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Cascading diversity effects transmitted exclusively by behavioral interactions

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ABSTRACT

Consumer diversity generally increases resource consumption. Consumers can also impact other species by altering their behavior, but it is unclear how such nonconsumptive effects scale with diversity. We independently manipulated predator species richness, and the consumptive and nonconsumptive effects of predator communities, to measure the role of each factor in protecting Brassica oleracea plants from caterpillar herbivory. Plant biomass was greatest when diverse predator assemblages induced antipredator behaviors in herbivores, an effect not further strengthened when predators could also kill caterpillars. Predators within diverse communities were more likely to forage on plants, and to disrupt herbivore feeding, reflecting greater aversion to foraging among conspecific than heterospecific competitors. Predator diversity, therefore, initiated behavioral changes at the predator and then herbivore trophic levels, both to the benefit of plants. Our results indicate that strong, emergent species richness effects can be transmitted entirely through behavioral interactions, independent of resource consumption.

Keywords: biodiversity, ecosystem function, interference competition, intraguild intimidation, nontrophic interaction, predator-prey, trait-mediated, trophic cascade

INTRODUCTION

The degree of resource extraction by diverse communities generally exceeds that of species-poor communities (Cardinale et al. 2006). In most cases this is attributed to niche partitioning among species, or interspecific facilitation of resource capture (Hooper et al. 2005). In either
case, diversity effects are generally envisioned as operating through the consumption of resources. However, multi-species interactions also may be mediated by changes in behavior, which may be reflected in biomass changes that are one or more trophic levels removed from the inciting species (Turner and Mittelbach 1990, Schmitz et al. 1997). For example, predator avoidance behaviors by herbivores often come at the cost of lost feeding opportunities, leading to reduced plant damage even when herbivore numbers do not change (Lima and Dill 1990, Werner and Peacor 2003, Preisser et al. 2005). Such behaviorally-mediated indirect effects can be as strong as those induced by the actual consumption of intermediary species (Schmitz et al. 2004). Analyses confined to predator and prey demographics, therefore, may significantly underestimate the effects of predators on community structure (Abrams 1995, Brown et al. 1999, Byrnes et al. 2006, Goudard and Loreau 2008).

For predators, there is good reason to expect complex interactions between consumer diversity and the relative strengths of nonconsumptive and consumptive effects (Schmitz 2007). Behavioral interactions are particularly important in predator communities (Sih et al. 1998), perhaps contributing to the widely varying diversity-consumption relationships observed at this trophic level (Ives et al. 2005, Bruno and Cardinale 2008). Prey consumption can decline at higher diversity levels when intraguild predation is common (Finke and Denno 2004), but increase when predators partition prey (Wilby et al. 2005, Bruno and O’Connor 2005, Finke and Snyder 2008) or facilitate one another’s prey capture (Soluk and Collins 1988). Thus, interactions between predator diversity and predator consumptive effects themselves often are non-additive (Ives et al. 2005). Similarly, the consumptive and nonconsumptive effects of single
predators can be positively or negatively non-additive (Peacor and Werner 2001, Griffin and Thaler 2006).

It is interesting that in terms of predator-predator intimidation, there is not a clear trend as to whether interspecific interactions tend to differ from intraspecific interactions. Studies isolating inter- and intraspecific competition among predators have shown that behavioral interference between heterospecific predators can be more severe than that among conspecifics (e.g., Soluk and Collins 1988, Peckarsky 1991, Wissinger and McGrady 1993); however, one study has suggested the opposite (Björkman and Liman 2005) while still others found inter- and intraspecific interference to be essentially equal (Evans 1991, Shaffer and Robinson 1996, Vance-Chalcraft et al. 2004, Vance-Chalcraft and Soluk 2005). Thus, it may be difficult to predict how changes in predator diversity will influence the nature and strength of predator-predator intimidation. Consideration of the various habitat domains and foraging modes of interacting predator species may provide a useful basis for predicting emergent diversity effects (Schmitz 2007). However, these factors may be dependent on the predator and prey community in which the species is embedded, necessitating natural history information on all potential predator-predator and predator-prey interactions (Schmitz 2007).

In various terrestrial and aquatic systems, there is evidence to suggest that nonconsumptive effects could largely underlie predator diversity effects (e.g., Sih et al. 1998, Byrnes et al. 2006, Prasad and Snyder 2006, Schmitz 2007), but the relative scaling of nonconsumptive effects across diversity levels has yet to be examined empirically (Borer et al. 2002, Goudard and Loreau 2008). This omission is unfortunate given that in nature, higher-order consumers are exerting both consumptive and nonconsumptive effects while imbedded in diverse communities.

