

Criteria determining behavioural responses to multiple predators by a stream mayfly

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We investigated prey responses to multiple predators by observing drift and positioning of *Baetis bicaudatus* (Ephemeroptera: Baetidae) nymphs in the presence of non-feeding brook trout (*Salvelinus fontinalis* chemical cues) and predatory stoneflies (*Megarcys signata* with glued mouths). In experiments carried out in stream-side channels *Baetis* antipredator behaviour differed markedly in response to each predator separately; they drifted away from stoneflies, and reduced exposure and drift levels in the presence of trout odour. When both predator cues were presented together the effect of stoneflies, which were observed more frequently at night, depended on the time of day and was modified by presence of fish odour. During the day *Baetis* responses to both predators were hierarchical as drift and the number on stone tops was reduced whenever fish odour was present regardless of the presence of stoneflies. At night when stoneflies were foraging, *Baetis* behaviour reflected a compromise between avoiding both predators, with responses to both predators being intermediate between that characteristic of stonefly avoidance (entering the drift) and that characteristic of fish avoidance (drift avoidance). The switch in antipredator behaviour from 'hierarchy' to 'compromise' between multiple predators reflects changes in the relative risk of predation during the day and night. During the day visually feeding trout present a higher predation risk, but at night when stoneflies are foraging and trout visual systems are less effective, the predation risks become more similar. We found no evidence for 'accumulative' avoidance where the magnitude of avoidance increases in response to the number of predators present, because avoiding stoneflies conflicted with avoiding trout. We predict that predator avoidance will take place in the form of a 'hierarchy' when one predator presents substantially higher predation risk than another; 'compromise' behaviour will be observed when the predation risk presented by two predators is more equivalent, but avoiding one predator conflicts with avoiding another; and 'accumulative' antipredatory responses will be observed only when responses to different predators do not conflict. We emphasise that investigation of the responses of prey to multiple predators is important to our understanding of natural communities, and is likely to reveal details about the mechanisms determining the outcome of interactions involving multiple predators.

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In natural situations animals are usually susceptible to predation by more than one type of predator (Cohen and Briand 1984, Pimm et al. 1991, Begon et al. 1996). The presence of multiple predators may significantly alter a prey animal's risk of predation. Furthermore,

avoiding one predator could conflict with avoiding another. Despite the potential importance of such dilemmas most empirical investigations of antipredator behaviour have focused on situations involving only one predator (see reviews by Sih 1987, Lima and Dill

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1990). Studies that have considered different predators foraging together have examined their impact on prey fitness (Peckarsky and McIntosh 1998), prey consumption by the predators (e.g. Soluk and Collins 1988a, Wissinger and McGrady 1993, Wissinger et al. 1996, Dahl and Greenberg 1997), prey foraging decisions (e.g. Soluk and Collins 1988b, Kotler et al. 1993, Scrimgeour and Culp 1994) or have modelled predator and prey populations (e.g. Matsuda et al. 1993). However, we need to investigate the criteria that prey use to determine the appropriate behavioural responses to multiple predator species. Those criteria are important because they not only determine prey behaviour, and ultimately individual prey fitness, but also affect the outcome of interspecific interactions at the community level (Peckarsky and McIntosh 1998).

A range of options are available to prey when faced with predation by multiple predators. First, prey could use a hierarchy of predation risk to determine their behaviour whereby they avoid predators in the order of the risk they pose. In this scenario, termed 'hierarchy control', a prey individual would forsake avoiding one predator that presents a lower risk to avoid a more dangerous predator. Hierarchy control should be common when the more dangerous predator modifies the risk presented by another predator leading to large disparities in the predation risk presented by the two predators. Alternatively, a vulnerable individual might adjust its behaviour by responding to the total or cumulative amount of predation risk it perceives from all predators (see Table 14.2 in Sih 1987). We might expect this scenario to be more common in situations where predation risk to multiple predators is more equivalent. On the other hand, we would not expect to observe 'accumulative control' of antipredator behaviour if the avoidance of one predator conflicts with avoidance of another. In situations where avoiding one predator results in increased vulnerability to another predator, prey might make a compromise involving a trade-off between avoiding one predator and avoiding another. Individuals could use 'compromise control' of antipredator behaviour that minimises their overall risk of predation.

In this study we investigated the antipredator behaviour used by a stream-dwelling benthic mayfly to avoid predatory fish, which feed from the water column, and to avoid predatory stoneflies, which occupy the substratum interstices of streams. Our aim was to identify the patterns of decision making that the mayfly used to determine their antipredator behaviour to the two predators. We designed experiments to distinguish whether the antipredator behaviour of the mayfly reflected 'hierarchy', 'compromise' or 'accumulative' control.

