Facilitation of periphyton production by tadpole grazing: functional differences between species

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SUMMARY

1. This study examined how interactions between resources that vary in edibility, and herbivores that vary in ability to acquire resources, control primary productivity. In a northern California river, grazing on *Cladophora glomerata*, a relatively inedible filamentous green alga, and its more nutritious epiphytic diatoms, was manipulated by exposing cobbles to tadpoles (*Rana boylii* or *Hyla regilla*) or excluding tadpoles.
2. *Rana* indirectly facilitated *Cladophora* by removing diatoms, whereas *Hyla* did not significantly change biomass relative to controls. Algal ash-free dry mass on cobbles in *Rana* treatments was 65 and 72% greater than on controls in two years of investigation (1991 and 1993). *Rana* decreased epiphytic diatom biovolume by 56% and detritus by 87%.
3. Because nitrogen excretion rates of *Hyla* and *Rana* were similar, the differences in effect between the two species were probably due to their roles as consumers rather than as recyclers.
4. The net effect of *Rana* on periphyton was a 10% increase in areal specific primary productivity (mg O_2 h^{-1} m^{-2}); *Hyla* caused an 18% decrease. *Rana* decreased biomass-specific productivity (mg O_2 h^{-1} g^{-1}) 44%; *Hyla* had no effect.
5. In tadpole exclosures, grazers such as baetid mayfly larvae (mostly *Centroptilum* sp.) were 4.7 (1991) and 1.8 (1993) times more abundant, and midge larvae (Chironomidae) were 2.5 (1991) and 2 (1993) times more abundant than in *Rana* enclosures. Invertebrate assemblages in *Hyla* enclosures, however, were similar to exclosures. Few predatory insects and fish colonized *Rana* enclosures. Path analyses indicated that *Rana* affected macroinvertebrates via both interference and exploitation of epiphytic diatoms.

Introduction

When only the direct effects of herbivores consuming plants are considered, grazer enhancement of primary production appears paradoxical. Indeed, a review of many experiments with periphyton grazers shows positive removal rates of algal biomass (Cattaneo & Mousseau, 1995). Grazing can increase primary production of an assemblage, however, when some individuals indirectly benefit from an environment in which their neighbours are grazed (Westoby, 1989). Indirect effects and feedbacks between trophic levels are necessary to understand the mechanisms of grazer-enhanced productivity (Power, 1990a, 1992a). These

indirect effects include nutrient regeneration (Lehman, 1980; Osborne & McLachlan, 1985; Bergquist & Carpenter, 1986; Sterner, 1986), removal of shading silt and detritus (Power, 1990a) and removal of epiphytes (Brawley & Adey, 1981; Shacklock & Doyle, 1983; Brönmark, 1985; Underwood & Thomas, 1990; Dudley, 1992).

The net impact of direct and indirect effects of one trophic level on another also depends on the traits of the diverse species within each level (Strong, 1992; Hunter & Price, 1992). For example, diverse aquatic grazers, such as tadpoles (*Rana utricularia* Harlan) and

Daphnia laevis Birge, vary in their ability to exploit heterogeneous algal resources and thus differ in their ability to propagate the effects of nutrient enrichment to higher trophic levels (Liebold & Wilbur, 1992). Even if diverse taxa consume similar diets, they may function quite differently as nutrient recyclers (Vanni & Findlay, 1990). When grazers vary in their mineral requirements and the extent to which they sequester a particular element (N or P), they can accentuate nutrient limitation of algae (Hessen & Andersen, 1992; Sterner & Hessen, 1994). Simultaneously, plants vary in their edibility and make trade-offs between defence and nutrient acquisition. Herbivory and plant competition can therefore interact to cause compensatory increases of inedible plants (Liebold, 1989; Grover, 1995).

This study examines how interactions between resources that vary in edibility, and taxonomically similar herbivores that vary in ability to acquire resources, affect primary productivity. In a northern California river, I manipulated which tadpole species grazed *Cladophora glomerata* (L.) Kuetzing, a filamentous green alga not readily edible by tadpoles (Savage, 1952), and its nutritious epiphytic diatoms (Kupferberg, Marks & Power, 1994). I tested the null hypothesis that larvae of foothill yellow legged frogs (*Rana boylii* Baird) and Pacific treefrogs (*Hyla regilla* Baird and Girard) do not differ in their effects as grazers on *Cladophora*. After rejecting this hypothesis because *Rana* facilitated *Cladophora* but *Hyla* did not, I compared the following possible mechanisms leading to the differences in effects. *Rana* tadpoles enhanced macroalgae by:

1 removing the negative shading effects of epiphytic diatoms, detritus and senescent tissue;

2 transforming these algal competitors into excreted nutrients thereby fertilizing the macroalgae; and

3 competing with and displacing macroinvertebrate grazers, such as tuft weaving midges.

Hyla, in contrast, had negative or no net effects on macroalgae because:

1 it did not effectively exploit epiphytes and detritus;

2 it did not recycle nutrients; or

3 it did not compete with macroinvertebrate grazers.

Materials and methods

Study system and site

Research was conducted in the South Fork Eel River at the Angelo Coast Range Reserve, near Branscomb,

CA (39°44′N, 123°39′W). During the summer low flow period, the rock-bedded river is clear and sunlit, and blooms of *Cladophora glomerata* occur. This filamentous green alga is prevalent and important ecologically in freshwater habitats worldwide (Whitton, 1970; Dodds & Gudder, 1992). In the South Fork Eel, *Cladophora* abundance is influenced by many factors, including grazing by invertebrates (Power, 1990b, 1991), flooding and nutrients (Power, 1992b). Nutrient inputs from the coniferous watershed are low (undetectable to $10 \, \mu g \, l^{-1}$ PO_4-P and 5–$15 \, \mu g \, l^{-1}$ NO_3-N; M. Power, unpublished data).