In the field within a community of predatory insects, we investigated how the consumptive and nonconsumptive effects of predators scaled with predator diversity. We independently manipulated consumptive and nonconsumptive predator effects, across two levels of predator diversity, and within a fully-factorial design. We then measured the resulting effects on predators, herbivores, and the basal resource of the system, *Brassica oleracea* L. plants. Nonconsumptive effects were isolated by replacing caterpillars killed by predators, and consumptive effects were isolated by hand-removing caterpillars in the absence of predators. These manipulations were catered to reproduce the magnitude of these effects in predator communities differing both in species richness and species composition, ensuring a factorial manipulation of predator nonconsumptive, consumptive, and diversity effects. We found that diverse predator communities promoted increased plant growth by reducing herbivory, but that this emergent diversity effect was transmitted entirely through a chain of nonconsumptive interactions at the predator and then herbivore trophic levels. Thus, when compared to species-poor predator assemblages, the more diverse predator assemblages increased system productivity without increasing herbivore mortality. Our results indicate that behaviorally-mediated effects can be influenced by diversity quite differently than, and independent from, those due to resource consumption. This supports the contention that nonconsumptive interactions warrant greater
attention in biodiversity studies (Borer et al. 2002, Schmitz et al. 2004, Bruno and Cardinale

METHODS

Natural history

Our study focused on the diverse community of predators attacking *Plutella xylostella* L. caterpillars on *B. oleracea* plants. These caterpillars are the dominant chewing herbivore on *B. oleracea* in the northwestern region of the United States (Biever et al. 1992). When threatened by a predator, *P. xylostella* caterpillars drop from the plant, anchored by a silk thread affixed to the leaf underside (Wang and Keller 2002). This behavior is readily observed, and the larvae may remain suspended for minutes to hours on this thread before climbing back up to the plant (Wang and Keller 2002). Since they cannot feed while dangling from the plant, deployment of this antipredator behavior likely carries a fitness cost.

Locally common predators attacking these caterpillars include the ladybird beetle *Hippodamia convergens* Guérin-Méneville, the predatory bugs *Geocoris pallens* Stål and *Nabis alternatus* Parshley, the golden-eyed lacewing *Chrysopa oculata* Say, and the specialist endoparasitoid *Diadegma insulare* (Cresson). This predator community encompasses a wide variety of body sizes, feeding rates, hunting modes, and foraging domains, of the type that has been demonstrated to yield complementary impacts on shared prey (Snyder et al. 2006, Straub and Snyder 2006, 2008, Preisser et al. 2007). At the same time, intraguild predation has been documented among several community members: lacewing larvae may succumb to *Geocoris* and *Nabis* attacks (Rosenheim et al. 1999), and *Geocoris* and *Nabis* prey upon one another (Snyder et
Further, prey may stand to benefit from various manifestations of intraguild intimidation among predators in more diverse communities (Sih et al. 1998, Berger et al. 2008). Assemblages of heterospecific predators, therefore, would be expected to endure antagonism yet also benefit from a degree of complementarity among their constituent species.

Our experiments included green peach aphids, *Myzus persicae* (Sulzer) (Hemiptera: Aphididae), as these aphids are nearly always present on *B. oleracea* plants at our study site and serve as an important prey for many of the predators (Snyder et al. 2006). In a survey of insect communities on *B. oleracea* plants conducted the same year as the field experiment, we found that 98.33% of caterpillar-infested plants also had aphids (*N* = 120 plants sampled; S. A. Steffan, unpublished data). Predators, therefore, would nearly always encounter aphids when hunting for caterpillars in nature, and thus the inclusion of aphid prey provided a more accurate reproduction of the *B. oleracea* insect community than would caterpillars alone. If some predator species are drawn to plants primarily to feed on aphids and, once there, feed opportunistically on caterpillars, this too would be a naturally occurring phenomenon, again justifying the inclusion of aphids.

**Experimental design**

In the field, four factors— predator diversity (*Low, High*), predator nonconsumptive effects (*Fear+, Fear−*), predator consumptive effects (*Lethal+, Lethal−*), and temporal block (1, 2)—were manipulated within a fully crossed, $2 \times 2 \times 2 \times 2$ factorial design, yielding 16 unique treatment combinations, each of which was replicated five times (total *N* = 80; Fig. 1). Our independent manipulation of these factors allowed us to calculate all main effects and
interactions. Experimental units were 60 × 60 × 60-cm field cages (BugDorm-3120, MegaView Science Education Services Co., Taiwan), enclosed on three sides by polyester netting (96 × 26 threads per cm mesh) and on the fourth with a clear plastic panel that allowed easy viewing of cage occupants. In the middle of the clear front panel, a mesh sleeve allowed access to the interior of the cage. Each cage housed, in addition to predators, two *B. oleracea* plants, 20 *P. xylostella* caterpillars, and 40 *M. persicae* aphids (see Methodological details below). The bottom edge of each cage was buried under 5-10 cm of packed soil to block arthropod movement.

