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The effects of nymphaeid (*Nuphar lutea*) density and predation by perch (*Perca fluviatilis*) on the zooplankton communities in a shallow lake

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SUMMARY

1. The effects of addition of juvenile perch (*Perca fluviatilis*) on the microcrustacean and rotifer communities associated with nymphaeid beds were studied, at three different plant densities [high (normal), medium (reduced by a half) and low (reduced to a third of normal)], in eighteen 2 m × 1 m enclosures in a shallow lake.
2. At the low and medium densities of lilies, *Daphnia* densities were high in the absence of perch but very low in the presence of perch. They increased, even in the presence of perch, to high densities (comparable with those in the absence of perch) at the highest plant density. Body sizes of *Daphnia hyalina* were consistent with high predation by perch at low and medium plant densities but reduced predation at high plant densities. Patterns of chlorophyll *a* concentration, in the presence of perch, inversely reflected those of *D. hyalina* density.
3. At naturally high densities but not at reduced densities, the plants appeared to act as refuges against predation for the *Daphnia*. Reductions in oxygen concentrations in the plant beds were not responsible for the refuge effect, nor could there be avoidance of the beds by the fish. The mechanism of the refuge effect must therefore lie in frustration of the process of capture of the *Daphnia* by the fish.
4. Numbers of other small Crustacea and rotifers were mostly unaffected by fish predation. Numbers of *Asplanchna* sp., *Chydorus sphaericus* and copepodites were higher in the presence of fish and, although there was no main effect of fish on numbers of *Diaphanosoma brachyurum*, there was a plant–fish interaction, with this species being less abundant in the presence of fish at low plant densities but more abundant in the presence of fish at medium and high plant densities. Main effects of plants were few, with only *Asplanchna* sp. and *Keratella* sp. decreasing in numbers with increasing plant density. Most taxa changed in numbers with time but interaction effects between time and plants, and fish and plants, were few.

Introduction

Nutrient supply and predation are among the more important agents in structuring biological communities in freshwaters (Hall, Cooper & Werner, 1970). In shallow, plant-dominated waters, nutrient supplies are often high (Sas, 1989). The open waters of such lakes, however, may be clear in summer and phytoplankton crops very small in relation to the growth potential set by the availability of nutrients (Jeppesen *et al.*, 1991). This may be because of the operation of factors such as progressive depletion of nitrogen by plant uptake, or denitrification (Ozimek, Gulati & van Donk, 1990), or allelopathic inhibition of algal growth by secretions from the plants (Hootsmans & Blindow, 1994). It may also be because the plants provide an environment in which potential grazers on the open water algae can find refuges against fish predation. Timms & Moss (1984) demonstrated that beds of nymphaeids, in a shallow lake in Norfolk, U.K., harboured large numbers of cladocerans, some of which appeared in the open water at night. The water had high algal growth potential and there was free access to fish from the lake and from interconnected lakes and a river system. Yet phytoplankton chlorophyll *a* concentrations were lower than 5 μg L^{-1} in the summer period when the biomass of nymphaeids was high.

There was circumstantial evidence that the nymphaeids were allowing large grazer cladoceran populations to persist. A linked lake, lacking plants, had few Cladocera, which were of smaller body size. Numbers and body sizes increased when the community was protected from fish predation in large polyethylene bags suspended in the lake. However, it was not clear how such a refuge mechanism might operate, for plant beds do not necessarily exclude fish and may indeed harbour concentrations of small zooplanktivorous fish against their own predators. Perch (*Perca fluviatilis* L.), compared with roach (*Rutilus rutilus* L.), hunt Cladocera effectively in weed beds (Winfield, 1986). One suggestion is that the refuge is, in fact, most effective for young zooplanktivorous fish, which are kept from the open water by the presence there of piscivores and hence are limited in their ability to feed on the open-water zooplankters (Venugopal & Winfield, 1993). Studies in enclosures in Little Mere, Cheshire (Beklioglu & Moss, 1996) have suggested that submerged plants (*Potamogeton berchtoldii* L.) provide some refuge for zooplankters against predation but that it is not so effective as that shown for nymphaeids by Timms & Moss (1984).