Vertebrate grazers of *Cladophora* and its epiphytes in the South Fork Eel River include larvae of *Rana boylii* and *Hyla regilla*. Adult *Rana* are common along the banks of the river and its tributaries. In late spring, *Rana* congregate at historic breeding sites in the main stem where oviposition is highly clumped (Kupferberg, 1996b). *Hyla* occur along the river as well as in the surrounding meadows and forests of the watershed. As the river recedes during summer, tadpoles become locally abundant in shallow, near-shore habitats.

Enclosure and experimental designs

The presence/absence of tadpoles in flow-through enclosures was manipulated. Each enclosure (12.7 l white plastic buckets with two screened windows 12 cm × 16 cm, 1.0 mm fibreglass mesh) was stocked with two algae-covered cobbles. Cobbles were selected which visually appeared equivalent in size and quantity of periphyton, predominantly *Cladophora*. Macroscopic invertebrates were removed by rinsing and picking with forceps. Tadpoles (stages 27–30; Gosner, 1960) were randomly assigned to enclosures at a density of five individuals/bucket (equivalent to 25 ind. m^{-2}). Ambient near-shore tadpole densities, measured by sampling with a bottomless bucket enclosure, were per enclosure 2.1 ± 2.2 (range 0–8, $n = 14$) for *Rana* and 2.5 ± 2.5 (range 0–8, $n = 15$) for *Hyla*. Water depth was maintained at about 20 cm. In 1991 there were two treatments: *Rana* tadpoles present and tadpoles absent. Treatment assignment was completely randomized. In 1993 there were three treatments: *Rana* only, *Hyla* only, and no tadpoles. Treatment assignment was in randomized blocks. Each treatment was replicated seven times in both years. Experiments ran for 4 weeks in 1991 (1 August to

2 September) and for 6 weeks in 1993 (20 July to 2 September), with response variables measured at the ends of the experiments.

Algal abundance

Periphyton response was measured in terms of area-specific ash-free dry mass (AFDM) and *Cladophora* filament length at the ends of the experiments. In 1991 mean initial AFDM was estimated by scraping algae from a haphazardly chosen 1 cm^2 patch. No significant differences in biomass were found between treatments. In 1993 initial algal standing crops were not quantified, but were visually similar. Average *Cladophora* filament length for each replicate was calculated from three evenly spaced measurements per cobble. At the final harvest, all periphyton was scraped from cobbles. Samples were oven dried at 60 °C for 24 h, and then incinerated in a muffle furnace at 510 °C for 1 h. The vegetated region of each rock was wrapped with aluminium foil and excess folds were carefully cut away. To calculate surface area, the foil was weighed and the mass was multiplied by the mean area per gram. Differences in vegetated surface area between treatments were not significant for either year.

For 1991 and 1993 samples, ash : biomass ratios were calculated. Ash : biomass ratio was significantly correlated with total diatom biovolume ($R = 0.598$, $P = 0.02$, $n = 14$) in the 1991 samples, so it was used as an index of diatom abundance for the 1993 samples.

Net primary productivity

In 1993, tadpole effects on oxygen liberation were measured using *in situ* incubation after 6 weeks of grazing. A larger scale version of the well-known light and dark bottle method (Hall & Moll, 1975) was developed by constructing chambers large enough to contain cobbles, using inverted Rubbermaid® cake servers (8 l, 380 cm diameter, 140 cm height). The centre of the opaque lid was cut away, leaving a plastic ring that formed an airtight seal when the opening was covered with clear 0.13 mm thick Visquene® plastic and secured with clips. Irradiance was measured with a Licor light meter inside and outside an enclosure vertically and in the four cardinal directions at equivalent water depths. Irradiance inside and outside the chambers was nearly identical. Light measured perpendicular to the water's surface was 1506 μmol^{-1}

m^{-2} inside and 1547 μmol^{-1} m^{-2} outside. Measured parallel to the water's surface in the four cardinal directions, mean ± 1 SD irradiance was 382.3 ± 226.1 μmol^{-1} m^{-2} inside, and 327.7 ± 230.9 μmol^{-1} m^{-2} outside.

Net productivity of periphyton on cobbles was estimated by subtracting community respiration from gross productivity. Respiration was measured using readings from a YSI dissolved oxygen meter before and after a 12 h dark incubation. Cobbles were placed in chambers completely filled with water, and with no air bubbles up against the plastic. Chambers were then wrapped in 0.20 mm thick black Visquene, submerged, and left in the river to incubate overnight. The next morning chambers were unwrapped and one edge of the lid was lifted to take an oxygen measurement while stirring the probe. Chambers were resealed and left in the light for 1 h, and oxygen concentration was measured again. In pilot experiments, photosynthesis for longer than 1 h caused supersaturation of the water with oxygen. The volume of water in the chamber was calculated by subtracting the volume of the rock, measured by displacement, and the exact time of each oxygen reading was noted.

Epiphytes and detritus

At the end of the experiment in 1991, several filaments from each enclosure were preserved with formalin, and then epiphyte cells were counted on twenty 1 mm lengths of *Cladophora* with a light microscope at ×100. Diatom cell dimensions were measured with an ocular micrometer using *c.* twenty-five cells of each taxa from each treatment. Volumes were calculated using geometric formulae for shapes that most closely approximated the taxa (Kovala & Larrance, 1966). Percentage cover of detritus and silt on each 1 mm transect of *Cladophora* was estimated by noting the presence or absence of detritus at 0.1 mm intervals along a grid micrometer. Detritus included empty gelatinous stalks of diatoms like *Gomphonema*, *Rhoicosphenia* and *Cymbella*.