**Manipulation of predator diversity**

Replicates of the *Low Diversity* treatment consisted of one species each (drawn from the pool of five predator species) in separate monocultures, while the *High Diversity* treatment included each of the unique draws of four species from our pool of five predator species (i.e., one species was absent from each polyculture) (Snyder et al. 2006). This design controls for the effects of species composition and identity by ensuring that no single composition or species was over- or underrepresented (Tilman 1997, Straub and Snyder 2006). Each of these species compositions was included once at each level of consumptive/nonconsumptive effect manipulation (Fig. 1). Predator diversity was manipulated within a substitutive design, with *Low Diversity* cages receiving four individuals of the same species, and *High Diversity* cages receiving one individual of each of four species.

**Manipulation of nonconsumptive predator effects**
We isolated nonconsumptive predator effects by systematically replacing caterpillars that had been killed. Each cage in this treatment (Fear+, Lethal−) included predators, assembled according to diversity level and species composition, that were allowed to forage freely. Each of these cages was randomly paired with a no-predator control (Fear−, Lethal−) cage, and every day, caterpillar densities in the Fear+, Lethal− cages were restored to no-predator levels by replacing any dead or missing caterpillars. This system of prey replacement allowed us to virtually eliminate prey losses in excess of background mortality, thereby neutralizing the effect of predator lethality in these cages. Every replaced larva was ensured to be the equivalent instar as that of its corresponding control cage, and all replacement larvae were chosen randomly from the same cohort used to originally populate the cages. Our methods obviated the need for physical manipulation of predators or the confinement of un-manipulated predators, and by allowing predators to kill prey, the system preserved the cues associated with predation, such as tactile vibration signatures (Castellanos and Barbosa 2006) and substrate-borne residues (Magalhães et al. 2005). Such cues have been shown to initiate antipredator behaviors in terrestrial communities.

Further, we attempted to minimize any disturbance to caterpillars during the transfer process by never physically grasping or handling the caterpillars. Using a fine camel-hair brush, we would gently touch a caterpillar with the brush tip to initiate a silk-drop. We then rolled this silk thread onto the brush, and with the caterpillar suspended in mid-air, transferred it to a new plant. This methodology allowed us to move caterpillars without risking handling trauma, and also allowed us to deposit caterpillars into cages without touching plant foliage (other than the
caterpillar itself touching down on the leaf). Caterpillar transfers were quick, taking less than 1 min to release a caterpillar into a cage.

Manipulation of consumptive predator effects

To isolate consumptive effects, a system of manual prey culling was employed in cages where predators were absent (as in Griffin and Thaler 2006). Each replicate of the Fear−, Lethal+ treatment (predators absent) was paired with the Fear+, Lethal+ replicate (predators present) with the same predator species composition. Each day of the experiment, we recorded caterpillar densities within Fear+, Lethal+ cages and then duplicated these densities within their corresponding Fear−, Lethal+ cages by manually removing caterpillars. Because the Fear−, Lethal+ cages did not contain predators, the prey experienced density reductions in the absence of any predation cues. To ensure that the culling of prey accurately mimicked predation in the Fear+, Lethal+ treatment (as suggested by Okuyama and Bolker 2007), the developmental stages (instars) of surviving larvae were recorded as well, allowing us to not only duplicate the population trajectory of the prey, but also the particular age structure of the survivors.

We again attempted to minimize disturbance to other cage occupants when removing a caterpillar. The caterpillar selected for removal was gently touched with a fine camel-hair brush to initiate a silk drop. Then, the silk thread was wound onto the brush and the suspended caterpillar lifted out of the cage. Thus, we were able to remove caterpillars without ever touching the plant foliage, and removals could be accomplished quickly (less than 1 minute per caterpillar being removed).
Methodological details

Experiments were conducted at the Washington State University Tukey Horticulture Orchard in Pullman, WA. Block 1 was initiated 27 July 2007, and Block 2 was initiated 25 August 2007.

First, two c. 16-cm-tall *B. oleracea* plants (3-4 unfurled true leaves) were planted in each cage. We released 20 green peach aphids, *M. persicae*, onto each plant and allowed three days for the plants and aphids to establish. We then released 10 *P. xylostella* caterpillars (3rd instars, reared on *B. oleracea* under the same greenhouse conditions) onto each plant in each cage (= 20 caterpillars per cage). This is well within the historical range of *P. xylostella* densities locally under open-field conditions (Biever et al. 1992), and 2007 surveys of local *B. oleracea* fields confirmed this, with per-plant densities ranging from 0 to 23 and averaging 11 caterpillars/plant (S.A. Steffan, unpublished data). On the same day, all enemy treatments were randomly applied to the cages, and the predators released. *Hippodamia, Geocoris,* and *Nabis* individuals were collected on adjacent vegetation within 48 h of the experiment’s start, and only adult females were used. *Diadegma* females and larval *Chrysopa* were collected from greenhouse colonies.