Study animals

The nymphs of the mayfly *Baetis bicaudatus* (Baetidae) are abundant and ubiquitous in the East River and its tributaries around the Rocky Mountain Biological Laboratory (RMBL) at 2459 m in Gunnison County of Colorado (Peckarsky 1991). At this elevation *B. bicaudatus* is bivoltine, with a fast-growing summer generation and a slower-growing generation that overwinters and emerges in early summer (Peckarsky et al. 1993). *Baetis* is the predominant mayfly prey of a predatory perlotid stonefly, *Megarcys signata* (Peckarsky and Penton 1989a). Encounters with *Megarcys* frequently result in *Baetis* abandoning foraging on the tops of rocks, where its diatom food is most abundant, and fleeing into the water column to escape predation (Peckarsky and Penton 1989b, Peckarsky 1996). Those disruptions can lead to sublethal reductions in *Baetis* fitness (Peckarsky et al. 1993, Peckarsky and McIntosh 1998).

Baetis is also vulnerable to predation by several species of trout. In the East River catchment around the RMBL brook trout, *Salvelinus fontinalis*, are the most common, but rainbow (*Oncorhynchus mykiss*), cutthroat (*O. clarki*) and brown trout (*Salmo trutta*) are also present (A. R. McIntosh, B. W. Taylor and S. Gross unpubl.). Visually feeding fish like trout present a higher predation risk to mayflies during the day than the night (Jenkins 1969, Ware 1973, Walsh et al. 1988, McIntosh and Townsend 1995, authors' unpubl. data). This heightened risk during the day typically reduces mayfly movement in the water column of streams (drift) and can also affect their positioning on the substratum surfaces (Flecker 1992, Culp and Scrimgeour 1993, Cowan and Peckarsky 1994, Douglas et al. 1994, McIntosh and Townsend 1994, Tikkanen et al. 1994). In the East River almost all *Baetis* drift occurs at night (Kerans et al. 1995) and nymphs typically appear on the tops of rocks primarily at night (Peckarsky and Cowan 1995, McIntosh and Peckarsky 1996, Peckarsky and McIntosh 1998). In streams without trout, mayfly behaviour is more variable; both aperiodic and nocturnal drift and positioning periodicities have been recorded (Cowan and Peckarsky 1994, McIntosh and Peckarsky 1996).

Methods

To determine how *B. bicaudatus* co-ordinate their antipredator behaviour against stoneflies and brook trout we observed *Baetis* behaviour using a factorial experiment manipulating the non-lethal presence and absence of both brook trout and stoneflies. The experiment was conducted in replicate artificial stream channels in the boreal summer of 1996. The speed of prey responses to predator cues was also measured to determine how

temporal variation in the presence/absence of the predator might affect antipredatory behaviour.

The artificial streams were constructed from grey plastic cattle watering tanks (width \times length: 68 \times 121 cm) that provided a wetted surface area of 0.823 m². We added a Plexiglas baffle to the centre of the tanks with a water jet on either side of the baffle directed to create circular flow (Fig. 1 in McIntosh and Peckarsky 1996). Water drained via a central standpipe with 0.5 mm-mesh windows thereby creating a flow-through system and a mean \pm SE water depth of 20.3 \pm 0.1 cm. The tanks received water filtered of large macroinvertebrates and gravity fed from a small fishless tributary of a fishless beaver pond. The fishless status of the water sources was checked regularly with visual observations and once by electrofishing. The intake filter and the standpipe outlets were cleaned daily to prevent clogging. During the experiments water velocity (measured at six locations around the tanks) was similar among tanks (mean \pm SE = 35 \pm 1 cm s⁻¹), but heterogeneous within tanks (range = 12–54 cm s⁻¹), depending on the proximity to the water jets. Water temperature ranged from 4°C to 11°C depending on the time of day. The bottom of each tank was covered with washed gravel that we collected from dry fishless stream banks and then cobbles were arranged in six clusters on top of the gravel. The experimental tanks were arranged in blocks of four tanks and the flow was regulated to each block by a gang valve system.

To prevent prey consumption from confounding our behavioural observations we used fish odour to simulate the presence of brook trout and glued the stonefly's mouthparts to prevent them from feeding (Peckarsky et al. 1993). Previous experiments in our system (Cowan and Peckarsky 1994, McIntosh and Peckarsky 1996, Peckarsky and McIntosh 1998) and in other streams (Tikkanen et al. 1996) indicate that *Baetis* mayflies from areas where trout are native alter their behaviour in the presence of chemical cues from trout. Nocturnal avoidance behaviour of *Baetis* from the East River is triggered in nymphs from fishless streams by the presence of brook trout odour, and the magnitude of nocturnal movement in nymphs from trout streams is reduced by brook trout odour (McIntosh and Peckarsky 1996, Peckarsky and McIntosh 1998). Once induced, nocturnal behaviour is maintained (for more than three weeks) even in the complete absence of fish cues (Cowan and Peckarsky 1994, McIntosh and Peckarsky 1996, Peckarsky and McIntosh 1998). Thus, to provide the best contrast among treatments we used *Baetis* from a fishless stream, Benthette Brook, in these experiments. Observations since 1979 and recent electrofishing indicate that this tributary of the East River has been consistently fishless.