There is also a complex mixture of potential phytoplankton grazers in and around plant beds. It includes largely open-water genera like *Daphnia* and *Bosmina*, loosely plant-associated genera like *Simocephalus*, *Sida* and *Eurycercus*, and firmly associated genera like *Graptoleberis* and *Pleuroxus* (Irvine, Balls & Moss, 1990). It is not clear what the relative roles of these organisms may be in supporting macrophyte dominance in shallow lakes. There is clearly much scope for field experimental work in elucidating exactly which mechanisms do operate to maintain clear water. This paper describes such an experiment in which the effects of perch predation on the associated crustacean community were studied at different densities of a nymphaeid, *Nuphar lutea* L. in enclosures in a shallow lake (Little Mere, Cheshire, U.K.). The hypothesis was that predation on the zooplankton community by perch would be increased with decreasing density of nymphaeids.

Methods

Eighteen experimental enclosures, each 1 × 2 m, were built in a uniform bed of *Nuphar lutea* in Little Mere (53°20′N, 2°24′W, maximum depth 1.7 m, mean depth 0.7 m, area 2.5 ha), which was still a very fertile lake when the experiment was carried out in 1993, despite diversion of poor quality sewage effluent from it in June 1991. The enclosures were of curtain netting (mesh size < 50 μm) fixed on to a wooden framework, pushed into the sediment in about 75 cm of water. The netting allowed exchange of water but not of animals, nor, because it clogged with periphyton, of planktonic algae.

Three densities of plants were established (Table 1) by leaving the stand intact in six enclosures (coded H for high), cutting about half of the leaves in a further six (M for medium) and cutting between three-quarters and two-thirds of the leaves in the remaining six (L for low). The remaining plants covered about 90%, 50% and 25% of the water surface, respectively. The surviving, floating leaves were marked with a waterproof marker and we cut any new floating leaves, as they were initiated, during the experiment, which ran from 10 June until 8 August 1993. Submerged leaves were allowed to grow but occupied only the lower part of the water column. Initial and final numbers of leaves are shown in Table 1. The intended differences in surface cover and petiole density in the upper part of the water column were maintained. Multiplication of submerged leaves led to a narrowing of the difference between some L and M treatments (Table 1) in terms of total leaf numbers, but the H treatment remained distinct throughout.

Each enclosure was initially electrofished until no more fish were recovered. Six perch, taken from the most abundant cohort in the population in the lake at the time, were then added to three enclosures in each plant treatment (Table 1). The remaining enclosures were left fish-free. Little Mere had only recently been invaded by fish from upstream,

Table 1 Density of *Nuphar lutea* (floating plus submerged leaves) per m^2 in the experimental enclosures at the start and end of the experiment, of chlorophyll *a* (μg L^{-1}) at the start of the experiment, and lengths (cm) and number of perch, *Perca fluviatilis*, measured (snout to fork) at the end of the experiment. Values are given + SD, *n* = 3

	Leaves			Perch	
Treatment	Start	End	Chlorophyll *a*	Number	Length
LC	10.2 ± 1.3	20.0 ± 6.0	10.1 ± 6.3	–	–
MC	17.3 ± 3.2	22.6 ± 0.8	7.8 ± 3.7	–	–
HC	31.6 ± 2.9	35.8 ± 4.1	4.6 ± 1.5	–	–
LF	10.5 ± 1.8	11.9 ± 3.9	10.0 ± 10.7	2.4 ± 0.8	14.8 ± 0.6
MF	15.2 ± 1.1	21.4 ± 1.8	10.9 ± 2.8	2.2 ± 0.3	14.9 ± 0.9
HF	30.5 ± 0.9	38.0 ± 6.5	4.8 ± 1.9	2.5 ± 0.5	14.8 ± 0.8