Benthic macroinvertebrates

Invertebrates colonized the enclosures through the mesh windows or by oviposition from above. At the end of each experiment, just prior to taking productivity measurements, macroinvertebrates were removed
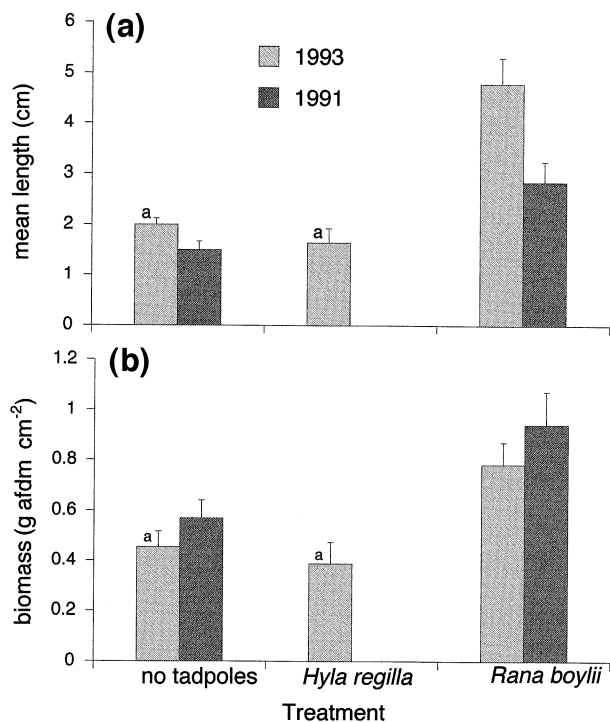
Fig. 1 Mean (± 1 SE) *Cladophora* length (a) and total periphyton mass (b) on cobbles grazed by invertebrates and tadpoles for 4 weeks in 1991 and 6 weeks in 1993. Treatment means with the same letter are not significantly different from each other ($P > 0.05$).

from cobbles, accumulated on a 0.1 mm mesh sieve and preserved in 70% ethanol. Macroinvertebrates were sorted and counted at ×10–70 magnification. Insects were identified to family or genus, and other invertebrates to class. For 1993 samples, insect head capsule width was measured to the nearest 0.1 mm with an ocular micrometer. Published regressions between head width and body mass (Smock, 1980) were used to estimate mass of each order of insects, except Hemiptera which lack good regressions. Predatory insects, mostly Hemiptera (*Ambrysus* sp.), are therefore reported as number per enclosure.

Nutrients

Effects of tadpoles on nitrogen cycling were assessed in two ways. In 1991, water samples were collected from each flow-through enclosure and the open river. No evidence of tadpole excretion (measurable levels of ammonium) was found, so tadpole excretion rates were measured using *in situ* incubations. On 27 August 1993, tadpoles from three *Rana* replicates and three

Hyla replicates were placed in sealed 500 ml polyethylene jars with filtered river water for 30 min ($n = 6$). Open river water was also collected. The average of five samples was used to compute background nitrogen concentration, $[N]_{ambient\ mean}$. Within 1 h of collection, water samples were vacuum filtered through Whatman GF/A glass microfibre filters and then through Gelman Metricel GN-6 0.45 μm filters to remove algal cells and particulates. Water samples were stored frozen in acid-washed Nalgene bottles. Colorimetric analyses (two replicates per sample) determined N concentration as ammonium (phenate method; American Public Health Association, 1985) and nitrate (hydrazine reduction method; Kellar, Paulson & Paulson, 1980). Absorbance was measured on a Bausch and Lomb spectronic 21 spectrophotometer. Rate of nitrogen excretion (μg N (tadpole h)$^{-1}$) was calculated as: $([N]_{sample} - [N]_{ambient\ mean})$ (5 tadpoles 0·5 h 0·5 l)$^{-1}$.

Statistical analyses

Unpaired *t*-tests were used to analyse algal and detrital response to *Rana* in 1991. Data were inspected for normality and homogeneity of variances. To meet these criteria, percentage coverage of detritus on *Cladophora* filaments was arcsin square root transformed. If transformation of a variable was unsatisfactory, Mann–Whitney *U*-tests were used. MANOVA tested *Rana* effects on the five most abundant groups of benthic macroinvertebrates. Macroinvertebrate numbers were natural log transformed to achieve homogeneity of variances. One-way ANOVAs tested *Rana*, *Hyla*, and block effects on algae in 1993 when normality and homogeneity of variance assumptions were met. Otherwise Kruskal–Wallis tests were used. MANOVA tested *Rana*, *Hyla*, and block effects on macroinvertebrate biomass. Experiment-wise error rates were maintained at α = 0.05 using the Bonferroni technique.

Path analysis, a multiple regression technique (Sokal & Rohlf, 1981; Hayduk, 1987), tested whether the observed correlations among tadpole grazing, primary production and macroinvertebrate abundances were best predicted by models of indirect or direct effects. Alternative models of algal–grazer interactions were compared by examining predicted correlation matrices among exogenous (i.e. manipulated) and endogenous variables. In 1991 there was one exogenous variable, presence *v* absence of *Rana*. In 1993 two exogenous

Table 1 Effects of tadpole grazing on epiphytic periphyton on *Cladophora* enclosed in 1 mm mesh cages for 4 weeks in 1991 and 6 weeks in 1993. For the 1991 tests, Bonferroni-adjusted α = 0.01

| | | Treatment | | | | | | | |
| | | No tadpoles | | *Hyla regilla* | | *Rana boylii* | | | |
Year	Response variable	Mean	SE	Mean	SE	Mean	SE	Test statistic	*P*
1991	Detritus on *Cladophora* (arcsin √%cover)	63.1	5.7			8.2	2.1	$t = 8.8$	<0.001
	Stalked diatoms (no. mm^{-1})	19.2	2.6			26.3	4.6	$t = 1.3$	0.212
	Adnate diatoms (no. mm^{-1})	5.04	1.0			1.26	0.5	$t = -3.3$	0.007
	Total diatom volume (μm^3 mm^{-1})	180,238	18,353			78,526	13,343	$t = 4.5$	<0.001
	Ash : biomass (ln(ash : biomass))	2.3	0.1			1.3	0.2	$t = 3.5$	0.004
1993	Ash : biomass (ln(ash : biomass))	3.4	0.5	3.1	0.8	2.3	0.4	$F(\text{trmt})_{2,12} = 3.1$	0.08
								$F(\text{block})_{6,12} = 3.9$	0.02

variables were used, the presence *v* absence of *Rana* and the presence *v* absence of *Hyla*. Predicted correlation matrices were compared with observed matrices using maximum likelihood goodness-of-fit χ^2 tests (appendix I in Wootton, 1994).

