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Food selection in freshwater omnivores: a case study of crayfish *Austropotamobius pallipes*

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With 4 figures and 7 tables

Abstract: The omnivorous habit and food selection in freshwater macroinvertebrates was investigated in the white-clawed crayfish, *Austropotamobius pallipes*, a threatened species. First, during an annual cycle we analysed gut contents for vascular plant detritus, moss, amorphous plant material, and animal remains, and evaluated their organic content. Second, we ran preference experiments in the laboratory, in which crayfish had to choose, in the first trial among three vegetal items, in the second trial among three animal prey, and in the third trial between the two preferred items of the previous two trials (i. e. moss and insect larvae). Third, we analysed whether the assimilation efficiencies of different food items affected crayfish food choice. Our results revealed that this species mostly acted as a detritus consumer, obtaining nutrition from the associated microbes, fungi, proto- and metazoans, but also showed herbivorous and carnivorous habits. However, at least when food resources were not limited, crayfish displayed distinct feeding preferences, often ruled by factors other than the nutritional quality and the assimilation efficiency of the selected food.

Key words: omnivory, food selection, freshwater communities, crayfish, *Austropotamobius pallipes*.

Introduction

Traditionally considered to be “rare in nature” (LAWTON 1989), to date omnivory (i. e., feeding on more than one trophic level, PIMM 1982) is seen as “a ubiquitous contravention of the trophic-level concept” (POLIS & STRONG 1996). Its prevalence in food webs of diversified habitats has reached the at-

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tention of community ecologists (e. g., POLIS 1991, DIEHL 1992). The potential of omnivory is to produce complex interactions within communities (DIEHL 1993). Multiple omnivorous links were shown to contribute to structuring freshwater food webs (DIEHL 1995).

Crayfish, usually defined as omnivorous consumers (HILL & LODGE 1994), play a relevant role in benthic food webs (NYSTRÖM et al. 1996, USIO 2000), constituting a large biomass in the littoral zones of lakes, ponds, and streams (LODGE et al. 1994). HUXLEY (1880) was the first author to describe crayfish feeding behaviour. After one century, a number of studies, based on gut content analysis, have been conducted on several species (e. g. SAFFRAN & BARTON 1993, O'BRIEN 1995, GUTIÉRREZ-YURRITA et al. 1998). Plant items more frequently occur in guts, enough to categorise crayfish as polytrophic decapods, mostly acting as detritivores/herbivores. In 1995, MOMOT suggested that crayfish species may be more carnivorous than previously thought and may be sufficiently predatory so as to potentially compete with fish for benthic macro-invertebrates.

The main purpose of this study was to describe food selectivity of omnivores through the analysis of the feeding behaviour of the white-clawed crayfish, *Austropotamobius pallipes*, a threatened species. Feeding preferences of other crayfish species have been analysed in previous laboratory studies that, however, provided only partial information on cost-benefits (ILHÉU & BERNARDO 1993), diverse digestibility of food items (ILHÉU & BERNARDO 1995), and differences in the chemistry of the ingested plants (CHAMBERS et al. 1991).

Here, a more extensive analysis of *A. pallipes*' selection has been conducted with both field and laboratory studies. Its diet was examined throughout a year from gut content analyses. Then, the organic carbon and nitrogen content of both guts and vascular plant detritus (the most exploited substrate during foraging, GHERARDI et al. 2001) was evaluated. We also investigated whether crayfish act as selective consumers, and whether assimilation efficiencies of different food items influence crayfish choice.

Materials and methods

The study animal

The white-clawed crayfish is native to Western Europe, where it occupies a wide geographical range from Dalmatia and the former Yugoslavia in the east, through Italy, France, Switzerland, northern and central Spain and northern Portugal to the British Isles (LAURENT 1988). In Italy, if we exclude some small populations of *A. torrentium* and *Astacus astacus*, it represents the only native species (GHERARDI et al. 1999 b). Notwithstanding this, the introduction of alien species spreading the crayfish plague and other factors such as pollution, over-fishing, floods, and drought are leading to a

pattern of distribution which becomes narrower each year (HOLDICH 1991). According to the European Council directive 92/43/ECC (21 May 1992), *A. pallipes* is declared "a species whose taking in the wild and exploitation may be subject to management measures".