L, low; M, medium; H, high; C, without fish; F, with perch.

following diversion of the effluent. Perch strongly dominated the community, with relatively few roach, tench (*Tinca tinca* L.) and pike (*Esox lucius* L.). The density of fish in shallow lakes is very heterogeneous; three per m^2 in plant beds is a probable population when overall density of small fish in a lake may be from 2 to 6 m^{-2} (Schriver *et al*., 1995). However, densities fluctuate and to allow for this and to simulate some inevitable mortality of a free-living juvenile perch population during the summer, attempts were made, on 16 July, to remove two fish from each enclosure to which fish had been added. This could not be uniformly completed without serious damage to the vegetation but was randomly achieved among the enclosures. The final number of fish is shown in Table 1. There was no significant difference (one-way ANOVA) in number of fish among treatments to which fish had been added.

There were thus six treatments: low plant density without fish (LC), medium plant density without fish (MC), high plant density without fish (HC), low plant density with perch (LF), medium plant density with perch (MF) and high plant density with perch (HF), each replicated three times, with treatments randomly dispersed among the enclosures, which were close together but not abutting. The design was conditioned by the need to establish treatments among an initially uniform stand of nymphaeids, which was of limited area and irregular in shape in a lake almost entirely colonized by submerged or floating-leafed plants.

The water from within the enclosures was sampled initially and subsequently on four occasions (29 June, 12 July, 26 July and 8 August 1993), using a 4.5-cm diameter, closable tube, which was lowered to just above the bottom then stoppered with a rubber ball at the lower end before being retrieved full of water. Samples were filtered through Whatman GF/C filters and the phytoplankton chlorophyll *a* was extracted in acetone and determined spectrophotometrically. Zooplankton was sampled using the same tube with at least four tubefuls bulked to make a combined 5-L sample. Two such bulked samples were taken per enclosure on each sampling occasion and filtered through a 67-μm mesh net. The zooplankters were narcotized with chloroform then preserved in formalin and later identified and counted. Body size of *Daphnia hyalina* Leydig was measured from the top of the head to the base of the spine to the nearest 10 μm.

At the end of the experiment the plants were harvested and the leaves counted, the fish were measured then killed and their stomach contents preserved for examination. Temperature and dissolved oxygen were measured to ±0.5 °C and ±0.1 mg L^{-1} with a thermistor and oxygen probe, respectively, at 09.00, 15.00, 21.00 and 03.00 h BST on 29/30 July just below the surface and just above the bottom in each of the enclosures.

One-way ANOVA was used on data for numbers of the more abundant zooplankters and chlorophyll *a* concentrations from the initial sampling to determine whether the initial conditions were uniform. Treatment effects were thereafter analysed first by repeated measures ANOVA, followed by Tukey tests where the ANOVA had shown significant differences. Regression analyses using mean data were used to determine relationships between phytoplankton chlorophyll *a*, leaf number and *D. hyalina* density.

Results

Table 1 shows that the intended differentials in plant density were maintained throughout the experiment, except that growth in submerged leaves had muted the difference between LC and MC treatments by the end. Differentials in surface coverage and petiole density in the upper parts of the water columns were inspected visually every week and were maintained. There were no significant differences in temperature and oxygen concentrations in the various treatments when these were measured over a 24-h period, midway through the experiment. Mean surface temperatures were then 19.1 ± 1.7 °C, and mean bottom temperatures 17.5 ± 0.5 °C. Mean surface oxygen concentrations were 8.7 ± 1.7 mg L^{-1} and bottom concentrations 2.4 ± 1.5 mg L^{-1}.