We used 16 tanks arranged in four blocks. The two factors were randomly allocated to tanks in a complete randomised block design creating four replicates of four

different predation regimes: (1) trout odour absent and stoneflies absent (control), (2) trout odour absent and stoneflies present, (3) trout odour present and stoneflies absent, and (4) trout odour present and stoneflies present. The foraging behaviour of *Megarcys* with mandibles glued with Barge cement is indistinguishable from those that do not have glued mouthparts (Peckarsky and Cowan 1995, Peckarsky 1996). We controlled for the presence of the Barge cement by placing pebbles containing Barge cement in tanks without stoneflies.

After allowing several days for algal colonisation of substrata, from 25–26 June we added 300 winter generation *Baetis* from Benthette Brook to each tank. We used only late instar larvae (stage III) without black wingpads (i.e. not about to emerge from the stream) and with a mean (\pm SE) headcapsule width of 0.85 (\pm 0.01) mm. Densities simulated mean densities of this stage of *Baetis* in the East River (365 m⁻², B. L. Peckarsky unpubl.).

On 27 June we added eight female *Megarcys* from Benthette Brook with a mean (\pm SE) head capsule width of 4.37 (\pm 0.03) mm to tanks designated to receive stoneflies. This density was within the range found in streams of the East River drainage (Peckarsky 1991). We also added two brook trout (between 140 mm and 180 mm fork length) to a large (110 L) holding tank that received the same fishless water as the experimental tanks. Water from the fish holding tank was delivered to the experimental tanks through spouts (mean \pm SE rate = 1.68 \pm 0.01 L min⁻¹). The tanks designated not to receive fish odour received fishless water delivered at a similar rate through identical spouts.

We observed the behaviour of both mayfly and stonefly nymphs in the tanks during the daylight at 10.00 h and during darkness at 22.00 h (mountain daylight saving time) for 6 d. Four people made observations and each person recorded behaviour in one block of tanks. During the day observers knelt motionless beside the tanks to make observations. Past experience (e.g. Cowan and Peckarsky 1994, Peckarsky and Cowan 1995, McIntosh and Peckarsky 1996) indicates that as long as observers do not make sudden movements (e.g. while moving between tanks) behaviour of *Baetis* and *Megarcys* is not affected by the presence of observers. At night observations were made using dim red light from headlamps covered with red filters. Red light can alter the behaviour of some mayflies (Heise 1992, A. R. McIntosh pers. obs.), but we and other authors have found that *Baetis* mayflies behave normally when observed with red light (Allan et al. 1986, Casey 1987, Cowan and Peckarsky 1994, McIntosh and Peckarsky 1996). The number of stoneflies visible and moving about the tanks on all surfaces, the number of mayfly nymphs visible in the tanks, and the number of mayflies drifting past a transect during a 2-min period were recorded from each tank.

To assess how quickly the mayflies responded to the presence of the predators we monitored the behaviour of mayflies in the tanks just before and after the predators were added. Observations were made at 75 min before the predators were added and at 5, 20, and 60 min after the predators were added. To complete observations on all tanks took approximately 15 min, so these measurements were made between 75 and 60 min before the predator additions and between 5 and 20 min, 20 and 35 min, and 60 and 75 min after the predator additions.

We tested for differences in behavioural variables between the predator treatments using a 2×2 factorial analysis of variance on the means of the 6 d of observations. Since the same populations were sampled during the day and night, "time of day" was added as an additional repeated measure to test for differences between the diurnal and nocturnal behaviour. Since our two response variables were interdependent and possibly correlated we first conducted a multivariate analysis of variance. Then, based on the outcome of the MANOVA, we tested both response variables with univariate ANOVA. When assessing the speed of behavioural changes, we were interested in mayfly behaviour at each particular time and not a general time effect that might be detected by a repeated measures design. Therefore, we tested for treatment effects using a separate two by two factorial ANOVA for each time. Accordingly we applied a Bonferroni correction to control for type I statistical errors and assessed statistical significance with $\alpha = 0.012$. We transformed variables where necessary (described in table legends) so that data satisfied the assumptions of analysis of variance. The term representing variation among blocks of tanks was included in ANOVA models when significant, but was otherwise excluded from ANOVA models to improve statistical power.

Results

The number of stoneflies observed on the substratum surface increased significantly at night (Fig. 1, Table 1). The presence of brook trout odour significantly reduced the number of stoneflies moving on the substrate surface on all occasions (Fig. 1, Table 1).

MANOVA indicated that all three independent factors had a significant effect on our behavioural response variables (Table 2a). *Baetis* drift during the day was nearly zero when brook trout odour was present, while mayfly nymphs in tanks without fish odour drifted similar amounts during the day and night (Fig. 2). Thus, there was a significant fish effect and a time by fish interaction since fish odour reduced drift only during the day (Table 2b, Fig. 2). The impact of stoneflies depended on both the time of day and the

presence of fish odour as indicated by a significant three-way interaction (Table 2b). This three-way interaction occurred because during the day fish odour reduced the number of *Baetis* drifting regardless of stonefly presence or absence, but at night the magnitude of the reduction of *Baetis* drift in the presence of fish odour depended on the presence of stoneflies (Fig. 2). Mayfly drift was lowest with fish odour and no stoneflies, highest with stoneflies and no fish odour, and intermediate (not different from controls) with both predators present.