Three response variables were measured every day, in each cage, during the course of this experiment: the number of surviving caterpillars, the number of caterpillars displaced from plants (i.e., silk-drops or otherwise displaced from plants), and the number of each predator species present on the plants. To minimize disturbance to cage occupants, insect counts were accomplished using small hand mirrors that allowed the observer to find insects on all plant and cage surfaces without ever touching plant foliage. Both caterpillars and predators were easily found using this method, such that counting these insects in each cage took only 2-3 minutes per cage per day. At this time, any missing predator was replaced in order to maintain predator
density and diversity levels. Caterpillars were exposed to the predator treatments for 4 days, at which time the caterpillars began spinning cocoons, and the experiment had to be concluded (as pupae, diamondback moths no longer feed and thus no longer transmit the predators’ effects to the plant). Despite the relatively short trial duration necessitated by the caterpillar’s rapid development, our study captured the majority of the diamondback moth’s herbivory potential by spanning the last two of its four instars, during which caterpillars are known to do 90% of their feeding (Boldt et al. 1975). First- and second-instar diamondback moth caterpillars are exceedingly small, and as first-instars, they feed entirely within the leaf (as leaf miners), emerging to feed on external leaf surfaces as second-instars (Wang and Keller 2002). At the conclusion of the experiment, cages were carefully searched and all remaining arthropods collected. Aphids were counted and removed from plants. Finally, the leaves (including petioles) were harvested, dried for at least 4 d at 90° C, and weighed.

Data analyses

Final plant biomass and the total number of caterpillars observed off of plants were analyzed as 2 x 2 x 2 x 2 fully-factorial ANOVAs, including the factors species richness (high, low), nonconsumptive effects (Fear+, Fear−), consumptive effects (Lethal+, Lethal−), and temporal block (1, 2). Total numbers of predators observed on plants was analyzed as a three-way ANOVA including the factors diversity treatment, species identity, and block. Caterpillar densities across the four days of each trial were analyzed using repeated measures MANOVA including the factors block (1, 2) and predators (absent, low diversity, high diversity). Here, only
Fear+, Lethal+ and Fear−, Lethal− cages were included in the analysis because caterpillar densities were directly manipulated in the other treatment combinations.

We tested for emergent biodiversity effects (overyielding) in the plant biomass and herbivore data, as transmitted through either consumptive or nonconsumptive channels. We calculated $D_T$, a metric of polyculture performance relative to the average of its constituent species in monoculture, and $D_{max}$, a metric of polyculture performance relative to that of the single most effective species in monoculture (Petchey 2003). For the $j^{th}$ polyculture, the $D_T$ value was calculated as $D_{Tj} = (O_j - E_j)/E_j$, where $O_j =$ observed effect of the $j^{th}$ polyculture, and $E_j =$ expected effect of the $j^{th}$ polyculture; $E_j =$ summed per-capita performances in monoculture of the species in the $j^{th}$ polyculture (see Appendix C). For each polyculture, a $D_{max}$ value was calculated; here, $D_{max} = (O_j - M_{max})/M_{max}$, where $M_{max}$ was the single “best” performance (greatest consumptive or nonconsumptive effect) of predators in monoculture, and again, $O_j =$ observed effect of the $j^{th}$ polyculture. One-sample $t$-tests were used to determine whether the respective $D_T$ and $D_{max}$ means differed significantly from zero. In the analyses, values significantly different than zero were taken as evidence of emergent diversity effects. All analyses were conducted in SYSTAT (Systat Software, Richmond, CA, USA) (SPSS 1999).

RESULTS

Because effects on plants of our predator manipulations represented the sum of direct and indirect effects through all channels, we first present the plant biomass data. We then work our way up through the other two trophic levels, presenting behavioral and density data for herbivores and then predators.
Plants

Final plant biomass was greatest where diverse predator communities initiated predator-avoidance behavior by caterpillars ( predator diversity × nonconsumptive effects interaction, $F_{1,64} = 6.88, P = 0.011$; Fig. 2a). Consumption of prey also increased plant biomass ( predator consumptive main effect, $F_{1,64} = 20.44, P < 0.001$; Fig. 2a), but this effect was not further strengthened with either greater predator diversity or the co-occurrence of nonconsumptive effects (all interactive consumptive effects $P > 0.05$; Appendix A1; Fig. 2a). Nonconsumptive effects were stronger in Block 2 than Block 1 ( nonconsumptive × block interaction, $F_{1,64} = 10.12, P = 0.002$), but block effects otherwise did not interact with other model terms ( Appendix A1, Appendix B1).

For plants, only the nonconsumptive effects of predators in high diversity assemblages significantly exceeded expectations, based on the summed per-capita performances of the constituent predator species in monoculture: the mean $D_T$ value for the indirect, nonconsumptive effect channel was significantly greater than zero ($t = 7.43, df = 9, P < 0.001$; Appendix C; Fig. 3a), while that of the consumptive (lethal) effect channel was not significantly different from zero ($t = 0.157, df = 9, P = 0.878$). The mean $D_{\text{max}}$ values for the indirect consumptive and nonconsumptive effect channels did not differ from zero ( consumptive: $t = -1.614, df = 9, P = 0.141$; nonconsumptive: $t = -0.069, df = 9, P = 0.946$), suggesting that in terms of either consumptive or nonconsumptive effects, the average performance of polycultures did not exceed that of the single most effective predator species ( Fig. 3a; Appendix C).