Table 2 shows mean numbers of organisms sampled from the water column and mean concentrations of phytoplankton chlorophyll *a* initially and as means of the data obtained on four subsequent sampling dates from each of the three replicates. Standard deviations ($n = 3$) are shown. Ten species of Crustacea, four of rotifers and one copepod in adult form (*Diaptomus gracilis* Sars) were recorded, although there may have been more species of copepods present. There was considerable patchiness in the less abundant forms but strong patterns soon emerged in the numbers of *D. hyalina* and in chlorophyll *a* concentrations. Repeated measures ANOVA (Table 3) showed general effects of time on animal numbers but surprisingly few main effects of plant density and of the presence or absence of perch. Increasing plant density decreased the numbers of two rotifer species and tended to reduce the concentration of chlorophyll *a*, especially in the presence of fish. The presence of fish increased the densities of the small crustaceans, *Alona affinis* Leydig and *Chydorus sphaericus* (O.F. Mull), of the rotifer, *Asplanchna* sp. and of copepodites. It decreased the densities of *D. hyalina* but had no significant effect on the majority of taxa. It increased the chlorophyll *a* concentrations. There were significant plant–fish interaction effects for *Diaphanosoma brachyurum* Lieven, which was scarce but increased with plant density in the presence of fish, and for both chlorophyll *a* and *Daphnia hyalina.* These are analysed in Tables 4 and 5. Chlorophyll *a* concentrations were significantly reduced by both plant density and the absence of fish (Table 4). The effects of fish were shown at low and medium plant densities but not at high densities, where fish had no effect. Concentrations at low and medium plant densities in the absence of fish were not significantly different from those at high densities in either the presence or absence of fish. In all these cases they were relatively low compared with values at low and medium densities in the presence of fish. A complementary inverse pattern was found for densities of *D. hyalina.* Densities were much higher in low and medium plant densities in the absence of fish but this effect was not found at high plant density. Fish were unable to reduce *Daphnia* numbers significantly at high plant density. There was an apparent reduction in *Daphnia* density in both high plant density treatments compared with densities in the low and medium densities where fish were absent, but the Tukey tests failed to show a significant difference.

Investigation of fish gut contents at the end of the experiment suggested that the effects of fish on *Daphnia* densities were exerted through predation because *Daphnia* was a frequent component of the gut contents in the LF (2.6 + 7.5 *Daphnia* per gut) and MF (29.3 + 89) treatments but significantly less abundant (*t*-tests) in the HF treatment (0.7 + 1.2). Benthic organisms, *Eurycercus* and copepodites were also found in the guts but there were no significant differences among treatments. Measurements of *Daphnia* body size are also consistent with predation effects (Table 6). Size differentials were small but significant (two-way ANOVA) with body sizes smaller in the presence of fish than in their absence at the low and medium plant densities on three out of four sampling dates, but insignificantly different in the presence or absence of fish in the high plant density treatments.

Regression analysis suggests a close link between *Daphnia* numbers, plant density and phytoplankton chlorophyll *a* in the presence of fish but not in their absence (Table 7).

Discussion

The results of this experiment suggest that perch are able to reduce *Daphnia* numbers and body sizes through predation when densities of *Nuphar lutea* are artificially reduced but not at naturally high densities. In consequence, phytoplankton chlorophyll *a* concentrations were increased when *Daphnia* densities were reduced by fish predation. This supports the hypothesis that nymphaeids provide refuges for daphnids

Table 2 Mean densities of animals (per L) and concentrations of chlorophyll *a* (μg L^{-1}) at the start and during the operation of the treatments in an experiment set up in Little Mere, Cheshire. Initial values are based on the means of two samples and counts from each of three replicate enclosures. Treatment means are based on similar sampling on four subsequent sampling occasions. All standard deviations are based on n = 3 replicates

