

Are mayfly anti-predator responses to fish odour proportional to risk?

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With 1 figure

Abstract: Adaptive prey behaviour should include responses to predators in proportion to the risk they present. We tested whether two salmonids fishes, the introduced brook trout (*Salvelinus fontinalis*) and native Colorado River cutthroat trout (*Oncorhynchus clarki pleuriticus*), presented different predation risks to *Baetis bicaudatus* mayflies, and whether mayflies used chemical cues to adjust their behaviour in proportion to that risk. In a feeding trial the native cutthroat trout consumed 60% more *Baetis* than brook trout. To test the antipredator responses of *Baetis* to fish chemical cues, we added water from holding tanks containing cutthroat, brook trout or goldfish (a non-predator), to microcosms with *Baetis* larvae. We measured rates of mayfly movement in the water column (drift), which is risky behaviour in the presence of drift-feeding predators. Nocturnal drift was substantially depressed in the presence of cutthroat trout odour, intermediate in microcosms with brook trout odour and relatively high in both goldfish and control treatments. Combined, these results indicate that risky behaviour decreased as predation risk increased. This result implies that *Baetis* used chemical cues produced by cutthroat trout, and brook trout to adjust their behaviour in proportion to the risk posed by those predators.

Key words: Chemical cues, predatory fish, predator-prey interactions, Salmonidae, mayflies, fish odour, stream invertebrate drift.

Introduction

A wide range of aquatic organisms gain information about potential predation risk from chemical signals present in the water (BRÖNMARK & HANSSON 2000). Chemical signals are useful in aquatic environments because the

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aquatic medium carries information to prey enabling them to assess predation risk (BURKS & LODGE 2002). Adaptive prey behaviour should include responses to predators in proportion to the risk they present (HELPMAN 1989), especially if anti-predator responses involve fitness costs (e. g., DAHL & PECKARSKY 2002). Such threat-sensitive responses to predators using chemical cues require that prey be able to differentiate among a multitude of chemical signals. Moreover, if chemicals associated with particular taxonomic groups of predators are similar (MIRZA & CHIVERS 2001), distinguishing between related species presenting different risks could be difficult. Exotic predators may also pose an additional problem, because prey may need to evolve appropriate responses to novel predators (KORPI & WISENDEN 2001).

Mayfly larvae are vulnerable to predatory trout that feed on prey drifting in the water column of streams (DOUGLAS et al. 1994). In many high-altitude headwater streams of western Colorado introduced brook trout, *Salvelinus fontinalis* (MITCHILL), have replaced native Colorado River cutthroat trout, *Oncorhynchus clarki pleuriticus* (COPE), as the main predatory salmonid. Although salmonids pose high predation risk during the day, *Baetis* spp. (Ephemeroptera; Baetidae) are also vulnerable to predation at night and reduce their nocturnal drift when brook trout odour is present (MCINTOSH et al. 1999, 2002). We tested whether brook trout and cutthroat trout presented different predation risks to *Baetis bicaudatus*, and whether *Baetis* were able to use chemical cues to differentiate between the two predators and thereby adjust their behaviour in proportion to the risks. We also measured mayfly responses to goldfish (*Carassius auratus*), as a completely novel, non-predatory fish to test the possibility of a general *Baetis* response to the 'smell of fish'.

Methods

Feeding experiment

To compare the predation risk posed by brook trout and cutthroat trout we measured their consumption of *Baetis* larvae in circular flow-through, plastic mesocosms (wetted surface area: 0.82 m², mean \pm SE water depth: 17.3 \pm 0.8 cm, mean \pm SE current velocity: 0.66 \pm 0.01 m/s; MCINTOSH et al. 2002). Filtered water (temperature range 4–9 °C) was gravity-fed to the mesocosms from a fishless stream near the Rocky Mountain Biological Laboratory in western Colorado; and cobbles (5–15 cm) were added to each mesocosm to provide cover.