All statistical analyses were conducted using SYS-TAT (1992).

Results

Algae

Tadpole grazing enhanced algal standing stocks but only in the case of *Rana boylii* (Fig. 1). After 4 weeks of grazing in 1991, algal AFDM on cobbles in *Rana* treatments was 65% greater (Mann–Whitney $U_1 = 6.2$, $P = 0.01$) and *Cladophora* turfs were 81% longer than on control cobbles ($t = 3.0$, $P = 0.01$). After 6 weeks of grazing in 1993, *Rana* increased AFDM 72% ($F(\text{treatment})_{2,12} = 7.1$, $P < 0.01$; $F(\text{block})_{6,12} = 1.0$, $P = 0.46$) and length 139% ($F(\text{treatment})_{2,12} = 42.4$, $P < 0.0001$; $F(\text{block})_{6,12} = 3.1$, $P = 0.04$). Differences in AFDM and length between *Hyla* and control replicates were not significant.

Rana grazing resulted in significant differences in the assemblage of epiphytes growing on *Cladophora* and the amount of detritus covering the filaments (Table 1). *Rana* reduced the numbers of adnate diatoms, mostly *Epithemia* and *Rhopalodia*. Stalked diatoms, prin-
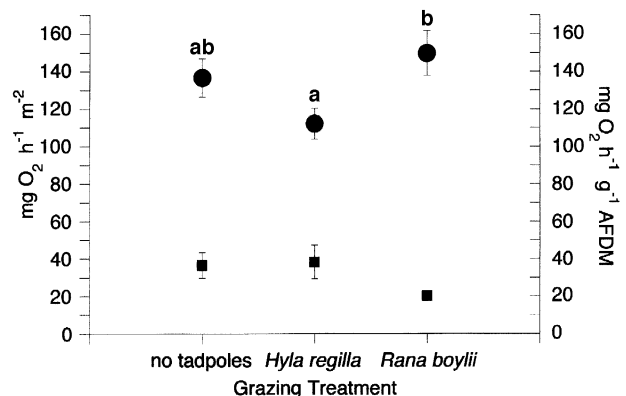
Fig. 2 Mean (± 1 SE) net primary productivity (NPP) as measured by area-specific (●) and mass-specific (■) oxygen liberation. a and b indicate significance of *R. boylii v H. regilla*, $P = 0.02$.

cipally *Gomphonema* sp. and *Rhoicosphenia* sp., were not significantly different between treatments, however. Total biovolume of diatoms per mm of *Cladophora* was 56% lower with *Rana* grazing. There was 7.7 times more surface area covered by detritus and silt on *Cladophora* filaments in tadpole exclosures than in tadpole enclosures. In 1991, *Rana* significantly decreased ash : biomass ratio, a correlate of diatom abundance. In 1993, *Rana* replicates again had the lowest ash : biomass ratio. *Hyla* replicates had intermediate values and controls the highest, but these differences were not significant and were outweighed by block effects.
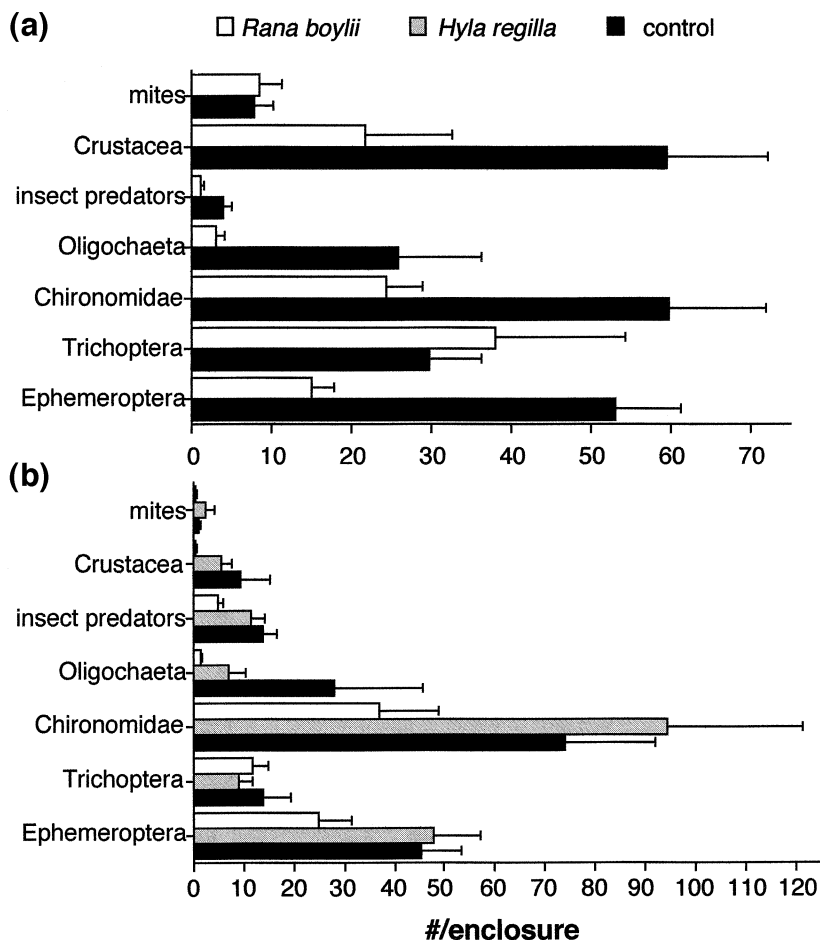
(a)

□ *Rana boylii*　▨ *Hyla regilla*　■ control

(b)

#/enclosure

Fig. 3 Mean abundance of macroinvertebrates (± 1 SE) colonizing enclosures in (a) 1991 and (b) 1993. Tests of treatment effects are given in Tables 2 and 3.

