct and indirect effects in prey, and on the basal trophic level. The relative contribution of pathways within this system ranged from primarily NCEs in the sit-and-wait predator to strictly CEs in the actively foraging spider. The authors concluded that the foraging mode of the predator was likely the primary factor driving transmission pathways (Schmitz & Suttle 2001). Our results strongly support the hypothesis that ultimately trophic cascades are determined by behavioural responses of prey to different predators (Schmitz, Krivan & Ovadia 2004). However, our results clearly show that even within predators that share the same foraging mode (i.e. both active foragers) and forage in the same microhabitat there can be considerable variation in the strength and nature of direct and indirect effects. In our study the most aggressive forager, *A. matricariae*, caused the greatest indirect impact on plants with the strength of the NCE antagonistically trading-off with the capability to directly generate CEs. In contrast the furtive forager, *A. abdominalis*, caused a lesser NCE and a higher CE. Thus the nature and strength of direct and indirect effects may primarily depend on the responses of prey to different predatory cues, not necessarily on specific predatory hunting modes or foraging habitats as this amalgamation of species may conceal the fundamental mechanisms responsible for the structure and stability of ecological communities.

Acknowledgements

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