There were dramatic nocturnal increases in the number of *Baetis* occupying positions on the substratum surface in all treatments (Fig. 3) as indicated by significant time effects (Table 2c). Similar to the drift responses, there were significant interactions between the presence of fish odour and time of day (Table 2c), because the effect of fish odour on the number of *Baetis* visible on the substratum differed between night and day (Fig. 3). The presence of stoneflies increased the number of *Baetis* visible, as indicated by a significant stonefly effect (Table 2c). However, there was also a three-way interaction between fish odour, time and stoneflies (Table 2c) because the effect of fish odour and stoneflies differed between night and day. Stoneflies had no effect on the number of *Baetis* visible during the day when fish odour was present but stoneflies did increase the number visible at night even when fish odour was present (Fig. 3).

In summary, trout-avoidance behaviour (decreased drift and exposure) took precedence during the day

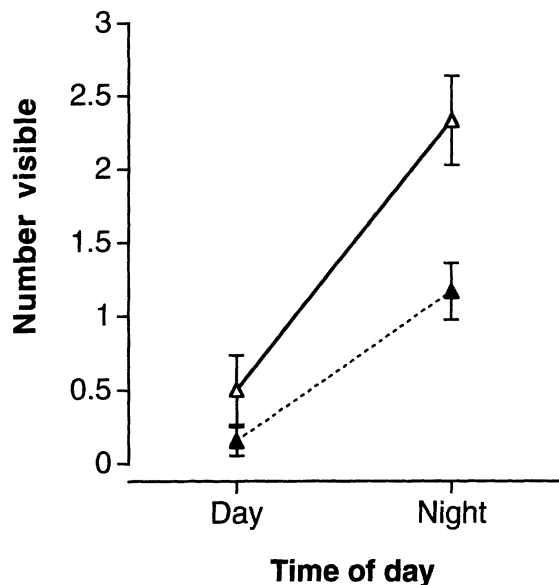


Fig. 1. The mean number of *M. signata* stoneflies observed on the substratum surface in the experimental channels during the day and night in tanks with (black triangles) and without (white triangles) brook trout odour. Error bars indicate one standard error.

Table 1. Factorial ANOVA using repeated measures on the number of predatory stoneflies observed on the substrate surface in experiments with and without brook trout odour (Fish) during the day and night (Time). Data were transformed $\sqrt{x+0.05}$.

	Source	df	MS	F	P
Between tanks	Fish	1	0.3283	7.851	0.031
	Error	6	0.0418		
Within tanks	Time	1	1.390	80.650	<0.001
	Time \times Fish	1	0.0440	2.555	0.161
	Error	6	0.0172		

when stoneflies were inactive, but at night the behavioural response of *Baetis* to both predators (black squares in Figs 2 and 3) was intermediate between that characteristic of stonefly avoidance (●) and that characteristic of trout avoidance (□).

Daytime observations at the start of the experiment indicated that the onset of antipredator behaviour was rapid (Fig. 4). Observations completed before the predator additions indicated there were some variations between tanks, but no significant treatment effects. Five minutes after predators were added fish odour caused a significant decrease in mayfly drift and the number of mayflies visible, whereas presence of stoneflies significantly increased mayfly drift (Tables 3 and 4). After 20 min we observed patterns of behaviour associated with fish odour that were typical of those observed consistently on following days of the experiment. That is, trout odour significantly reduced the number of *Baetis* visible and drifting. Stoneflies had no significant effect, except drift increased in the 5-min sample. Thus, both trout odour and stoneflies had an almost immediate effect, but stoneflies did not consistently alter prey behaviour until sometime greater than one hour after they were introduced.

Discussion

Most empirical studies of prey antipredator strategies concern responses to single predators and the subsequent trade-offs involving resource acquisition (e.g. Fraser and Huntingford 1986, Lima and Dill 1990, Werner and Anholt 1993). Meanwhile, investigations in community ecology are revealing that complex indirect interactions are common, and that a knowledge of pairs of species interactions is seldom sufficient to explain the patterns observed (Billick and Case 1994, Wootton 1994a, b). Thus, developing models that predict how prey respond to multiple predators in complex food webs presents an important challenge for behavioural ecologists. Even in the relatively simple system we have examined prey react quite differently depending on the combination of predators present and the time of day. Nevertheless, those patterns of behaviour are predictable based on the characteristics of the predators and the predation risks they present.

Responses to one predator species

The responses we observed to single predators reflect two well-established patterns of prey behaviour. First, mayflies often hide and rarely move in the water column during the day when the risk of predation by visually feeding fish is high (Flecker 1992, Culp and Scrimgeour 1993, Douglas et al. 1994, McIntosh and Townsend 1994, Tikkanen et al. 1994). Second, *Baetis* mayflies flee into the water column to escape from stoneflies (Peckarsky and Penton 1989b, Peckarsky 1996). Those patterns of behaviour are typical of most prey associated with visually feeding fish and benthic feeding invertebrates (see review by Wooster and Sih 1995). However, our experiments with two predators revealed several new details about those single pairs of predator-prey interactions.