14
Herbivores

Mirroring the results for plant biomass, herbivore displacement from the host plant was greater among treatments coupling nonconsumptive effects with diverse predator communities (diversity × nonconsumptive effects interaction, $F_{1,64} = 8.51, P = 0.005$; Fig. 2b). Again, nonconsumptive predator effects were stronger in Block 2 than Block 1 ($F_{1,64} = 5.30, P = 0.025$; Appendix A2, Appendix B2), though the relationship between diversity and caterpillar behavior did not change across blocks (diversity × nonconsumptive × block interaction, $F_{1,64} = 0.212, P = 0.647$; Appendix A2). Displaced caterpillars were observed almost exclusively in cages bearing predators, indicating that the caterpillars generally did not stray from their host plants unless induced by predators (Fig. 2b).

The magnitude of predator-induced mortality grew through time (predator × time interaction; $F_{3,108} = 29.65, P < 0.001$; Appendix A3; Appendix D), but caterpillar suppression was not further strengthened with greater predator diversity (Appendix A4; Appendix D). Overall caterpillar mortality was significantly higher in Block 1 than Block 2 (Appendix A3).

The consumptive effects of predators in polyculture did not exceed what would be expected based on the summed per-capita performances of their constituent species in monoculture: the mean $D_T$ value for the direct consumptive effect channel was not significantly greater than zero ($t = 1.788, df = 9, P = 0.107$; Appendix C; Fig. 3b). Conversely, the mean $D_T$ value for direct nonconsumptive predator effects was significantly greater than zero ($t = 2.67; df = 9, P = 0.026$; Fig. 3b), providing evidence of consistent overyielding among predator polycultures. The mean $D_{max}$ value for the direct consumptive effect channel was significantly less than zero ($t = -4.047, df = 9, P = 0.003$; Appendix C; Fig. 3b), while that of the nonconsumptive effect channel was...
not significantly different from zero ($t = -1.781, df = 9, P = 0.109$). This indicates that the consumptive effects of predators in polyculture were significantly less than, and nonconsumptive effects no different than, the single most voracious or intimidating predator species in monoculture (Fig. 3b).

Aphid densities were reduced in cages including predators (nonconsumptive main effect: $F_{1,64} = 42.06, P < 0.001$; Appendix A5; Appendix E1). However, unlike plant biomass and caterpillar silk-drop behaviors, aphid density was not impacted by an interaction between predator diversity and nonconsumptive effects (diversity × nonconsumptive interaction: $F_{1,64} = 0.25, P = 0.621$; Appendix A5). A significant interaction between predator diversity and block (diversity × block: $F_{1,64} = 5.719, P = 0.020$) appeared to reflect somewhat lower aphid densities in diverse predator communities in Block 1, but somewhat higher aphid densities in diverse predator communities in Block 2 (Appendix E2).

**Predators**

Predator species differed in how diversity impacted their likelihood to forage on plants ( predator diversity × species identity interaction; $F_{4,20} = 3.80, P = 0.019$; Fig. 4a; Appendix A6). This interaction appeared to be driven by the responses of two species, the lady beetle *Hippodamia* and the parasitoid *Diadegma*, both of which exhibited higher per-capita visitation of plants when foraging within diverse communities (Fig. 4a); plant visitation by the other predators was unaffected by diversity treatment. Overall, plant-visitation by predators was significantly higher in polyculture than in monoculture ($F_{4,20} = 8.33, P = 0.009$; Appendix A6).
Predator species differed in their likelihood of dying during the experiment ($F_{4,20} = 7.14, P = 0.001$; Fig. 4b), but there was no difference in mortality, for any species, between Low and High Diversity treatments (diversity × species identity interaction, $F_{4,20} = 1.91, P = 0.147$; Fig. 4b Appendix A7). Thus, there was no evidence that predator diversity influenced predator mortality rates.

When in monoculture, predator species differed both in their likelihood of triggering caterpillar displacements ($F_{4,15} = 4.71, P = 0.012$; Fig. 4c) and in the total caterpillar mortality that they exerted ($F_{4,15} = 4.74, P = 0.011$; Fig. 4d). The lady beetle *Hippodamia* and the parasitoid *Diadegma* were most likely to evoke displacement (Fig. 4c), while the lacewing *Chrysopa* was the most lethal predator (Fig. 4d). In diverse predator communities, displaced and dead caterpillars could not be attributed to a particular predator species.