		Treatment					
Organism	Sampling	LC	MC	HC	LF	MF	HF
Alona affinis	Initial	0.8 ± 2.0	0.8 ± 2.0	0	0	0	0.7 ± 1.6
	Treatment	0	0.4 ± 0.8	0	1.2 ± 2.4	2.0 ± 4.0	2.1 ± 3.0
Asplanchna sp.	Initial	0	0	0.7 ± 1.6	0.8 ± 2.0	4.5 ± 2.4	0
	Treatment	2.1 ± 3.9	1.9 ± 2.5	0.5 ± 1.0	16.0 ± 21.0	12.8 ± 12.7	0.2 ± 0.4
Bosmina coregoni	Initial	0	0	0	0	0	0
	Treatment	4.6 ± 4.0	2.3 ± 3.1	0	8.1 ± 7.0	2.4 ± 3.0	0.4 ± 0.4
Brachionus sp.	Initial	0	0	0	0	0	0
	Treatment	0.5 ± 1.0	0.5 ± 1.0	0	2.6 ± 4.4	0.8 ± 1.5	0.3 ± 0.5
Ceriodaphnia sp.	Initial	0	0	0	0	0	0
	Treatment	2.1 ± 2.5	4.0 ± 3.8	3.3 ± 3.5	1.3 ± 2.3	1.6 ± 2.1	8.5 ± 12.4
Chydorus sphaericus	Initial	0	0	0	0	0	0
	Treatment	0.5 ± 0.4	0.3 ± 0.4	0.9 ± 1.3	5.3 ± 6.3	2.6 ± 2.1	0.6 ± 0.8
Copepodites	Initial	17.0 ± 11.6	10 ± 11.5	8.2 ± 6.8	9.2 ± 11.2	9.5 ± 2.0	13.8 ± 14.2
	Treatment	27 ± 17	32 ± 27	29 ± 20	83 ± 33	99 ± 107	27 ± 3
Daphnia hyalina	Initial	99.3 ± 57.6	191 ± 45.2	58.2 ± 14.8	158 ± 85.0	75.8 ± 41.8	120 ± 54.3
	Treatment	192 ± 84	140 ± 45	79 ± 29	7.2 ± 5.6	14.9 ± 7.7	80 ± 20
Diaphanosoma brachyurum	Initial	0.8 ± 2.0	0	0	0	0	0
	Treatment	1.0 ± 2.0	0.6 ± 1.3	4.2 ± 2.1	0.5 ± 0.9	35.5 ± 4.1	2.0 ± 3.5
Diaptomus gracilis	Initial	0	5.0 ± 12.2	0	0	0	0
	Treatment	2.0 ± 2.5	0.4 ± 0.8	0.2 ± 0.4	1.6 ± 2.4	3.2 ± 4.6	0.3 ± 0.5
Eurycercus lamellatus	Initial	0	0	0	0	0	0
	Treatment	0	0.2 ± 0.4	0	0.2 ± 0.4	0.2 ± 0.4	0.8 ± 1.5
Keratella sp. 1	Initial	0	2.5 ± 4.2	0.7 ± 1.6	1.7 ± 4.1	11 ± 13.9	1.2 ± 2.9
	Treatment	31 ± 25	18 ± 15	4.8 ± 3.0	274 ± 115	55 ± 48	3.6 ± 3.1
Keratella sp. 2	Initial	0	0	0	1.0 ± 2.4	0.5 ± 1.2	0
	Treatment	17 ± 30	4.5 ± 5.1	1.4 ± 2.6	40 ± 56	10 ± 15	1.6 ± 1.6
Nauplii	Initial	13.8 ± 12.3	6.7 ± 10.6	15.7 ± 8.2	14.7 ± 13.6	34.5 ± 30.8	15.2 ± 12.9
	Treatment	12 ± 11	18 ± 17	22 ± 24	25 ± 17	32 ± 28	17 ± 15
Pleuroxus sp.	Initial	0.8 ± 2.0	0.8 ± 2.0	0	0	0	0.7 ± 1.6
	Treatment	0.2 ± 0.4	0.3 ± 0.5	0	0.5 ± 0.9	0.6 ± 1.3	0.4 ± 0.4
Polyphemus pediculus	Initial	6.8 ± 6.3	58 ± 37.6	15.0 ± 14.8	12.0 ± 11.6	24.7 ± 31.0	26.7 ± 24.5
	Treatment	21 ± 11	13 ± 10	8.8 ± 15	20 ± 12	15 ± 19	11 ± 23
Scapholeberis mucronata	Initial	0	0	0	0	0	0
	Treatment	7.3 ± 7.6	10 ± 8.9	7.7 ± 8.3	3.2 ± 3.4	5.0 ± 3.6	7.1 ± 9.7
Chlorophyll *a*	Initial	10.1 ± 6.3	7.8 ± 3.7	4.6 ± 1.5	10.0 ± 10.7	10.9 ± 2.8	4.8 ± 1.9
	Treatment	15.3 ± 13.4	10.4 ± 9.8	12.1 ± 10.3	41.6 ± 17.0	35.5 ± 12.0	12.9 ± 10.6