First, stoneflies induce increased *Baetis* drift during the day even though stoneflies do not forage during the day (Martinez 1987, Peckarsky and Cowan 1995, L. A. Martinez and B. L. Peckarsky unpubl.). The increased daytime drift of *Baetis* in fishless environments with high densities of stoneflies during the day is probably associated with foraging triggered by hunger as an aftermath of intense predator-avoidance of stoneflies during the previous night. Previous experiments indicate that increased *Baetis* drift resulting from encounters with stoneflies, as we observed at night, leads to reductions in *Baetis* gut fullness (Peckarsky et al. 1993). Thus, in fishless streams the increased foraging of stoneflies at night probably forces *Baetis* to forage more during the day. This mechanism would also explain why mayflies taken from fishless streams that have never experienced fish odour, hide during the day in treatments with no fish odour and no stoneflies (Fig. 3 and McIntosh and Peckarsky 1996). We suspect that they do not take the risk of daytime foraging unless they are very hungry (Peckarsky 1996), and that their hunger is a function of avoidance of stoneflies on the previous night. Interestingly, in the short-term observations (Fig. 4) we observed a response of *Baetis* only in the first 5 min after addition of stoneflies, and not in subsequent observations up to 75 min after stoneflies were placed in the tanks. We suspect that this initial mayfly response reflected unnatural daytime movement of stoneflies that were searching for refuges rather than prey, but caused drift of *Baetis* due to elevated preda-

tor-prey encounter rates. This is a different mechanism than the one we believe causes the longer term elevation in daytime drift of mayflies when stoneflies are present in their environment (Figs 2 and 3).

Second, the speed of the response to fish odour was fast, less than 12 min. It may even be faster but the logistics of counting mayflies in all 16 tanks prevented finer resolution. Since we found that nocturnal behaviour was induced in fishless stream mayflies, we agree with Tikkanen et al. (1996) that diel drift periodicities are not necessarily fixed behavioural patterns. However, unlike the study of Tikkanen et al. (1994), where visual or hydrodynamic cues in addition to chemical cues were needed to elicit a strong nocturnal periodicity, fish odour alone was sufficient to induce nocturnal avoidance responses in our experiments.

Third, our experiments with both predators demonstrated that the presence of fish odour decreased stonefly foraging. This observation is consistent with another study indicating reduced fitness in stoneflies that coexist with trout (Feltmate and Williams 1991), but does not agree with the suggestion that low stonefly densities in trout streams result from behavioural avoidance (dispersal) of predators (Feltmate and

Williams 1991). It does, however, provide a behavioural mechanism to explain the reduced response of *Baetis* to stoneflies when both predators are present. Thus, the hierarchical avoidance of trout over stoneflies during the day may be driven by the effect of trout on stonefly foraging behaviour.

Responses to two predator species

The patterns of prey behaviour associated with the two predators separately indicate that avoidance responses to trout and stoneflies conflict, since a mayfly entering the drift to avoid a stonefly is exposed to high risk of trout predation primarily during the day. Thus, the costs and benefits of each predator-avoidance response differed between day and night. During the day the presence or absence of stoneflies had no impact on mayfly behaviour, as long as fish odour was present in the environment. We term this pattern "hierarchy control" in which prey respond entirely to the most dangerous of two predators. We suggest that *Baetis* nymphs use this criterion because the predation risk presented by trout exceeds that presented by stoneflies during the day that they do not risk trout predation to avoid a stonefly. In foraging experiments brook trout

Table 2. Factorial MANOVA (a) and ANOVA using repeated measures on (b) the number of *Baetis* nymphs drifting in the water column and (c) the number of nymphs visible on the substrate surface in experiments with different predators (\pm stoneflies and \pm brook trout odour) during the day and night (Time). Data were square root transformed.

	Source	df	MS	F	P	
(a) MANOVA						
Between tanks	Fish	2,11	0.198*	22.27	<0.001	
	Stonefly	2,11	0.530*	4.877	0.03	
	Fish \times Stonefly	2,11	0.746*	1.877	0.20	
Within tanks	Time	2,11	0.014*	375.9	<0.001	
	Time \times Fish	2,11	0.140*	33.77	<0.001	
	Time \times Stonefly	2,11	0.885*	0.7127	0.51	
	Time \times Fish \times Stonefly	2,11	0.537*	4.741	0.03	
	Error					
(b) drift						
Between tanks	Fish	1	32.21	44.81	<0.001	
	Stonefly	1	4.647	4.466	0.026	
	Fish \times Stonefly	1	1.466	2.040	0.179	
	Error	12	0.7187			
Within tanks	Time	1	8.844	51.46	<0.001	
	Time \times Fish	1	10.46	60.85	<0.001	
	Time \times Stonefly	1	0.1032	0.6006	0.453	
	Time \times Fish \times Stonefly	1	1.041	6.059	0.030	
	Error	12	0.1719			
(c) number visible						
Between tanks	Fish	1	8.318	27.50	<0.001	
	Stonefly	1	5.506	18.21	0.002	
	Block	3	1.163	3.845	0.050	
	Fish \times Stonefly	1	0.025	0.082	0.781	
	Error	9	0.3024			
Within tanks	Time	1	141.2	1160	<0.001	
	Time \times Fish	1	1.701	13.93	0.012	
	Time \times Stonefly	1	0.1609	1.318	0.280	
	Time \times Block	3	0.4030	3.302	0.071	
	Time \times Fish \times Stonefly	1	0.6766	5.544	0.043	
	Error	9	0.1220			

* Wilk's lambda.