**DISCUSSION**

Nonconsumptive interactions are increasingly recognized as ubiquitous and important ecological phenomena, influencing not only how species interact but also how communities function (Lima and Dill 1990, Sih et al. 1998, Schmitz et al. 2004, Preisser et al. 2005). However, it has yet to be resolved how the effects of behavioral interactions scale with changes in species diversity. Our fully-factorial manipulation of predator diversity and predator impacts operating through nonconsumptive (behavioral) and consumptive (lethal) channels, allowed us to explore the independent and interactive effects of these factors. We found that trophic cascades were strengthened significantly through an interaction between predator diversity and the nonconsumptive component of the predator effect, with caterpillars significantly more likely to
engage in antipredator behavior, both eating less (Fig. 2a) and dropping from the plant more often (Fig. 2b), when facing diverse than single-species predator communities. While predators also protected plants by killing caterpillars (Fig 2a; Appendix D), predator effects through the consumptive channel were not influenced by predator diversity or the occurrence of antipredator behavior. In monoculture, two predator species were particularly strong disruptors of caterpillar feeding, the parasitoid *Diadegma* and the lady beetle *Hippodamia* (Fig. 4c), and these same two predator species were more likely to forage on plants when embedded within diverse communities (Fig. 4a). Together, these results suggest that predator diversity effects were conducted through a chain of behavioral interactions, with the most intimidating predators more likely to occur on plants, and thus to disrupt herbivore feeding, when within diverse communities.

The cascading diversity effects we observed appear to have been initiated by a reduction in the degree of interference among predators in the diverse communities. In our system, plant-visitation by predators served as an indicator of predator-predator interactions. Given that caterpillars (and aphids) remained on their host plant until confronted by a threat (Fig. 2b), predators were generally forced to converge on the plants in order to locate food. Our finding that plant-visitation was greater in the high diversity treatment (Appendix A6) suggests that interference was relaxed to some extent among predators in the multi-species communities. In our low diversity arenas, predators interacted only with conspecifics, whereas in diverse arenas predators interacted only with heterospecifics. Thus, intraspecific competition among predators was entirely eliminated within the diverse communities. To the extent that intraspecific competition may exceed interspecific competition for some species, relief from intraspecific
interactions may facilitate greater foraging efficiency. It was evident in our study that the predators *Diadegma* and *Hippodamia* were much more likely to forage for prey within cages housing heterospecific than conspecific competitors (Fig. 4a), suggesting that the release from intraspecific interference in diverse communities allowed these species to forage more effectively. There is good evidence in the literature of strong intraspecific interference among arthropod predators (Sih et al. 1998), including for many of the predators used in this study when foraging on *B. oleracea* plants (Northfield et al., in revision). Both lady beetles and parasitoids have the ability to recognize and respond to cues left by earlier-arriving conspecific competitors (Rosenheim 1998, Hodek and Michaud 2008), which is thought to be adaptive because of the high risk of cannibalism for progeny oviposited where conspecifics already occur. In our experiments predator mortality was rare overall, and we saw no evidence that intraguild predation exceeded mortality (if any) due to cannibalism (Fig. 4b). This strengthens our argument that predator-predator interactions were primarily behavioral in nature. At least one other study has provided theoretical evidence that relief from intraspecific competition can generate positive diversity effects (Weis et al. 2007); the mechanism in that case was the reduction of exploitative competition with increasing diversity, rather than the reduction of intraspecific interference.

Nonetheless, we were curious whether higher plant-visitation rates by *Diadegma* and *Hippodamia* could entirely explain the behaviorally-mediated diversity cascade that we observed. We can examine this quantitatively by calculating an “expected” number of displacements for each polyculture replicate, under the simplifying assumption that a plant visit by a predator incites the same level of intimidation regardless of diversity treatment. We first
calculate the number of caterpillar displacements per predator, per plant-visit for each species in monoculture (displacements could not be attributed to particular predator species in the polycultures). Then, multiplying this value by the observed plant visitation rate of each predator species in a given polyculture, and summing these values across all species in that polyculture, yields the expected number of caterpillar displacements for that replicate. Intriguingly, observed caterpillar displacement per-visit in polyculture was greater than predicted by this calculation ($t = 2.41$, df = 19, $P = 0.026$; Fig. 5). Thus, accounting for higher per-capita visitation rates by the two most intimidating predator species does not entirely explain the greater nonconsumptive effects of diverse predator assemblages, suggesting that another factor was also at work. One possibility is that predator consumptive and nonconsumptive effects scale differently with predator density. Behaviorally-mediated effects could remain relatively intense as the density of particular predator species declined, as happens at higher diversity levels within substitutive designs. This could occur, for example, because a predator can simultaneously intimidate many prey individuals but kill only one at a time (Peacor and Werner 2001). Consistent with this explanation, in our experiments nonconsumptive effects were similar to those of the single most intimidating predator species, whereas consumptive effects reflected the average predation rates across species (Fig. 3). A relative insensitivity of nonconsumptive effects to predator density would allow these effects to remain consistently intense in diverse communities, even when greater diversity dilutes densities of the more intimidating predator species. This points to the possibility that a highly intimidating species may effectively saturate a system with fear, rendering other predators variously redundant. If predators’ nonconsumptive effects were indeed partially redundant in both monoculture and polyculture, then our finding that $D_T$ values were
significantly greater than zero (in terms of nonconsumptive effects; Fig. 3) indicates that the more intimidating predator species were less redundant in polyculture than in monoculture. So, not only did *Diadegma* and *Hippodamia* spend more time on plants when embedded in diverse communities, but their nonconsumptive effects on caterpillars and plants were, to some extent, complementary.