L, low; M, medium; H, high densities of nymphaeids; C, control (without fish), F, with fish.

against fish predation and allow large densities to be maintained to the advantage of maintenance of a favourable light climate for the plants. Schriver *et al.* (1995) have also demonstrated that, except where the fish density is very high, fish predation has reduced effects on the zooplankton community in submerged macrophyte beds than in the open water.

Few effects were found for other crustaceans or rotifers. Neither plant density nor the presence or absence of fish influenced the numbers of most of the fifteen taxa named. This may mean that fish predation has little influence on the population dynamics of these organisms, whose numbers may be controlled by food supply or invertebrate predation. Many were small (rotifers, *Bosmina*, *Diaphanosoma*, *Pleuroxus*) and might be expected to escape visual predation by vertebrates. However, others were even larger than the vulnerable *D. hyalina*, for example *Polyphemus pediculus* (L.) and *Eurycercus lamellatus* (O.F.Mull) or occupy niches associated with the surface tension film (*Scapho-*

Table 3 Results of repeated measures analysis of variance of densities of Crustacea and rotifers and concentrations of phytoplankton chlorophyll *a* from an experiment set up in Little Mere, Cheshire. Values are probabilities (Huynh-frldt epsilon criterion); ns, $P > 0.05$

	Effects of:					
	Plant density	Presence of fish	Plant × fish	Time	Time × plant	Time × fish
Alona affinis	NS	< 0.05	NS	< 0.01	NS	< 0.05
Asplanchna sp.	< 0.05	< 0.01	NS	< 0.001	< 0.05	NS
Bosmina coregoni	NS	NS	NS	< 0.01	NS	NS
Brachionus sp.	NS	NS	NS	< 0.01	NS	NS
Ceriodaphnia sp.	NS	NS	NS	< 0.05	NS	NS
Chydorus sphaericus	NS	< 0.05	NS	< 0.05	NS	< 0.05
Copepodites	NS	< 0.05	NS	< 0.001	< 0.001	NS
Daphnia hyalina	NS	< 0.001	< 0.01	< 0.05	NS	< 0.05
Diaphanosoma brachyurum	NS	NS	< 0.01	NS	NS	NS
Diaptomus gracilis	NS	NS	NS	< 0.01	< 0.05	NS
Eurycercus lamellatus	NS	NS	NS	NS	NS	NS
Keratella sp. 1	< 0.05	NS	NS	< 0.001	< 0.05	NS
Keratella sp. 2	NS	NS	NS	< 0.05	NS	NS
Nauplii	NS	NS	NS	< 0.001	NS	NS
Pleuroxus sp.	NS	NS	NS	< 0.01	NS	NS
Polyphemus pediculus	NS	NS	NS	< 0.001	NS	NS
Scapholeberis mucronata	NS	NS	NS	< 0.001	NS	NS
Chlorophyll *a*	< 0.01	< 0.001	< 0.05	< 0.001	NS	NS