Treatment effects on production, as measured by oxygen liberation, mirrored the impacts on *Cladophora* and diatom standing stock (Fig. 2). On an areal basis, productivity was 10% higher on cobbles grazed by *Rana* than on those grazed by *Hyla*, reflecting the overall increase in biomass (ANOVA of ln(area-specific NPP): $F(\text{treatment})_{2,12} = 5.1$, $P = 0.03$; $F(\text{block})_{6,12} = 2.4$, $P = 0.09$; *R. boylii v H. regilla*, $P = 0.02$). On a per gram basis, however, net primary productivity was reduced by *Rana*, reflecting the decreased diatom abundance. The overall treatment effect was significant (ANOVA of ln(mass-specific NPP): $F(\text{treatment})_{2,12} = 3.8$, $P = 0.05$; $F(\text{block})_{6,12} = 3.8$, $P = 0.4$), but after adjusting α for experiment-wise error rate, the *post-hoc* comparisons were not significant (control *v R. boylii*, $P = 0.08$; *H. regilla v R. boylii*, $P = 0.08$).

Invertebrates

Total abundance of macroinvertebrate consumers was lower in the presence of *Rana* than *Hyla* tadpoles or controls in both years (Fig. 3a). In 1991, three of the five most common taxonomic groups, Ephemeroptera (mostly *Centroptilum* sp., a baetid mayfly), Diptera (mostly chironomid midges), and oligochaete worms associated with midge tufts, were significantly less abundant in the presence of *Rana* (Table 2). Treatment effects on Trichoptera (dominated by *Mystacides* sp., a leptocerid caddisfly) and Crustacea (amphipods, ostrocods, cladocerans, and copepods) were not significant. In 1993, treatment effects on benthic invertebrates were qualitatively similar (Table 3). Abundances of all groupings of common taxa excluding Trichoptera were lowest in the presence of *Rana*, but the highest abundances were not consistently in controls (Fig. 3b). *Hyla* replicates also had high numbers of chironomids, Ephemeroptera and predatory insects. Predators included gomphid dragonfly nymphs, megalopteran larvae (*Sialis* sp.), and naucorid bugs (*Ambrysus* sp.).

Three nested scenarios of mechanisms by which tadpoles may influence primary and secondary

Table 2 One-way MANOVA for the 1991 data set concerning the response of benthic invertebrates to grazing by *Rana boylii* tadpoles. The five most numerically abundant groups of taxa were analysed. For the five univariate tests, Bonferroni-adjusted α = 0.01

Tests	Source	Wilk's λ	F	df	P
Multivariate	Treatment	0.1	14.4	5,8	0.0008
Univariate	Ephemeroptera (ln(no. encl.$^{-1}$))		10.6	1,12	0.007
	Trichoptera (ln(no. encl.$^{-1}$))		0.2	1,12	0.705
	Chironomidae (ln(no. encl.$^{-1}$))		16.0	1,12	0.002
	Oligochaeta (ln(no. encl.$^{-1}$))		11.0	1,12	0.006
	Amphipoda, Cladocera, Copepoda, Ostrocoda (no. encl.$^{-1}$)		3.1	1,12	0.106

Table 3 One-way MANOVA for the 1993 data set concerning the response of benthic invertebrates to grazing by *Rana boylii*, *Hyla regilla*, or no tadpoles. For multivariate contrasts Bonferroni adjusted α = 0.025. For univariate tests and contrasts, Bonferroni-adjusted α = 0.005

	Source	Wilk's l	F	df	P
Multivariate test	Treatment	0.14	2.4	10,14	0.06
	Block	0.14	0.9	30,30	0.89
Contrasts	*Rana boylii v* control	0.28	3.7	5,7	0.06
	Hyla regilla v control	0.36	2.5	5,7	0.14
Univariate tests					
Ephemeroptera (ln(mg encl.$^{-1}$))	Treatment		2.4	2,11	0.14
	Block		1.4	6,11	0.30
Trichoptera (ln(mg encl.$^{-1}$))	Treatment		1.2	2,11	0.35
	Block		0.6	6,11	0.72
Chironomidae (ln(mg encl.$^{-1}$))	Treatment		4.6	2,11	0.04
	Block		3.0	6,11	0.06
Oligochaeta (ln(no. encl.$^{-1}$))	Treatment		2.4	2,11	0.14
	Block		1.6	6,11	0.25
Predators (no. encl.$^{-1}$)	Treatment		5.4	2,11	0.02
	Block		2.4	6,11	0.10

Contrasts	Ephemeroptera	Trichoptera	Chironomidae	Oligochaeta	Predators
R. boylii v control	0.05	0.47	0.05	0.06	0.01
H. regilla v control	0.25	0.16	0.64	0.13	0.48