The specimens used in this study came from a population inhabiting Fosso di Farfereta, a third order stream in the Tuscan-Emilian Apennine, 40 km from Florence (Italy) at an altitude of 450 m above sea level. Recent genetic studies (GRANDJEAN et al. 2002, FRATINI et al., unpubl.) suggested to assign the study population to the new species *A. italicus*. Awaiting for a formal acceptance of this new taxonomic status, we will still refer to this population as *A. pallipes*.

The structure and size of the population inhabiting this area have been monitored since 1996 and crayfish density was estimated to be 0.16/m² (GHERARDI et al. 1997). The area under study is unaffected by human interventions. The stream is mostly composed of shallow riffle sections, interspersed with pools, that seldom exceed 200 cm in depth and have an average width of 3 m. Water hardness is about 672 ppm with pH in the range 6.5–7. The substrate is composed of stones, a few rocks, and sandy or clay bottom. The study area is forested on both sides (*Corylus avellana*, *Carpinus betulus*, and *Acer campestre*) and is also surrounded by riparian vegetation consisting mostly of shrubs of eglantine (*Rosa canina*), elder (*Sambucus nigra*), and fescue (*Festuca heterophylla*). Vascular plant detritus (patches of decaying riparian shrubs and leaf material from deciduous trees), as well as moss (*Fontinalis* spp.), are the main sources of vegetal food available for crayfish. Potential animal prey that can be abundant are insect larvae (Ephemeroptera, Trichoptera, and Plecoptera), rainbow trout fry (the introduced *Oncorhynchus mykiss*), and toad tadpoles (*Bufo bufo*). Predators can be grey heron (*Ardea cinerea*) and adult rainbow trout (FOSTER & SLATER 1995).

Gut content analysis

Samples of crayfish consisting of 16–27 specimens were captured in each season during 1998. Crayfish were collected by hand after sunset, when the foraging activity of *A. pallipes* is high (BARBARESI & GHERARDI 2001). Each sample was composed of individuals of both sexes and mature and immature stages (in the population under study, males and females reach their sexual maturity at a cephalothorax length of, respectively, 30 and 25 mm, VILLANELLI & GHERARDI 1998).

Foreguts (called guts throughout the text and figures) were removed immediately after capture, to interrupt digestion and to preserve the ingested material in a recognisable condition. Only contents of the foregut were examined, because nearly all items in the mid- and hindguts were digested beyond recognition. The gut of each crayfish was split open, the content placed in a Petri dish containing a small amount of water, distributed as evenly as possible, and viewed under a dissecting microscope (magnification 30×).

The food items that we found in guts were classified as vascular plant detritus (i.e. recognisable leaf fragments), moss, amorphous plant material (i.e. plant items in a highly decomposed state), and animal remains. We estimated by sight the relative content of each food item and distinguished four classes of occurrence, i.e. 0–25%, 25–

50%, 50–75%, and 75–100%. On the basis of these classes, guts were assigned to one of the four categories from 0 to 3 (when guts had, respectively, 0–25% and 75–100% of their volume replenished with a given food item).

Gut contents and a quantity of vascular plant detritus collected from the study habitat were frozen to allow the subsequent quantitative study. These samples were dried for two days in an oven at 80 °C and weighed with an electronic balance. Then, the percentage of organic carbon and nitrogen contents of both guts and detritus were evaluated using a gas-chromatography technique (Carlo Erba NA 1500 Analyser).