Table 4 Results of Tukey tests of effects of treatments on chlorophyll *a*. Mean values are given, with values underlined if not significantly different at $P = 0.05$ in the cases of main effects

Plants	Low	Medium	High			
	28.4 ± 19.9	23.0 ± 16.8	12.5 ± 9.7			
Fish	Present	Absent				
	30.0 ± 17.7	12.6 ± 10.4				
Plants × Fish						
	LC	LF	MC	MF	HC	HF
Mean	15.3 ± 13.4	41.6 ± 17	10.4 ± 9.8	35.5 ± 12	12.1 ± 10.3	12.9 ± 10.6
LC		< 0.01 NS	< 0.05	NS	NS	
LF			< 0.01	NS	< 0.01	< 0.01
MC				< 0.01	NS	NS
MF					< 0.01	< 0.05
HC						NS

leberis mucronata O.F.Mull), which would appear to make them potentially vulnerable. *Polyphemus* may be able to escape predation by rapid movement. It has a very large eye which may equip it to avoid attack even in the absence of vegetation. It was also not reduced by the presence of perch in experiments in polyethylene bags in Little Mere where there were no plants (Beklioglu & Moss, 1995). Rapid movement also appears to be advantageous for adult copepods but for *Scapholeberis* and *Eurycercus* it may be that even a reduced plant cover supplies sufficient refuge for escape from predation to be possible. This might also contribute to the invulnerability of some of the smaller taxa. Neither *Scapholeberis* nor *Polyphemus* was found in perch guts but occasional remains of the slower-moving *Eurycercus* were found. Many of these

Table 5 Results of Tukey tests of effects of treatments on *Daphnia hyalina*. Mean values are given, with values underlined if not significantly different at $P = 0.05$ in the cases of main effects

Plants	Low	Medium	High			
	99.5 ± 113	77.5 ± 73.6	79.3 ± 23.2			
Fish	Present	Absent				
	34.0 ± 36	137 ± 71				
Plants × Fish						
	LC	LF	MC	MF	HC	HF
Mean	192 ± 84	7.2 ± 5.6	140 ± 45	14.9 ± 7.7	78.9 ± 29	80 ± 20
LC		< 0.01	NS	< 0.01	NS	NS
LF			< 0.05	NS	< 0.01	< 0.01
MC				< 0.05	NS	NS
MF					< 0.01	< 0.01
HC						NS

Table 6 Effects of plant density and presence or absence of perch on body lengths of *Daphnia hyalina* in an experiment in enclosures in Little Mere. Values, in mm ± SD are means of three replicates of each treatment on each of four sampling dates. For each replicate at least 100 animals were measured, where sufficient were available. Otherwise all animals in the sample were measured, with $n > 30$ in all cases. Results were analysed by two-way ANOVA, followed by t-tests for data on paired with and without fish treatments. In these cases values, within dates, with shared superscript letters are not significantly different at $P = 0.05$

Date	29 June	7 July	26 July	9 August
LC	1.13 ± 0.70^{a}	1.26 ± 0.03^{a}	1.04 ± 0.09^{a}	1.30 ± 0.10^{a}
LF	1.00 ± 0.06^{b}	1.06 ± 0.07^{b}	1.02 ± 0.17^{a}	1.08 ± 0.11^{b}
MC	1.22 ± 0.11^{a}	1.24 ± 0.08^{a}	1.07 ± 0.07^{a}	1.31 ± 0.08^{a}
MF	1.12 ± 0.05^{b}	1.06 ± 0.16^{b}	1.08 ± 0.07^{a}	1.14 ± 0.06^{b}
HC	1.12 ± 0.04^{a}	1.11 ± 0.05^{a}	1.04 ± 0.06^{a}	1.15 ± 0.07^{a}
HF	1.13 ± 0.02^{a}	1.12 ± 0.11^{a}	1.07 ± 0.08^{a}	1.18 ± 0.85^{a}
Fish	$P < 0.001$	$P < 0.001$	NS	$P < 0.001$
Plants	$P < 0.001$	$P < 0.025$	NS	$P < 0.001$
Interaction	$P < 0.001$	$P < 0.001$	NS	$P < 0.001$