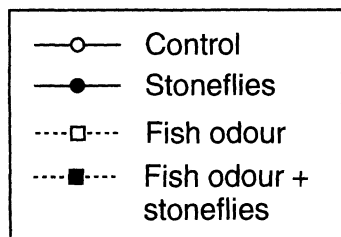
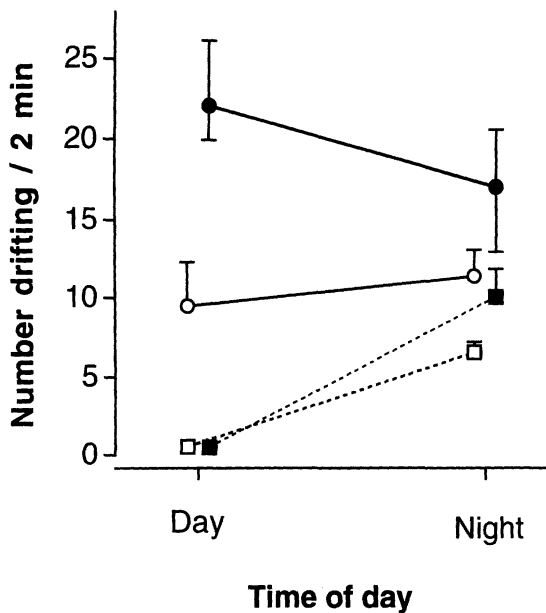


Fig. 2. The mean number of *B. bicaudatus* mayflies drifting in the water column in the experimental channels during the day and night in tanks with different predators. Error bars indicate one standard error.

captured over 40% of *Baetis* released into the drift during the day (authors' unpubl. data), resulting in a very high potential cost to using the wrong predator-avoidance behaviour. In contrast, stonefly foraging is low during the day and most predation takes place during the night (Martinez 1987, Martinez and Peckarsky unpubl.), which further reduces the benefits of moving away from predatory stoneflies during the day.

Similarly, hierarchical control of antipredator behaviour may be favoured when multiple predators are present and post-contact defences are inadequate protection against one of the predators. In our study, *Baetis* is relatively safe from stonefly attacks during the day, and at night, unless closer than 2 cm to the stonefly (Peckarsky et al. 1994). Furthermore, mayflies have a low probability of being consumed even within this close distance from stoneflies (Peckarsky and Penton 1989a). Thus, post-contact defences (swimming and drifting) are used predominately to avoid active

stoneflies. In contrast, the precise location of a fish in the water column is difficult to predict, trout have reaction distances up to 90 cm (McIntosh and Townsend 1995), and during the day their probability of capturing mayfly prey once detected is very high (> 0.9 – McIntosh and Townsend 1995, authors' unpubl. data). In this situation post-contact defences are of little use. Thus, *Baetis* may use a hierarchical response during the day when both predators are present because post-contact defences towards trout are inadequate. This situation is essentially analogous to one predator presenting a substantially higher predation risk.

In contrast, at night when stoneflies were more active, *Baetis* displayed behaviour characteristic of a compromise between avoidance of both predators. We contend that this response reflects the reduction in the disparity in vulnerability between the two predators at night. The foraging and therefore the predation risk posed by stoneflies increases dramatically at night (Peckarsky and Cowan 1995); and the effectiveness of a visual predator like trout is dramatically reduced at