More generally, dissimilarity in hunting domain and foraging mode decreases the probability of predator-predator encounters and thereby may reduce interference competition (Björkman and Liman 2005, Preisser et al. 2007, Schmitz 2007). Interference can be direct, manifesting as aggressive or despotic behavior among predators (Shaffer and Robinson 1996, Switalski 2003), or indirect, with intraguild prey assessing their surroundings for evidence of intraguild predators and actively avoiding encounters (Soluk and Collins 1988, Siddon and Witman 2004, Vance-Chalcraft and Soluk 2005, Berger et al. 2008). Given that conspecific predators would be expected to be more similar than heterospecifics, intraspecific competition should be greater than interspecific competition (MacArthur 1958). Hence, if foraging in a more diverse predator community can spare an actively hunting species from frequent encounters with conspecifics (e.g., Björkman and Liman 2005), then such predators should be more efficient in polyculture. Our results are consistent with this expectation, yet our data also represent a departure from studies isolating the cascading effects of diversity. Predator diversity effects have been shown to cascade via lethal predator-prey interactions (Bruno and Cardinale 2008), as well as by lethal predator-predator interactions (Finke and Denno 2004). Our data suggest that cascading diversity effects can also be initiated by nonconsumptive interactions among predators, which dovetails
well with recent work investigating how the effects of complementary predator-prey relationships may cascade via nonconsumptive channels (Byrnes et al. 2006).

Our experimental arenas included green peach aphids as a second, non-focal herbivore species because caterpillars nearly always co-occur with aphids on *B. oleracea* plants at our study site and are common prey of the predators (Snyder et al. 2006, Straub and Snyder 2006, 2008). Although the magnitude of the difference is relatively small (Appendix E), predators significantly reduced aphid densities in *Fear+* cages because these were the only cages that included predators. These lower aphid densities may have reinforced the strength of the significant trophic cascade that we observed in these cages (see nonconsumptive main effect for plant biomass, Appendix A1). However, in contrast to the plant biomass (Appendix A1) and caterpillar displacement findings (Appendix A2), the aphid data showed no evidence of an interaction between predator diversity and nonconsumptive effects (Appendix A5), suggesting that the significant diversity effects we observed were unlikely to have been mediated by aphid densities. For aphids, a significant interaction between Diversity and Block appeared to result from aphid densities being slightly lower in diverse predator communities in *Block 1*, but slightly higher in diverse predator communities in *Block 2* (Appendix A5). So, there was no consistent predator diversity main effect influencing aphids, and there were no significant interactions between Diversity and any other factor. Since aphid densities were similar everywhere predators foraged, any impact of aphid density on predator foraging behavior would be similar across all treatments. Thus, it does not appear that the direct or indirect effects of aphids could explain the strong interaction between predator diversity and nonconsumptive effects that impacted our trophic cascades (Fig. 2).
We used removals and replacements of herbivores to isolate predator trophic versus nontrophic effects. This precluded the need to render predators nonlethal and thereby avoided many of the potential problems resulting from such manipulations (e.g., altered predator behavior, absence of substrate-borne cues associated with prey capture). However, there are limitations to what can be achieved using our techniques. When removing prey from no-predator cages to isolate lethal (consumptive) predator effects, there is always a short lag between when predation events occur and when the prey are removed to mimic predation (Griffin and Thaler 2006). As a result, prey culling techniques slightly underestimate the true strength of consumptive effects, as prey that should be “dead” continue feeding until the daily predation simulation can be levied. Similarly, when adding prey to isolate nonconsumptive effects, there is a lag between the moment a prey individual is killed and when it is replaced, leading to an overestimation of the strength of nonconsumptive predator effects. These biases heighten the importance of frequently censusing herbivores such that lag times are minimized (< 1 day), and suggest caution in placing too much weight in estimates of the relative strengths of consumptive versus nonconsumptive effects. Despite these limitations, our methodology can be particularly useful when looking for interactions between these two effects and other factors (e.g., predator diversity), where it is less important to precisely reproduce the magnitudes of consumptive/nonconsumptive effects, but more important to isolate them from each other.

