

Impacts of an introduced predatory fish on mayfly grazing in New Zealand streams

Abstract—The replacement of native common river galaxias (*Galaxias vulgaris* Stokell) by introduced brown trout (*Salmo trutta* L.) in many streams of the Taieri River system, New Zealand, has altered the predation regime for *Deleatidium* mayfly nymphs. Brown trout present a higher predation risk to mayfly prey during the day than at night but common river galaxias present similar risks both day and night. To determine whether this alteration has changed mayfly grazing behavior, we investigated the diel gut fullness of *Deleatidium* nymphs in five galaxias streams and five trout streams. Gut fullness was measured by the fluorescence of algal pigments extracted from *Deleatidium* guts. Nymphs from trout streams generally had fuller guts at night, whereas nymphs from galaxias streams tended to have more in their guts during the day or similar amounts both day and night. The difference reflects the high predation risk presented by brown trout during the day, restricting mayfly access to the algae-rich upper surfaces of stones. These results indicate that the influence of predatory fish on mayfly grazing is detectable at a large spatial scale in the Taieri River system.

A large array of behavioral interactions between predators and prey have been identified in stream communities. Predators can restrict prey movement, microhabitat selection, positioning, and the ability of prey to harvest resources, causing important alterations in prey diet, growth, fecundity, and drift (e.g. Macchiusi and Baker 1992; Peckarsky et al. 1993; Cowan and Peckarsky 1994; Forrester 1994). However, most of these studies have been confined to comparisons of pairs of streams (*but see* Flecker 1992; Douglas et al. 1994) or have been conducted in artificial laboratory settings. To determine the real importance of such predator-prey interactions it is necessary to ascertain whether they occur in natural communities and across larger spatial scales. Here we describe a study to determine whether the influence of predatory fish on mayfly grazing periodicity is present at an inter-tributary spatial scale.

In the Taieri River catchment (South Island, New Zealand) the replacement of a native predatory fish, the common river galaxias (*Galaxias vulgaris* Stokell) by introduced brown trout (*Salmo trutta* L.) in many streams has resulted in a significant change in the predation regime affecting prey (McIntosh and Townsend 1995). Experiments in artificial stream channels and observations of field gut contents have shown that brown trout forage from positions in the water column, rely principally on vision to capture prey and have larger reaction distances and rates of consumption during the day (McIntosh and Townsend 1995). Common river galaxias, which move around more at night, forage from benthic positions, probably use mechanical cues to detect prey, and consume

similar numbers of prey and have similar reaction distances both day and night (McIntosh and Townsend 1995).

Experiments in laboratory stream channels and in situ stream channels have shown that *Deleatidium* and *Nesameletus* mayfly nymphs avoid the upper surfaces of stones during the day in the presence of trout. They primarily graze algae-rich upper surfaces at night (McIntosh and Townsend 1994; McIntosh 1994). These experiments have also shown that the number of *Nesameletus* and *Deleatidium* occupying the substratum surface is reduced when galaxias are present compared to when no fish are present, especially at night when galaxias are most active, but the number on the substratum surface is lower still when trout are present. In the Shag River (adjacent to the Taieri drainage) these changes in grazing behavior have a major impact on the trophic cascade identified by Flecker and Townsend (1994). Reductions in grazing pressure result in higher algal abundances on the sides and tops of cobbles in channels with trout (McIntosh 1994). This trophic cascade was mediated by the impact on mayfly behavior described above because there was no difference in the biomass of grazers in channels with and without trout.

To determine whether the replacement of *G. vulgaris* by *S. trutta* has resulted in a general change in grazing behavior, we examined the grazing periodicity of *Deleatidium* mayflies in five streams in the Taieri River catchment containing common river galaxias (but not trout) and five streams containing brown trout (but not galaxias). Our study animal, the leptophlebiid mayfly *Deleatidium* spp., is a ubiquitous member of New Zealand stream faunas, comprising as much as 90% of total macroinvertebrate numbers in streams of the Taieri River catchment (Scarsbrook and Townsend 1993). It is impossible, given current taxonomy, to identify individual species of *Deleatidium* as nymphs, although two species groups are recognized (Winterbourn and Gregson 1989). *Deleatidium* is a collector-browser and consumes a large amount of algae (Rounick et al. 1982; Winterbourn et al. 1984; Collier and Winterbourn 1990). If brown trout were inhibiting mayfly grazing during the day, then we would expect mayflies from streams with trout to have more algae in their guts at night than during the day. Thus, we tested the hypothesis that the ratio of night : day gut contents in *Deleatidium* nymphs was higher in streams with trout than in streams with galaxias.