On 6 August 1996 we added 200 *Baetis bicaudatus* larvae (mean \pm SE head capsule width: 0.85 \pm 0.10 mm) from the adjacent East River to each of 10 mesocosms, simulating natural densities (244/m², MCINTOSH et al. 2002). The East River contains moderate densities of brook trout (0.11 fish/m², see MCINTOSH et al. 2002), but only 2 cutthroat trout were recorded among the >200 brook trout captured in quantitative electrofishing in the East River in 1996–97. At 17:10 h (mountain daylight saving

time, MDST), one brook trout (mean \pm SE fork length, 255 ± 7 mm) was added to four randomly selected mesocosms, one cutthroat trout (254 ± 13 mm) to four other mesocosms, and two mesocosms were left as fishless controls. The non-predatory fish (goldfish) were not included in this experiment. Brook trout were captured by electrofishing the East River and cutthroat trout were captured from Copper Creek, a tributary of the East River, and acclimated in holding tanks prior to the experiment. During the experiment the mesocosms were covered with translucent covers (2 mm diameter white mesh) and left undisturbed overnight. After 17 hours (6 h daylight) the fish were removed and all remaining mayflies were counted.

Response experiment

To test the antipredator responses of *Baetis bicaudatus* to fish chemical cues, we added water from holding tanks containing brook trout (introduced predator), cutthroat trout (native predator) or goldfish, (a novel, non-predatory fish) to microcosms each stocked with 15 *Baetis* larvae (mean \pm SE head capsule width = 1.01 ± 0.10 mm) from the East River. The microcosms were circular (15 cm diam.), plexiglass, flow-through chambers housed in a translucent greenhouse beside the East River (see PECKARSKY & COWAN 1991 for a diagram and full description). Jets in the microcosms produced circular flow and a central standpipe with a 0.5 mm mesh outlet allowed water to drain but retained the *Baetis*. Fishless stream water was gravity-fed to 40 microcosms (velocity and depth ranges: 9–11 cm/s and 3–4 cm, respectively) each containing two periphyton-covered rocks (≈ 5 cm diameter). Each of four fish cue treatments (2 brook trout, 2 cutthroat trout, 2 goldfish and fishless controls) was randomly assigned to one of four 96 L plastic tanks located on a hill above the greenhouse, receiving 0.11 L/s of the same fishless water. Water from these tanks was piped to the greenhouse and dripped into the microcosms at a mean \pm SE rate of 9.2 ± 1.00 mL/s. Brook (combined biomass: 220 g) and cutthroat trout (210 g) were captured by electrofishing from the East River and Copper Creek, respectively, and acclimated before the experiment in a large holding tank where they were fed *ad libitum* on mayflies. Goldfish (240 g) were obtained from a local pet shop and acclimated separately. They were also offered mayflies, but did not consume any.

Mayflies were introduced to the microcosms on the morning of 14 July 1997 and the fish were added to the odour tanks at 16:00 h. While in the odour tanks all fish were also offered *Baetis* mayflies *ad libitum* to ensure that any cues produced in the predation process were also present. Counts of larvae drifting past a fixed transect during one minute were made by two observers on two occasions at night on 14 July (22:00 and 22:30 h) and during daylight (10:00 and 10:30 h) the next day. Observers used a dim red light, which does not affect *Baetis* behaviour, to make night observations (MCINTOSH et al. 2002).

Results and discussion

The fish took up foraging stations and actively consumed *Baetis* in the mesocosms during the feeding experiment. The number of mayflies disappearing

from the experimental tanks was significantly different between treatments (one-way ANOVA: $F_{2,7} = 11.94$, $P < 0.01$). Tukey post-hoc tests indicated that significantly more mayflies disappeared from brook trout and cutthroat trout tanks than control tanks ($P < 0.05$). An average of five mayflies disappeared from the control tanks, so estimates of fish consumption were adjusted accordingly. Cutthroat trout consumed on average 60 % more *B. bicaudatus* than brook trout during the feeding trial (Fig. 1), but the Tukey post-hoc tests indicated there was no significant difference in the number missing from brook trout tanks compared to cutthroat tanks ($P = 0.14$). In the antipredator response experiment, during the day no *Baetis* drifted and most larvae resided under rocks in the microcosms. At night the larvae actively moved around the microcosms and grazed periphyton on the rock tops. Nocturnal movement (drift) was significantly different among treatments (one-way ANOVA: $F_{3,36} = 10.12$, $P < 0.001$), and was lowest in the presence of cutthroat trout odour, intermediate in the brook trout microcosms and relatively high in both the goldfish and control treatments (Fig. 1). Tukey post-hoc tests indicated that mayflies in both brook trout and cutthroat trout tanks drifted significantly less than those in control tanks ($P < 0.01$ in both cases), that drift in goldfish tanks was not significantly different to control tanks ($P = 0.60$) and that drift in brook trout and cutthroat trout treatments was not significantly different ($P = 0.16$). How-