productivity were compared using path analysis (Fig. 4). The first hypothesis is one of exploitative competition between tadpoles and insect grazers and subsequent release of *Cladophora* from the negative effects of epiphytes. In the path diagrams (Fig. 4) the hypothesized cause-and-effect relationships are indicated by one-headed arrows. The diagram for hypothesis 1 (H_1) implies that tadpoles determine diatom abundance, and that diatom density determines the amount of *Cladophora*, and the abundance of grazers and predators. The second hypothesis (H_2) includes H_1 as well as the direct effects of tadpoles as consumers of macroalgae.
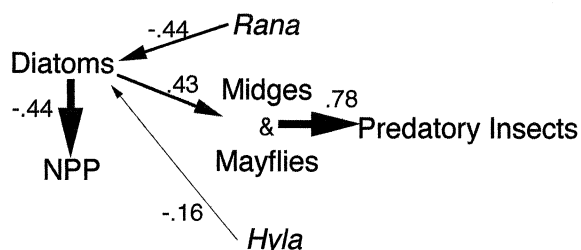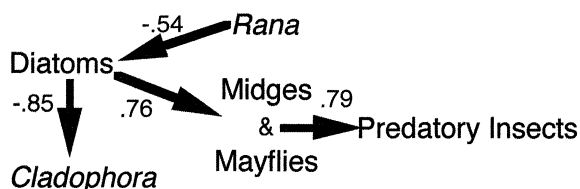
Hypothesis 3 (H_3) includes H_2 plus direct interference of tadpoles with invertebrates. The predicted correlation between any two variables is the sum of the direct path and the products of path coefficients associated with the indirect pathways. For example, the $r_{predicted}$ between *Rana* and grazers under H_3 was:

$$-0.63 \text{ (direct path)} + [-0.54 \text{ (\textit{Rana} to diatoms)} \times 0.42$$
$$\text{(diatoms to midges and mayflies)}] =$$
$$-0.857 \; v \; r_{observed} = -0.86$$

The models incorporating interference competition (shaded scenarios in Fig. 4) produced the best matches between observed (Tables 4 and 5) and
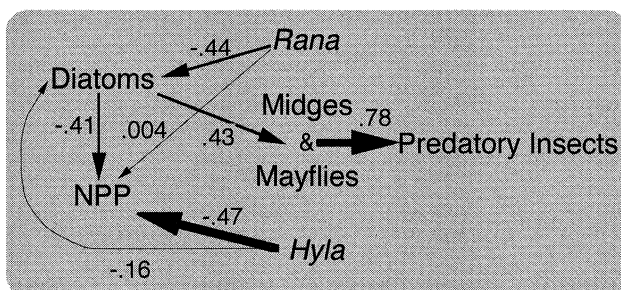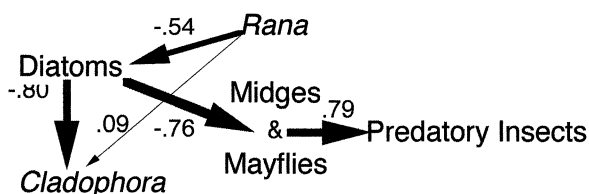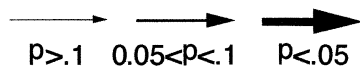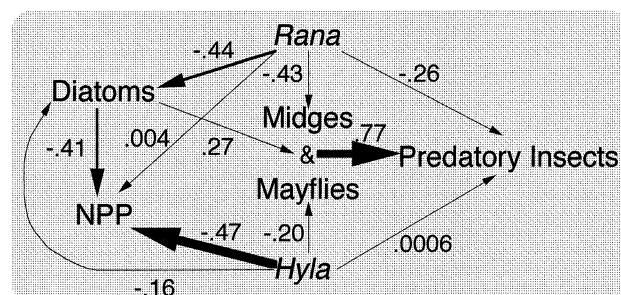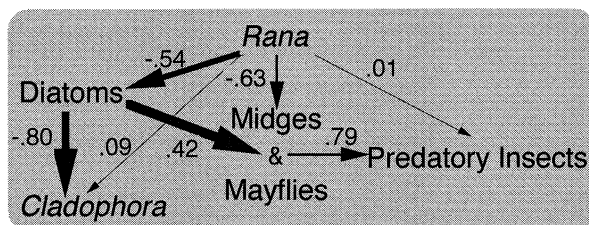
Fig. 4 Path diagrams of interactions among variables in grazing experiments in (a) 1991 and (b) 1993. Path coefficients are adjacent to arrows. Arrow thickness indicates level of statistical significance of the coefficients. Shading indicates which models predict correlations among variables that are not significantly different from the observed correlations in Tables 4 and 5. Dark shading indicates $P > 0.5$, and light shading indicates $P > 0.05$. For 1993, H_2 is not statistically better fit than H_3 (restricted $\chi^2 = \chi^2(H_2) - \chi^2(H_3) = 5.1$, df = 2, $0.1 < P < 0.5$).

predicted correlation matrices. In 1991, H_3 was the best fit model: $\chi^2(H_1) = 17.4$, df = 6, $P < 0.01$; $\chi^2(H_2) = 17.5$, df = 5, $P < 0.005$; $\chi^2(H_3) = 2.5$, df = 3, $P > 0.5$. In 1993, H_2 and H_3 produced good fits, indicating that both exploitation and interference

were important: $\chi^2(H_1) = 10.2$, df = 8, $P < 0.025$; $\chi^2(H_2) = 10.2$, df = 6, $P > 0.1$; $\chi^2(H_3) = 5.1$, df = 2, $P > 0.05$. H_2 is not statistically better fit than H_3 (restricted $\chi^2 = \chi^2(H_2) - \chi^2(H_3) = 5.1$, df = $2.1 < P < 0.5$).