Five streams with trout and five with galaxias were selected to be similar in their physical characteristics. These were either different tributaries of the Taieri River separated by large distances (> 1 km) or by waterfalls. All were relatively small (avg width, 1.3–6.2 m), but the set included first-, second-, and third-order streams. They all drained tussock and pasture grass catchments and were

dominated by schist substrates. The taxonomic status of some populations of common river galaxias is under review so the galaxias streams were chosen to contain the same genetic group of galaxias (Allibone et al. 1995).

Two weeks before mayfly sampling we measured the physical characteristics of a 40-m riffle stretch from each stream to ensure any differences in mayfly grazing observed between streams with galaxias and trout could not be attributed to differences in the abiotic environment. Substratum characteristics were assessed by classifying 100 randomly collected particles according to the Wentworth scale (as described by Gordon et al. 1992). Width and depth were measured on five equally spaced transects. Current velocity was estimated by measuring the time it took fluorescein dye to travel the length of the site (judged visually). Discharge was calculated from the current velocity (as described previously) and cross-sectional area of the depth-width transects. The proportion of the stream bank undercut together with the bed geology and the nature of the surrounding vegetation were all assessed visually. Potential differences in the physical characteristics of sites with trout and galaxias were assessed by discriminant functions analysis on the scores of principal components calculated from these physical measurements. Bed geology and surrounding vegetation were not included in this analysis because they were similar in all streams (schist rocks and tussock grassland). The streams had very little canopy cover because the tussock grasses do not extend very far over the water.

Mayfly sampling took place in three 24-h periods spread over 10 d in December 1993 centered around a dark phase of the lunar cycle in the austral summer. Between 10 and 12 *Deleatidium* nymphs were collected from each stream with a kick net between 1200 and 1500 hours (the day sample) and 0000 and 0300 hours (the night sample). Nymphs were large, late instar individuals of both sexes that did not have black wing pads (i.e. were not immediately about to emerge). Both groups of *Deleatidium* were present in samples and our observations of gill morphology indicate there were unlikely to be distinct species in trout and galaxias streams. Similar sizes of nymphs were sampled in galaxiid and trout streams (mean head capsule widths \pm SE = 1.71 ± 0.04 mm and 1.75 ± 0.02 mm, $t_8 = -1.02$, $P = 0.35$). After capture, nymphs were placed in individual vials and immediately frozen.

We used the gut fluorescence technique described by Cowan and Peckarsky (1990) to quantify the amount of algal pigment in the guts of *Deleatidium*. After measuring their head capsule widths, whole nymphs were macerated to maximize exposure of the gut contents to the solvent and then extracted for 24 h in 90% ethanol after initial boiling at 78°C (the boiling point of the solvent) for 5 min. Extracts were then filtered (Whatman GF/F glass-fiber filter, 2.5-cm diam) and read on a Turner model 450 fluorometer, fitted with Turner NB440 excitation and SC665 emission filters, and calibrated in the range of 0.25–100 $\mu\text{g liter}^{-1}$ with a standard solution of pure chlorophyll *a* (Chl *a*) from *Anacystis nidulans* (Sigma Chemical No. C-6144). Total pigment in the gut (ng ind.^{-1}), calculated as the sum of Chl *a* and its degradation prod-

ucts, pheopigments, was used as the measure of feeding activity on algae (Cowan and Peckarsky 1990).

Successful detection of diel differences in foraging requires that the passage of pigment through the gut is faster than the time between sample intervals. Otherwise the estimates of foraging rates may be confounded by feeding at times other than during the sampling period. To test this assumption we measured gut clearance times for *Deleatidium*. Eighty four *Deleatidium* were collected at 0300 hours, when we expected gut contents to be full, from a stream containing brown trout (Water of Leith—NZMS 260 grid ref.:I44 162824) on 15 November 1993. Twelve randomly selected animals were frozen immediately to determine initial gut fullness. The rest were placed in groups of 12 in aerated containers (20 \times 20 cm) that had been thoroughly scrubbed to remove any potential food and transported to the laboratory. The animals from one randomly selected container were then sampled at 0.5, 1, 2, 4, 8, and 12 h after the initial collection to measure gut pigment.

A pilot study using *Deleatidium* from the Water of Leith (where we have previously observed *Deleatidium* to feed nocturnally—unpubl. data) was conducted to determine the sample size required to detect diel differences in gut fullness. Twelve animals were sampled at 0300 hours and another 12 at 1500 hours on 15 November 1993 and gut pigment was quantified by the above technique.