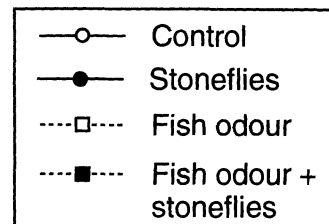
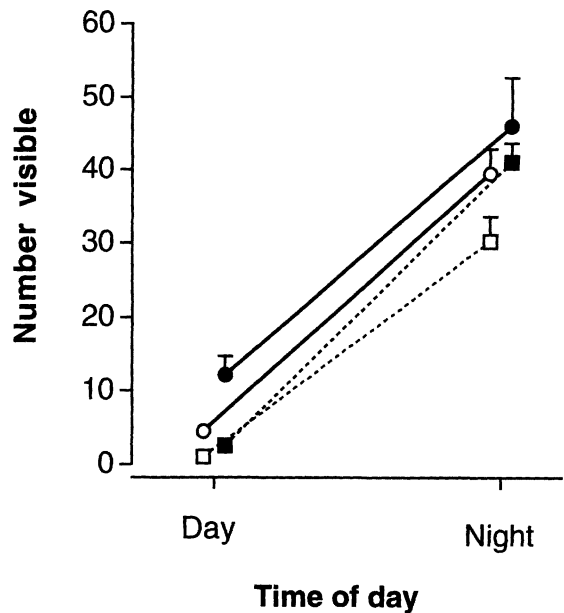
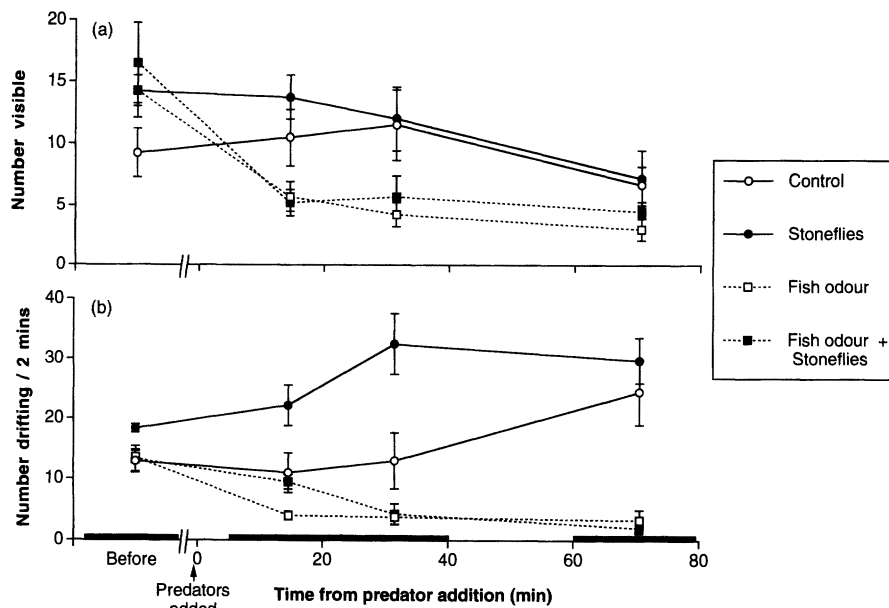


Fig. 3. The mean number of *B. bicaudatus* mayflies observed on the substratum surface in the experimental channels during the day and night in tanks with different predators. Error bars indicate one standard error.

Fig. 4. The mean number of *B. bicaudatus* mayflies (a) visible on the substratum and (b) drifting before and after the addition of the predators. Error bars indicate one standard error and the black bars on the time axis indicate the time taken to measure behaviour in all 16 tanks.



night (Jenkins 1969, McIntosh and Townsend 1995, authors' unpubl. data). Brook trout capture less than 10% of large *Baetis* nymphs released into the drift at night, and the reaction distance of trout to *Baetis* at night also decreases to < 7 cm. Thus, at night the area within which both predators detect prey is more similar, and the actual predation risks presented by trout and stoneflies converge.

Compromise antipredator behaviour has been shown by gerbils, *Gerbillus pyramidum*, which increase their use of open microhabitats when predatory snakes are present even though this makes them more vulnerable to predatory owls, which hunt over open ground (Kotler et al. 1992). Interestingly, when lights were added to open microhabitats to increase gerbil vulnerability to owls in this experiment, gerbil use of open microhabitat decreased to a level more similar to a situation with owls and lights only. Those observations are consistent with our prediction that when one predator presents much higher predation risk, the prey should switch from compromise to hierarchy antipredator behaviour to avoid the more dangerous predator.

In other systems prey have shown compromise avoidance behaviour even if the predation risk presented by one predator is dramatically higher than that presented by another if the compromise is relatively safe. For example, johnny darters (*Etheostoma nigrum*) avoid small-mouth bass (*Micropterus dolomieu*) by decreasing movement and hiding more, but they compromise and increase their movement when both crayfish (*Orconectes rusticus*) and bass are present to avoid the crayfish, which evict them from refuges (Rahel and

Stein 1988). This compromise is possible even though bass present a much higher risk because it does not significantly increase their vulnerability to either predator (Rahel and Stein 1988).

We did not observe any accumulative response to both predators where the amount of avoidance observed increased with the number of predators present (see Table 14.2 in Sih 1987 for examples). This is probably to be expected since avoiding stoneflies by entering the water column would make them more vulnerable to trout predation. Thus, increasing the amount of avoidance to either predator would be ineffective.

Predictions of responses to multiple predators

The observed patterns of response support a number of predictions about how prey should modify their behaviour when multiple predators are present.

1. Predator avoidance will take place in the form of a hierarchy when: (a) One predator presents substantially higher predation risk than another, (b) one predator modifies the predation risk of another resulting in condition (a), or (c) when post-contact defences are inadequate for one predator (i.e., it presents a much higher predation risk once encountered), which warrants a more conservative avoidance strategy to that predator.
2. Compromise antipredator behaviour, where behaviour is intermediate between that associated with

either predator, should be observed when there is not a large disparity in the predation risk presented by the two predators, but avoiding one predator conflicts with avoiding another. It may also be used when one predator presents substantially higher predation risk than another but a relatively safe compromise is available that will reduce vulnerability to both predators.