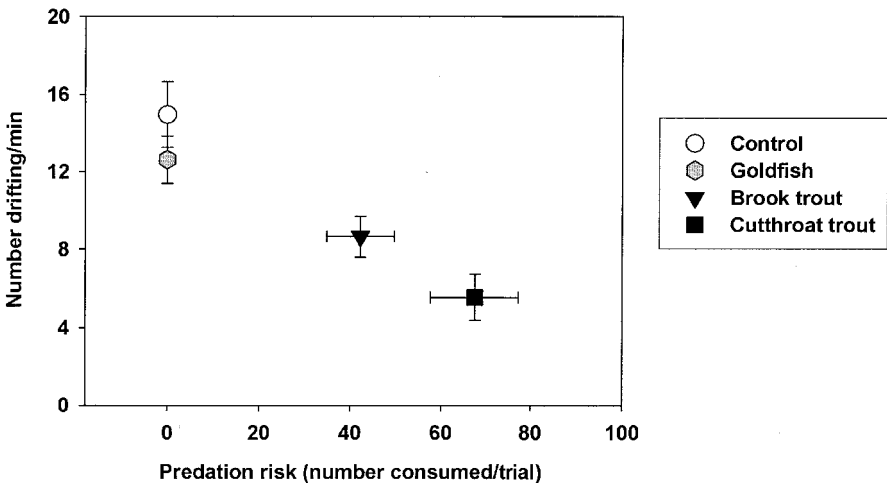


Fig. 1. The relationship between mean (\pm SE) predation risk measured by the number of *Baetis* mayfly larvae consumed in a feeding trial and behaviour of *Baetis* measured by mean (\pm SE) nocturnal drift rate in microcosms receiving chemical cues from four different predation regimes. *Baetis* are most vulnerable to predatory fish when drifting in the water column.

ever, a significant negative correlation between vulnerability to predation and nocturnal drift behaviour indicated that nocturnal drift rate decreased as predation risk increased (Fig. 1, $r = 0.96$, $P < 0.05$).

The observation that *Baetis* reduces nocturnal drift in response to odour from predatory fish is consistent with previous experiments and observations (e. g., McINTOSH et al. 1999, 2002), and is not surprising given that predatory salmonids consume substantial numbers of drifting mayflies at night (GIRoux et al. 2000, McINTOSH et al. 2002). Drifting *Baetis* were moving circularly around the microcosms so they may have reduced their rate of entry to the water column, the distance they drifted or both, leading to a reduction in their subsequent vulnerability to trout predation.

The negative correlation between predation risk and nocturnal drift also implies that *Baetis* adjusted their behaviour in proportion to the risk posed by the predators. Prey frequently adjust their behaviour according to the level of predation risk (SIH 1987), but the most interesting aspect of this study is that prey responded to chemical cues alone. A wide range of aquatic prey species use chemical cues to distinguish between predators that pose different risks (see reviews by BRÖNMARK & HANSSON 2000 and WISENDEN 2000). Some of the most sophisticated behaviours come from fish living in lakes. For example, PETERSSON et al. (2000) observed that crucian carp could detect and respond appropriately to different sizes of predatory pike. Our results indicate that prey detection of predators using chemical cues may be widespread and that some insects from running waters show threat-sensitive, graded responses to predators according to risk (HELFMAN 1989).

The chemicals used by *Baetis* to assess predation risk are unknown; but previous microcosm and field experiments have shown that *Baetis* can detect variations in the concentration of brook trout odour in stream water (McINTOSH et al. 1999, 2002). To do this, prey may detect cues related to the number of prey consumed by a predator (see ROCHETTE et al. 1997). Similarly, prey fish readily detect and respond to novel predators by detecting alarm substances or cues from injured conspecifics in the water (e. g., PETERSSON et al. 2000, MIRZA & CHIVERS 2003).

Under this scenario *Baetis* antipredator behavior may be triggered directly by consumption rates of predatory salmonids. Thus, the graded behavioural response to the three different fish species may have been prompted by variation in the concentration of 'victim cues' related to the rates of consumption of *Baetis*. Interestingly, *Baetis* did not respond to goldfish; thus, the lack of 'victim cues' in the water may provide a reliable mechanism for recognising non-predatory fish. Finally, the greatest antipredatory response by *Baetis* was to cues from native cutthroat trout, which are rare but still pose considerable risk to *Baetis*. Our study suggests that investigation of prey responses to chemical cues from predators may be very fruitful for predicting whether they

will recognise threats and ultimately be vulnerable to exotic predators in aquatic systems.

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