As mayflies collected during the day and at night in some sites were of different sizes, we corrected total gut pigment for differences in *Deleatidium* ash-free dry weight (AFDW) based on head capsule width with the equation

$$\text{AFDW}(\text{mg}) = 0.001784 \times [\text{head width} + 1 (\text{mm})]^{6.8254}.$$

This equation was generated from a regression line ($r^2 = 0.94$, $N = 30$) relating the head width of *Deleatidium* before drying with AFDW after drying for 24 h at 60°C and ashing at 550°C for 1 h. We expressed the weight of gut pigment as $\text{ng Chl } a \text{ ind.}^{-1}$ (mg AFDW) $^{-1}$. Patterns in gut pigment were then assessed between streams with trout and galaxias by means of nested ANOVA (Zar 1984).

Further sampling of the algal community was not undertaken because of a flood just after the completion of mayfly gut sampling.

Examination of *Deleatidium* gut clearance rates showed that over half of the pigment disappeared within the first hour (Fig. 1). Because mayflies could not feed during these trials, our estimate of gut clearance time is likely to overestimate the actual time. In the pilot study we observed a significant difference between day and night gut fullness (means \pm SE = $1,601 \pm 279$ and $4,023 \pm 654$ ng ind.^{-1} , $t_{22} = -3.41$, $P = 0.004$). Thus, the sampling interval, sample size, and methods were likely to detect diel differences in *Deleatidium* gut fullness.

There were no consistent differences in the physical characteristics of the galaxias and trout streams (Table 1, Fig. 2). Discriminant functions analysis, with cross-validation, could not correctly classify the sites according to

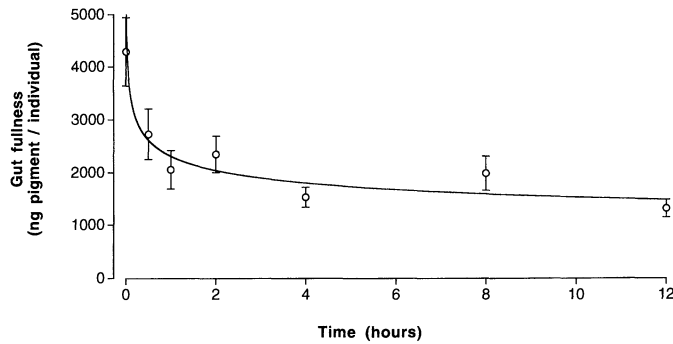


Fig. 1. Gut clearance of *Deleatidium* over 12 h. Values are means (\pm SE). The equation for the curve is $y = 2,311x^{-0.18}$, $r^2 = 0.66$.

fish regime from the scores of the first four principal components which together accounted for 96% of the variation in physical variables.

There were considerable differences between streams in the median amounts of pigment in *Deleatidium* guts but no consistent differences between the galaxias and trout streams in overall gut fullness (Fig. 3). ANOVA showed there was a significant fish by time interaction (Table 2). This interaction indicates that *Deleatidium* from galaxias streams have different diel patterns of gut fullness than *Deleatidium* from trout streams. Figure 3 shows that nymphs from the streams with galaxias either had similar amounts in their guts during the day and at night or more during the day, whereas nymphs from the streams with trout generally had more in their guts at night.

To test our hypothesis that the ratio of night : day gut content is higher in trout streams, we divided the mean gut content from each stream during the night by that

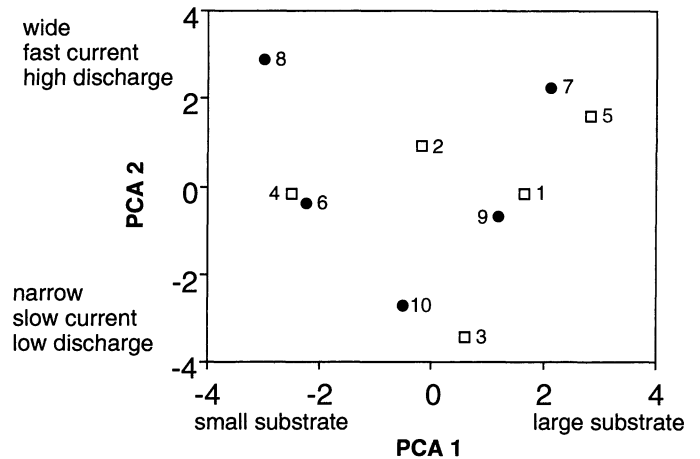


Fig. 2. The scores from the five galaxias stream sites (□) and five trout stream sites (●) for principal component 1 (PCA 1) vs. principal component 2 (PCA 2). Numbers correspond to the site labels in Table 1. These two components account for 75% of the variation in measured physical variables.

during the day to produce a night : day gut fullness ratio for *Deleatidium* from the two categories of stream. The ratios were significantly higher [Mann-Whitney $U_{0.05(1),5,5} = 23$, $P = 0.025$] in streams with trout compared to streams with galaxias (Fig. 4).