Table 7 Regression relationships among *Daphnia hyalina* (numbers L^{-1}), plant density (leaves per enclosure, taken as mean of initial and final numbers) and phytoplankton chlorophyll a (mg L^{-1}) in the experimental enclosures. All regressions are based on mean data ($n = 3$ per treatment)

y	x	Slope	Intercept	r	P
(a) Control enclosures					
Daphnia	Plants			0.60	> 0.05
Chlorophyll	Plants			0.28	> 0.25
Chlorophyll	*Daphnia*			0.21	> 0.25
(b) Enclosures with perch					
Daphnia	Plants			0.70	> 0.05
Chlorophyll	Plants	–0.38	45.9	0.74	< 0.05
Chlorophyll	*Daphnia*	–0.27	37.3	0.91	< 0.0005

organisms are closely associated with vegetation and may be much more cryptic than *Daphnia*, which is essentially an open-water genus.

The significant, if modest, size-selection in the predation on *Daphnia,* even at the lowest lily density, suggests that the predation was less intense than it might have been in open water, where such size-selective effects are common (Brooks & Dodson, 1965; Hall *et al*., 1976). In a similar experiment, in which Diehl (1992) used three densities of perch (zero, low, high) in the presence or absence of vegetation, it was found that perch ate macroinvertebrates in the presence of vegetation but moved to microcrustaceans in the absence of vegetation. The perch in the present experiment were of a size that might be expected to feed on benthic animals as well as plankton, although perch of even greater sizes in large lakes turn mainly to zooplankton in the summer (Fryer, 1991).

The nature of the refuge mechanism is not revealed by the experiment. It clearly did not involve repulsion of the fish by chemical secretions of the plants, for the fish could not escape and survival of fish was high. Fish are clearly not generally excluded from plant beds (Killgore, Morgan & Rybicki, 1989) and indeed may use them as refuges from their own predators (Rozas & Odum, 1988; Persson, 1991). Nor was it a function of deoxygenation in the beds, for there were no significant differences in oxygen concentrations related to plant density.

Differential light intensity may have been involved but there was sufficient light at depth, even in the highest density stands, to allow growth of submerged leaves and hence probably enough for visual predation. In dense plant beds, pH may be increased through photosynthetic activity. Experiments with changed pH in polyethylene bags in Little Mere have shown that pH 10.0 is sufficient to inhibit the feeding of perch without killing them (Beklioglu & Moss, 1995) and such pH values are not unknown in plant beds. It is possible that such a pH was attained in the lily beds but the simple effect of physical obstruction cannot be eliminated. Perch have been shown to hunt more effectively among structures (Winfield, 1986; Diehl, 1988) than some cyprinid fish so it may be expected that the refuge will operate even more effectively with fish such as roach and bream.

Daphnia has been shown elsewhere commonly to be the major component of the diet of perch in the size range used here (Koepke *et al*., 1988), although there is much flexibility in its diet and a tendency to concentrate on the most available prey (Terlecki, 1987). Thus another mechanism in the operation of the refuge, where perch, at least, is concerned, may be the existence of alternative food sources in the plant beds in the forms of sedimentary and epiphytic invertebrates. Benthic organisms were also found in the guts (see also Rask, 1983). If these animals are easier to catch than mobile ones in dense vegetation, then this may give an additional refuge mechanism for forms such as *Daphnia*. This emphasizes the need to carry out studies on phenomena such as refuges in the full complexity of natural situations rather than in the simplified conditions of laboratory aquaria where key variables may be excluded for the sake of simplicity.

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