Table 4 Observed correlations and descriptive statistics of variables used in the path analysis of the 1991 grazing manipulation (n = 14 enclosures)

	R	D	C	G	P
Rana boylii (R) (absence = 0, presence = 1)	1				
Diatom abundance (D) (ln(ash : biomass))	−0.54	1			
Area-specific *Cladophora* production (C) (ln(mg AFDM cm^{-2}))	0.53	−0.85	1		
Grazer abundance (G) (no. of midges and mayflies encl.$^{-1}$)	−0.86	0.76	−0.70	1	
Predatory insect abundance (P) (no. of odonates, megalopterans, and hemipterans encl.$^{-1}$)	−0.67	0.49	−0.52	0.79	1

Table 5 Observed correlations of variables used in the path analysis of the 1993 grazing manipulation (n = 21 enclosures)

	H	R	D	NPP	G	P
Hyla regilla (H) (absence = 0, presence = 1)	1					
Rana boylii (R) (absence = 0, presence = 1)	−1					
Diatom abundance (D) (ln(ash : biomass))	0.04	0.39	1			
Area-specific net primary productivity (NPP) (ln(mg O$_2$ h^{-1} m^{-2}))	−0.59	0.36	−0.51	1		
Grazer mass (G) (mg of midges and mayflies encl.$^{-1}$)	0.04	−0.43	0.43	−0.09	1	
Predatory insect abundance (P) (no. of odonates, megalopterans, and hemipterans encl.$^{-1}$)	0.17	−0.55	0.40	−0.12	0.78	1

Fish

In 1993 fish fry differentially colonized enclosures without tadpoles. Densities were per enclosure 1.67 ± 0.49 in control enclosures, 0.29 ± 0.29 in *Hyla* enclosures, and zero in *Rana* enclosures (Kruskal–Wallis = 13.7, df = 2, P =0.001). The California roach (*Hesperoleucas symmetricus* Baird and Girard) and three-spine stickleback (*Gasterosteus aculeatus* L.) were ≈ 1–2 cm standard length, too large to swim through the 1 mm^2 mesh. They probably entered as smaller fry and outgrew the mesh size.

Nutrients

Tadpole excretion did not elevate ambient levels of nitrogen in the flow-through enclosures (Fig. 5a, $F_{2,13}$ = 0.4, P = 0.7), despite tadpoles of both species excreting similar (t = 0.44, P = 0.68) amounts of N, i.e. 15–20 μg h^{-1} (Fig. 5b).

Discussion

Compensation response of macroalgae to tadpole grazing

The results of this study indicate the occurrence of biomass compensation by *Cladophora* in response to tadpole grazing on it and its epiphytes. *Rana boylii*

tadpoles decreased the abundance of diatoms and detritus on *Cladophora*, but this grazing activity resulted in an increase in total periphyton biomass and area-specific primary productivity on cobbles. *Hyla regilla* did not significantly reduce *Cladophora* abundance relative to controls, which may indicate some degree of *Cladophora* growth, at least enough to balance its depletion by grazing. In contrast, several other tadpole grazing experiments, conducted within ranges of natural densities, have indicated that tadpoles reduce periphyton standing crop (Dickman, 1968; Morin, Lawler & Johnson, 1988; Brönmark, Rundle & Erlandsson, 1991; Liebold & Wilbur, 1992). The experiments conducted by Brönmark *et al.* (1991), in enclosures similar to those in the present study, explicitly tested the effects of tadpole density. Low density (five *R. temporaria* tadpoles per bucket) caused more than 60% reduction of periphyton dry weight compared with no-tadpole controls, and high density (ten per bucket) caused ≈ 85% reduction. When tadpole grazing has been reported to increase total producer biomass, it was the result of increasing the abundance of relatively inedible macrophytes and macroalgae (Werner, 1994; Kupferberg, 1997).

The effect of *Rana boylii*, where growth of relatively inedible algae resulted in an increase of total periphyton biomass, was contrary to expectations. For algae, trade-offs between grazing-resistant structures and
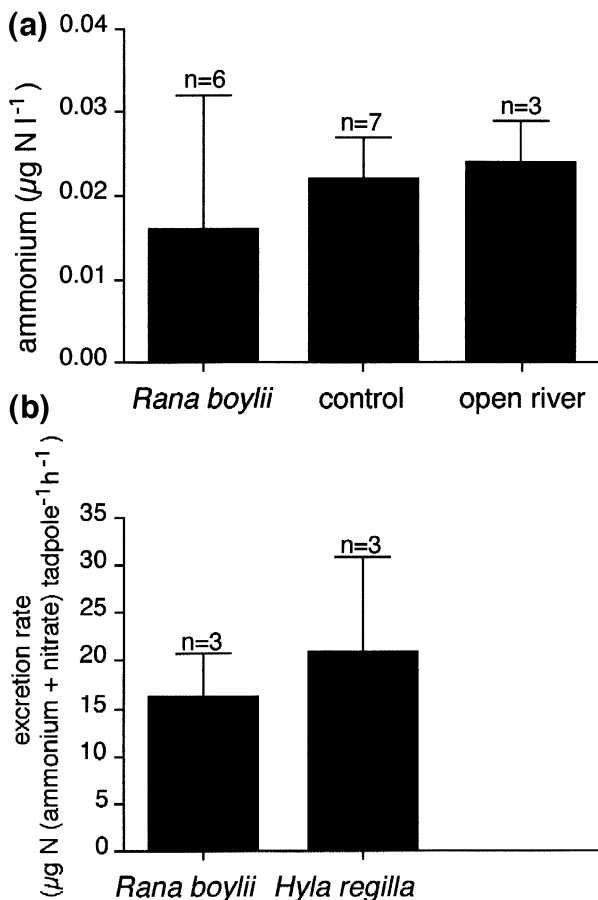
(a)

(b)

Fig. 5 (a) Ammonium concentrations (± 1 SE) in the open river, tadpole enclosures, and exclosures. (b) Rate of total nitrogen (ammonium and nitrate) excretion during closed incubations. Open river total N = 6.1 ± 0.5 µg l⁻¹.

competitive ability of algae have been shown to cause inedible taxa to increase in abundance but not to increase total periphyton biomass (Rosemond, Mulholland & Elwood, 1993). In streams, the benefits of grazers to uneaten periphyton have included increased chlorophyll-specific or AFDM-specific productivity (Hunter, 1980; Lamberti & Resh, 1983; Lamberti *et al.*, 1987; Hill & Harvey, 1990) rather than biomass compensation. The mechanisms described below may have operated simultaneously to produce this unexpected result.