Our study correlates the species of predatory fish present in a stream with patterns in mayfly behavior. To be confident that differences in the nature of the predation risk are responsible for the observed patterns it is important to rule out the effect of some other covarying factor. We found no consistent differences in the physical characteristics of streams with trout and galaxias. In the Taieri River sites, trout are larger than galaxias (mean

Table 1. Grid references (NZMS 260 map series), physical measurements, and fish densities (m^{-2}) from streams containing common river galaxias and streams containing brown trout. Fish densities were obtained by electrofishing (method of Townsend and Crowl 1991).

Site	Grid ref.	Stream*			Part. size (mm)	Streambed†			Flow‡		Bank undercut (%)	Fish
		Alt.	W	D		Bo.	Co.	Gr.	Vel.	Dis.		
Galaxias streams												
1 Three O'clock Stream	I43:093118	380	3.1	11	8.0	4	78	18	0.34	0.114	43	2.35
2 Linn Burn	H42:656323	450	2.8	16	8.0	9	82	9	0.35	0.155	35	1.97
3 Kye Burn (above waterfall)	I41:015805	780	1.3	7	8.5	15	63	23	0.12	0.011	57	1.81
4 Healy Creek	I43:993783	740	2.6	17	9.0	20	70	10	0.26	0.118	15	1.32
5 Taieri River	H43:586098	770	4.8	15	7.5	4	67	29	0.55	0.421	52	0.69
Trout streams												
6 Sutton Stream	H43:686068	620	2.9	17	9	21	65	14	0.26	0.126	18	0.69
7 Shepherds Hut Creek	H43:633101	850	4.2	17	8	6	74	20	0.51	0.340	50	0.79
8 Styx Creek	H43:709272	570	6.2	19	9	23	70	7	0.44	0.527	25	0.29
9 Heeney Creek	I42:916323	240	2.67	9.8	8	7	74	19	0.32	0.082	5	0.83
10 Kye Burn (below waterfall)	I41:012803	740	1.33	7.2	8.5	15	63	23	0.12	0.011	58	0.62

* Altitude (m), width (m), and depth (cm).

† Percent boulders, cobble, and gravel.

‡ Current velocity ($m s^{-1}$) and discharge ($m^3 s^{-1}$).

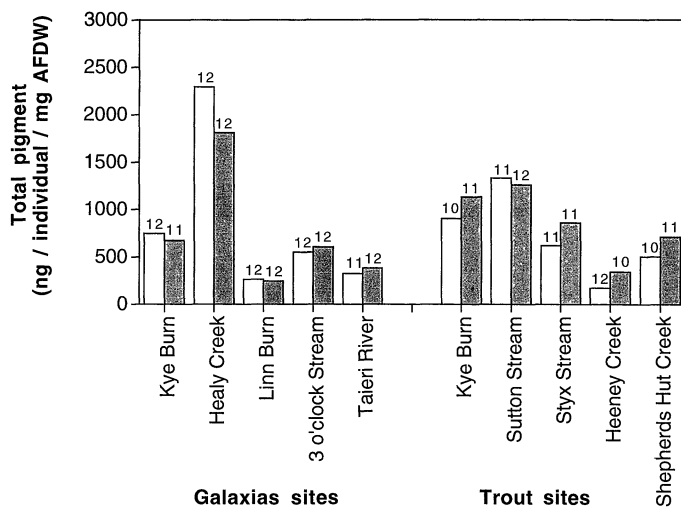


Fig. 3. The mean gut fullness measured in terms of total gut pigment for mayfly nymphs from streams with common river galaxias and trout during the day (unshaded bars) and at night (shaded bars). Number of nymphs sampled indicated above each bar.

weight \pm SE: galaxias = 3.72 ± 0.08 g; trout = 13.8 ± 0.99 g), but streams with galaxias and trout have similar fish biomasses present (4.05 ± 0.4 and 3.42 ± 0.37 g m^{-2} —C.R. Townsend unpubl. data) so the patterns are unlikely to be attributable to differences in fish biomass. Thus, the diel differences in mayfly gut fullness are probably associated with differences in the predation risk presented by trout and galaxias.