Food preference experiment

Preference was studied in the laboratory on the basis of the differential consumption of six food categories by mature crayfish only. The food categories chosen shared the properties of being: commonly available in the stream where crayfish originated and/or highly represented in the gut contents and/or mostly preyed on as shown through direct observations (GHERARDI et al. 2001) and pilot tests. Crayfish preference for the different items was calculated by measuring the quantity (dry weight) of each food ingested.

During Spring 1998, sixty specimens of both sexes (cephalothorax length ranging 31.5–40.0 mm) were isolated immediately after capture and starved for 5 days in rectangular polypropylene containers (20 × 15 cm). Three trials (each composed of 20 replicates conducted on different specimens) were carried out in a thermostatic cell (18–19 °C) under a natural light: dark regime (6:00 light on, 20:00 light off). Experimental aquaria were circular (40 cm in diameter). Food items were fixed to the bottom with a metal weight, equidistant between each other and near to the border, following an order which was randomised among replicates. This assured that the crayfish, once put into the centre of the container, had the same chance of encountering each type of food and avoided any experimental bias. To eliminate prey mobility and thus the influence of differential encounter rates, prey were killed immediately before the experiment in a freezer at –30 °C, where death occurred after a few seconds. Pilot tests showed that defrosted prey were still attractive to the crayfish.

Crayfish had to choose, in the first trial, among moss (*Fontinalis antipyretica*), fresh leaves of the hazel (*Corylus avellana*, a common riparian tree) and plant detritus (composed of decaying leaf material from deciduous trees), and in the second among insect larvae (3 Ephemeroptera, 3 Trichoptera, and 3 Plecoptera), toad (*Bufo bufo*) tadpoles (5 individuals) and rainbow trout (*Oncorhynchus mykiss*) fry (1 specimen), and in the third between the two items which were preferred in the previous two trials. To eliminate any bias due to a differential handling time, Trichoptera were extracted from their cases.

In each replicate, wet pre-weighed amounts of the different food items having a similar volume were placed in each container, allowing crayfish to feed ad libitum. Food, which was not consumed after two days, was dried for 48 hours in an oven at 80 °C and then weighed with an electronic balance. As a control, we kept for two days the same amount of the six food items in single containers without crayfish to see whether food was subject to change in its weight in this experimental condition independently of the crayfish presence. The relationship between wet and dry weight was

calculated for each type of food. The consumption of each item was obtained by subtraction of the remaining amount from the supplied food quantity.

Organic content and assimilation efficiency

Percentages of both the quantity (in dry weight) and the organic content (carbon and nitrogen) assimilated by crayfish when they were free to feed on animal and plant items offered in the food preference experiment were evaluated in a study conducted during June 1999. Thirty-two adult crayfish (25 males and 7 females, with a cephalothorax length ranging 30–39.5 mm) were isolated and starved for 6 days in rectangular polypropylene containers (20×15 cm). Pre-weighed amounts of each food item used in the preference experiment, having approximately the same volume, were placed in the containers and crayfish were allowed to eat *ad libitum* for two days. To prevent coprophagy, a 2-cm-high section at the basis of the aquarium was separated from crayfish by fixing a polyethylene net of 4×4 mm mesh that however permitted the passage of faeces. Faeces were collected several times a day, during the foraging period and three days after. Non ingested food and faeces were treated as in the preference experiment in order to obtain the dry weight of ingested and defecated matter. Using these values, the dry matter assimilation efficiency for each food category was determined, following the equation (WHITLEGGE & RABENI 1997):

$$AE (\%) = 100 \times (\text{ingested} - \text{defecated})/\text{ingested}.$$

The assimilation efficiency of organic matter for each food category was computed using the above equation knowing the amount assimilated, the mean organic carbon and nitrogen percentages of the food items and the organic carbon and nitrogen content of individual faeces (evaluated using the methods as described above in the Gut content analysis section).