3. Accumulative control will be used only when responses to different predators do not conflict. Thus, only when a change in behaviour (e.g., an increase in refuge use) enhances a prey's ability to avoid both predators should there be a further increase in that behaviour when both predators are present.

When testing those predictions behavioural ecologists will need to use natural predator-prey combinations, accurately assess relative risks of predation and the temporal variation in predation risks, and measure behaviours that reflect responses in a 'natural setting'. For example, even though a predatory benthic fish, the longnose dace *Rhinichthys cataractae*, attacked *B. tricaudatus* nymphs at a rate three times higher than a predatory stonefly (*Claassenia sabulosa*) in a laboratory experiment, *B. tricaudatus* nymphs did not adjust their foraging behaviour to respond to the presence of the different predators (Scrimgeour and Culp 1994). The authors explained this anomalous result by suggesting that attacks may not have been a good measure of predation risk. However, another behavioural response variable (dispersal from risky patches) more closely reflected expected avoidance behaviour of these two predators. In another experiment with a predatory benthic sculpin, *Cottus bairdi*, and a predatory stonefly (*Agnatina capitata*), *B. tricaudatus* did not respond to the sculpin even though it was capable of consuming more prey than the stonefly (Soluk and Collins 1988b). In this case the authors suggested that *Baetis* did not

respond to the fish because the sculpin is not normally a major predator of *Baetis* in nature. Thus, prey will only respond to predators that they recognise as a threat.

Importance of multiple predators in natural systems

The type of antipredatory behaviour that a prey animal uses will ultimately influence its individual fitness and its distribution and abundance in nature. In our system the natural drift and positioning periodicities of mayflies observed in natural streams represent behavioural trade-offs between maximising resource acquisition while avoiding both drift-feeding trout and benthic-feeding stoneflies (Peckarsky 1996). Both trout and stoneflies alone and together have been shown to affect *Baetis* fitness (Peckarsky et al. 1993, Peckarsky and McIntosh 1998). However, a behavioural trophic cascade, whereby the combined nonlethal effects of stoneflies and trout can lead to a reduction in grazing that increases algal abundances, was only observed when both predators were present (Peckarsky and McIntosh 1998). Models of predator-prey systems involving multiple predators also indicate that the type and magnitude of antipredator behaviour shown by prey can alter the fitness and coexistence of predators (Matsuda et al. 1993).

Due to the interactive effects of multiple predators on prey behaviour, patterns of predator effects at the population level will not necessarily follow those predicted by responses to individual predators. This is particularly important in streams where predators clearly affect prey dispersal. We predict that a range of different behavioural options will be used by prey to deal with multiple predators in different situations. Those predictions now need to be tested amongst a range of different predator-prey systems.

Table 3. Factorial ANOVA for the number of *Baetis* nymphs drifting in experiments with different predators (\pm stoneflies and \pm brook trout odour) at different times before and after the addition of predators. Data were square root transformed.

	Source	df	MS	F	P
Before	Fish	1	0.2051	1.011	0.336
	Stonefly	1	0.5287	2.607	0.134
	Fish \times Stonefly	1	0.7065	3.483	0.089
	Error	11	0.2028		
5 min	Fish	1	8.105	19.68	0.002
	Stonefly	1	6.510	15.81	<0.001
	Fish \times Stonefly	1	0.1421	0.3452	0.568
	Error	12	0.4118		
20 min	Fish	1	27.38	29.54	<0.001
	Stonefly	1	5.261	5.676	0.035
	Fish \times Stonefly	1	4.795	5.173	0.042
	Error	12	0.9270		
60 min	Fish	1	58.09	58.48	<0.0001
	Stonefly	1	0.0805		0.781
	Fish \times Stonefly	1	0.7375		0.406
	Error	12	0.9934		

Table 4. Factorial ANOVA for the number of *Baetis* nymphs on top of stones in experiments with different predators (\pm stoneflies and \pm brook trout odour) at different times before and after the addition of predators. Data were square root transformed.

	Source	df	MS	F	P
Before	Fish	1	0.9771	2.857	0.117
	Stonefly	1	1.062	3.105	0.104
	Fish \times Stonefly	1	0.2505	0.7325	0.409
	Error	12	0.3420		
5 min	Fish	1	5.103	14.19	0.003
	Stonefly	1	0.1644	0.4574	0.512
	Fish \times Stonefly	1	0.403	1.122	0.310
	Error	12	0.359		
20 min	Fish	1	5.854	9.713	0.009
	Stonefly	1	0.127	0.211	0.654
	Fish \times Stonefly	1	0.0286	0.0474	0.831
	Error	12	0.6027		
60 min	Fish	1	1.853	5.133	0.043
	Stonefly	1	0.243	0.673	0.428
	Fish \times Stonefly	1	0.165	0.458	0.511
	Error	12	0.361		

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