The significantly higher night : day gut fullness ratios in streams with brown trout compared to streams with galaxias indicate that *Deleatidium* in trout streams forage relatively more at night than do *Deleatidium* in streams with common river galaxias. This difference in behavior reflects the diel variation in predation risk imposed by brown trout and common river galaxias. Brown trout,

Table 2. Nested ANOVA for ln-transformed mayfly gut pigments [ng Chl *a* ind.⁻¹ (mg AFDW)⁻¹] in relation to the fish predation regime (galaxiids vs. trout: "fish"), the time of day (day vs. night: "time"), and the stream sampled ("site"). Streams are nested inside the predation regime, the "site(fish)" term is used as the denominator for the *F*-ratio test of the effect of the fish predation regime and the "time \times site(fish)" term is used as the denominator for the *F*-ratio test of the fish predation regime by time interaction. Mean-square terms are adjusted (adj. MS) for differences in the number of mayfly guts examined in each sample (as shown in Fig. 3).

Source	df	Adj. MS	<i>F</i> -ratio	<i>P</i>
Fish	1	0.5134	0.042	0.84
Site(fish)	8	12.1040		
Time	1	2.2102	5.82	0.017
Fish \times time	1	2.8651	8.30	0.02
Time \times site(fish)	8	0.3450		
Error	206	0.3798		
Total	225			

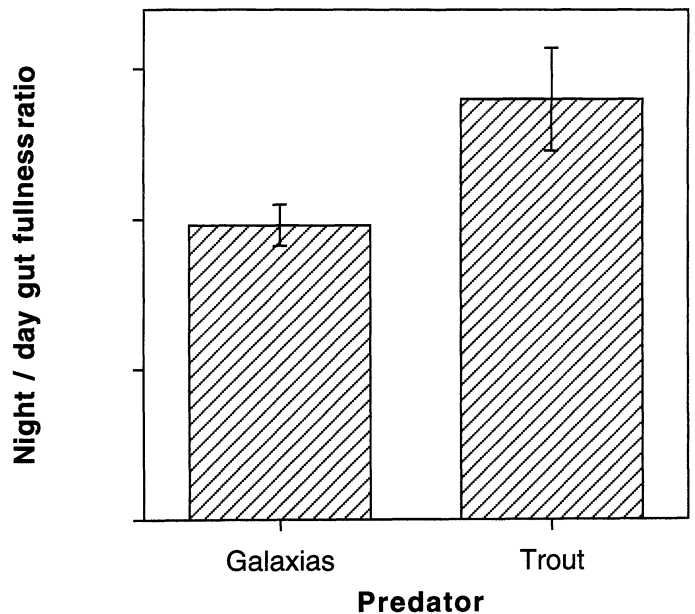


Fig. 4. Mean (\pm SE) night : day gut fullness ratios [measured in ng of total pigment ind.⁻¹ (mg AFDW)⁻¹] for streams with common river galaxias or brown trout.

like most visually feeding salmonids, present a higher predation risk during the day (Ware 1972; Walsh et al. 1988; Angradi and Griffith 1990); however, there is no diel difference in the predation risk presented by common river galaxias despite diel variations in their activity (McIntosh and Townsend 1995). Thus, when brown trout are present, both *Nesameletus* and *Deleatidium* mayfly nymphs avoid the upper surfaces of the substratum, where algal abundance is higher, during the day (McIntosh and Townsend 1994; McIntosh 1994).

Our results indicate that changes in mayfly positioning observed in in situ channels (McIntosh 1994) and in artificial laboratory stream channels (McIntosh and Townsend 1994) can cause changes in algal consumption which are detectable at a large scale in natural streams. Thus, the behavioral mechanisms important in producing a trophic cascade in the Shag River, where algal biomass was higher in in situ channels with trout (Flecker and Townsend 1994), are present generally in streams of the Taieri River catchment.

The impact of the fish predation regime on the diel periodicity of gut contents in our study was modest. However, the phenomenon seems to be common because studies on a third mayfly genus, *Baetis*, indicate nocturnal increases in gut contents in nymphs from trout streams compared to those from troutless streams (Cowan and Peckarsky 1994; Culp and Scrimgeour 1993). These results bolster our conclusion that the fish predation regime can influence mayfly foraging. Furthermore, our study shows that it is not only the presence or absence of fish that modifies mayfly foraging but that variations in the type of fish predator can have specific effects.

A growing list of studies (e.g. Power 1992; Peckarsky et al. 1993) reveals large potential for predators to influ-

ence important aspects of stream communities through their effects on prey behavior. Our study adds weight to the argument that such patterns are detectable at the large spatial scale of multiple stream comparisons (see also Flecker 1992; Douglas et al. 1994; Friberg et al. 1994). Ecologists should now determine whether behavioral interactions such as those described here affect other aspects of stream ecology including algal biomass and prey abundance.

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