To our knowledge, no previous studies have isolated nonconsumptive from consumptive effects while also manipulating consumer diversity. Theoretical and empirical evidence suggest that consumer species richness may commonly influence the nature and magnitude of nontrophic interactions between predators and prey, as well as among predators (Sih et al. 1998, Borer et al.
2002, Byrnes et al. 2006, Schmitz 2007, Goudard and Loreau 2008). It is striking how often behavioral interactions underlie emergent diversity effects for predators and other animal consumers. For example, inherent differences in foraging behavior and/or prey choice can lead to a positive relationship between predator diversity and herbivore exploitation (Preisser et al. 2007, Finke and Snyder 2008). Similarly, prey species may exhibit predator-specific antipredator behavior, modulated to reflect the differing risks posed by different predator species (Shaffer and Robinson 1996, Byrnes et al. 2006, Castellanos and Barbosa 2006). This suggests that behavioral interactions may mediate diversity effects to the same degree that consumptive interactions may, as has been found when looking at simpler subsets of consumer communities (Preisser et al. 2005).

Our results bring us to several conclusions. First, behaviorally-mediated diversity affects can cascade across multiple trophic levels, as do density-mediated effects (Ives et al. 2005, Duffy et al. 2007). This reinforces suggestions that nonconsumptive interactions warrant greater attention in ecological research (Peckarsky et al. 1991, Brown et al. 1999, Borer et al. 2002), and that it may be unwise to rely entirely on abundance or biomass metrics when quantifying diversity effects (Byrnes et al. 2006, Goudard and Loreau 2008). Second, consumer effects operating via nonconsumptive channels might act quite differently than, and in our study were independent from, those due to resource consumption. This raises the possibility that diversity effects attributed to consumptive interactions may instead be partially, or entirely, due to nonconsumptive interactions. It appears that many cases of consumptive diversity effects in animal communities have complex behavioral underpinnings (Bruno and Cardinale 2008), and
only a more consistent consideration of how behavior influences diversity effects will allow incorporation of these effects into theory (Borer et al. 2002, Goudard and Loreau 2008).

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REFERENCES


FIGURE LEGENDS

**Fig. 1** Factorial structure of the experiment: predator diversity (Low, High) × consumptive effects (Lethal+, Lethal−) × nonconsumptive effects (Fear+, Fear−) × temporal block (1, 2). All factors were fully-crossed, achieving a complete $2 \times 2 \times 2 \times 2$ factorial design.

**Fig. 2** (a) Final plant biomass and (b) Total number of caterpillars observed off of plants, for three factors: predator diversity (Low, High), predator nonconsumptive effects (Fear+, Fear−), and predator consumptive effects (Lethal+, Lethal−). Data are shown averaged across the two temporal blocks; data are presented separately by block in Appendix B. Data are means ± 1SE.

**Fig. 3** Two measures of overyielding, $D_T$ and $D_{max}$, for consumptive and nonconsumptive predator effects on (a) plant biomass and (b) herbivore density (Lethal channel) and behavior (Fear channel). See Appendix C (Ecological Archives) for calculations of $D_T$ and $D_{max}$ values (data are means ± 95% CI).

**Fig. 4** Occurrences, per-capita, of each predator species (a) observed foraging on plants, and (b) dying during the course of the experiment in high and low diversity compositions. Total number of caterpillars (c) displaced and (d) killed by each predator species when in monoculture. Data are for the predator species *Hippodamia* (Hip), *Diadegma* (Dia), *Chrysopa* (Chr), *Nabis* (Nab), and *Geocoris* (Geo). Data are means ± 1 SE.
Fig. 5 Predicted and observed caterpillar displacements in high diversity species compositions.

Predicted displacement for a given polyculture was calculated by summing the predicted displacements generated by each species constituting the polyculture: \[ \sum \left( \frac{\text{observed displacement}_i}{\text{observed visits}_i} \right) \] for the \( i^{th} \) species in monoculture, and the \( i^{th} \) species within the \( j^{th} \) polyculture. Data are means ± 1 SE.
Figure 1

|                | |                 |
|----------------||-----------------|
| **Consumptive**| \(\times\) | **Predator Diversity** (High, Low) \(\times\) **Temporal Block** (1, 2) |
| **Non-consumptive** | \(+\) | \(-\) |
| **Control**     | | | **Lethality** |
| **Fear**        | \(+\) | **Lethality** + **Fear** |
Figure 2

(a) Lethal- Plant biomass (g)

(b) Nonconsumptive component (-/+)

Consumptive component (-/+)

Fear - Fear +

Fear - Fear +

No. caterpillar displacements

Plant biomass (g)

0.2 0.4 0.6

High diversity

Low diversity
Figure 3

(a) Indirect effects

(b) Direct effects

Effect channel

Metric value

Lethal  Fear

0.4
0.2
0.0
-0.2
-0.4

-1
0
1
2
3

DT  Dmax
Figure 4

(a) Per-capita visits to plant

(b) Per-capita predator mortality

(c) No. caterpillar displacements

(d) No. caterpillars killed

Legend:
- Low diversity
- High diversity

Predator species:
- Hip
- Dia
- Chr
- Nab
- Geo

Different letters indicate significant differences among treatments.
Figure 5

Predicted vs. Observed Caterpillar Displacements per Composition (mean ± 1 SE)