Cladophora enhancement via epiphyte removal. Grazer removal of epiphytes generally has positive effects on host plants. Although the ability of grazers to mitigate epiphyte effects may vary with seasonal changes in temperature and nutrients (Neckles, Wetzel & Orth, 1993), there is much evidence that epiphytes compete

asymmetrically with many host taxa (Shacklock & Doyle, 1983; Orth & van Montfrans, 1984; D'Antonio, 1985; Dodds, 1991; Underwood, Thomas & Baker, 1992). The hypothesis that *Rana* benefits *Cladophora* by epiphyte removal is supported by observations that tadpoles preferentially graze on epiphytes. In patch-choice experiments, *Rana* chose to forage in patches of *Cladophora* with epiphytes rather than on the host alga alone or on taxa of filamentous green algae not supporting epiphytes (Kupferberg, 1996a). Similarly, some snails prefer epiphytes over *Cladophora* (Underwood & Thomas, 1990; Brönmark *et al.*, 1991). Grazer preference for epiphytes over hosts illustrates the mutualistic relationship between *Cladophora* and some grazers (Dodds, 1991; Dodds & Gudder, 1992). Grazers remove epiphytes releasing *Cladophora* from competition. As *Cladophora* increases in length, more substrate for epiphyte attachment becomes available, benefiting the grazers.

Why was total periphyton biomass not increased by *Hyla*, even though these tadpoles also consume diatoms when enclosed with cobbles supporting *Cladophora* (Kupferberg *et al.*, 1994)? Mouthpart morphology and size differ between the two tadpoles. *Rana* has six rows of keratinized 'teeth' above its mouth and seven rows below, and these may make them very efficient scrapers. *Hyla* has only two and three rows of 'teeth' above and below its mouth, respectively. *Hyla* are smaller and thus may process smaller volumes of material, remove sediment and diatoms with lower efficiency, and have shorter gut passage time allowing more diatoms to survive ingestion. Even though replicates were stocked at relevant natural densities with equal numbers of tadpoles, tadpole biomass was greater in *Rana* replicates. *Hyla* at higher densities might have caused an effect. For loricariid catfish, the beneficial effect of sediment removal on algal growth depended both on grazer density and the size class of the fish (Power, 1990a). Only large fish were able to clear enough area to facilitate primary production in places where rates of sediment accumulation on the river bed were high.

Nutrient regeneration. Cladophora, which is strongly nitrogen limited in the Eel River (Hill & Knight, 1988; Power, 1991), may have been stimulated by the nitrogen tadpoles mineralized from diatoms and detritus. In ponds and rock pools, tadpoles can influence nitrogen concentrations (Seale, 1980; Osborne &

McLachlan, 1985). In closed containers, investigations of macrophyte–epiphyte systems have shown that snail grazing can facilitate macrophyte growth, in part by nutrient regeneration (Kairesalo & Koskimies, 1987; Underwood, 1991). I could not detect changes in ambient concentrations of nitrogen, possibly due to dilution or rapid uptake by nutrient-limited algae and bacteria. The shift observed in epiphyte composition away from *Epithemia* may support the tadpole nutrient cycling hypothesis, however. *Epithemia*, which contains nitrogen-fixing endosymbionts (Dodds, 1991), may lose its competitive advantage over stalked diatoms if nitrogen becomes more available in the presence of tadpoles. Nutrient addition experiments in the South Fork Eel River resulted in similar decreases in the relative abundance of *Epithemia* (Marks, 1995).

Release of Cladophora from midge damage. Chironomid midge larvae have been shown to dramatically reduce standing crops of *Cladophora* when they are released from predation in the Eel River (Power, 1990b). If midges are more effective at limiting *Cladophora* than tadpoles, then tadpole reduction of midge numbers could lead to an increase in *Cladophora* abundance. This might explain the differences between *Rana* and *Hyla* effects, because *Hyla* did not reduce midge abundance.

Tadpole effects on benthic assemblages

As the path analysis (Fig. 4) indicates, the models best explaining the effects of *Rana* tadpoles on the benthos incorporate both exploitation and interference as mechanisms. For example, exploitation of detritus and diatoms by *Rana* directly decreased food for other primary consumers. The grazer taxa that were lower in abundance (larvae of the mayfly, *Centroptilum* sp., chironomid larvae and the oligochaete worms associated with them) are collector–gatherers of diatoms and detritus (Cattaneo, 1983; Merrit & Cummins, 1984). Competition between tadpoles and insects for periphyton has also been shown in experimental cattle tanks (Morin *et al.*, 1988; Morin, Lawler & Johnson, 1990). Periphyton colonizing glass slides was more abundant in the presence of *Hyla andersonii* tadpoles alone than in the presence of both *Hyla* and aquatic insects (Morin *et al.*, 1990). In my experiments, the effects of competition for periphyton and detritus were also manifest at higher trophic levels, with fewer

invertebrate predators and fish fry colonizing *Rana* enclosures than controls.

The specific mechanisms of interference could include tadpole inhibition of insect oviposition (Petranka & Fakhoury, 1991) and consumption of insect larvae (Jenkins & Kitching, 1990). Faecal pellets from the present experiment, and wild caught tadpoles, provided no evidence (insect head capsules or other sclerotized body parts) of predation, however. Behavioural interference, such as mechanical disruption of invertebrate foraging by tadpoles, was more likely. As shown with a snail, *Physella gyrina* Say (Gresens, 1995), the presence of a large grazer can cause larval chironomids to decrease activity even without direct physical contact.

Appreciation of functional differences between species (*Rana boylii v Hyla regilla*) is relevant to conservation. As amphibians decline (Wake, 1991), we rarely know whether sensitive species, such as *R. boylii*, which is listed as a species of special concern in California (Jennings & Hayes, 1994), are strong interactors. As demonstrated here, *R. boylii* has unique effects on the complex indirect interactions between algae and grazers, and can influence primary productivity with consequences for higher trophic levels.

Acknowledgments

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