The estimate of nitrogen in faecal matter appeared unbiased, because the ammonia released into the water during the 5 days of the experiment as the result of crayfish excretion, faeces leaching, and food decomposition was very low (following the Nessler's method – see APHA et al. 1976: 0.001 mg/l in the presence of either detritus, or fish or insect larvae; 0.002 mg/l in the presence of leaves; and 0.003 mg/l in the presence of either moss or tadpoles).

Statistical analyses

Parametric statistical techniques were used following ZAR (1984), when the scores appeared to be drawn from a normally distributed population.

Student's *t*-test (statistic: *t*) and a one-way ANOVA (after arcsine square root transformation for percentage values, statistic: *F*) were used to compare each of the gut content parameters between sexes, between maturity stages, and among seasons, as well as to compare the organic content of the six food items offered in the preference experiment and in the assimilation efficiency study. We used a two-way ANOVA (after arcsine square root transformation) to analyse differences in the organic content between detritus and guts among seasons. The Tukey test was applied to show hierarchies when a difference was found.

When the assumptions of normality and homoscedasticity in the population distribution were not met, statistical inferences were made through non-parametric tests (SIEGEL & CASTELLAN 1988). In the preference experiment choices were not independent, thus the Friedman test (statistic: F_r) (LOCKWOOD III 1998) and the Wilcoxon signed ranks test (statistic: z for large samples) were used when crayfish were presented with, respectively, three and two food items. When differences were found after the Friedman test, we applied a test of Multiple Comparisons among Conditions to show hierarchies among the food items chosen. The Wilks test (statistic: G) was used when data were on an ordinal scale.

The level of significance at which the null hypothesis was rejected was $\alpha = 0.05$.

Results

Gut contents

We classified guts into 4 categories of increasing relative content (0–3) of every food item (vascular plant detritus, moss, amorphous plant material, and animal remains) (see Materials and methods section). Crayfish having a larger volume of detritus in their gut (gut category 3) were more numerous than

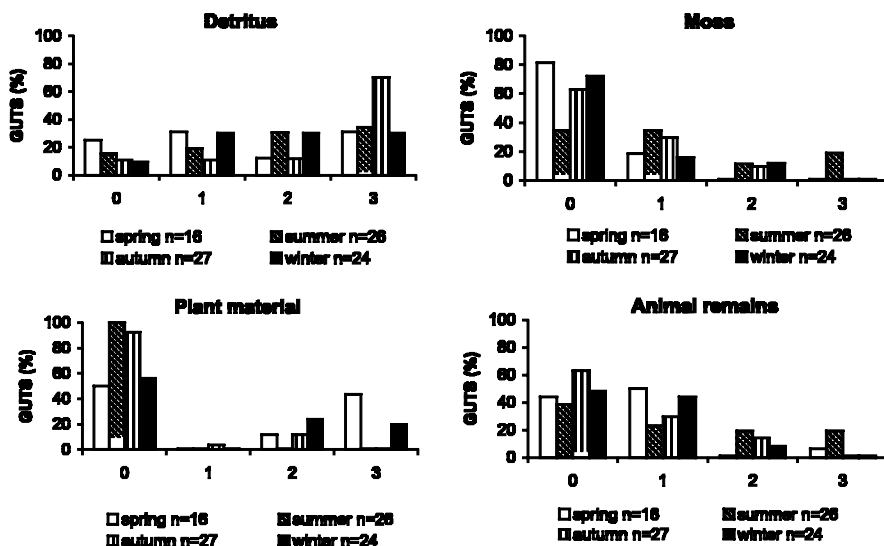


Fig. 1. Frequency of guts (%), classified into 4 categories (0–3), distinguished per season and food content (i. e. vascular plant detritus, moss, amorphous plant material, and animal remains). Gut categories were defined from the relative content of every food item. Relative content (and gut category in parenthesis) were: 0–25 % (0), 25–50 % (1), 50–75 % (2), and 75–100 % (3). For instance, guts of the category 0 for a given food item were replenished with that food item for 0–25 % of their volume. Data from all the collected crayfish per season were pooled.

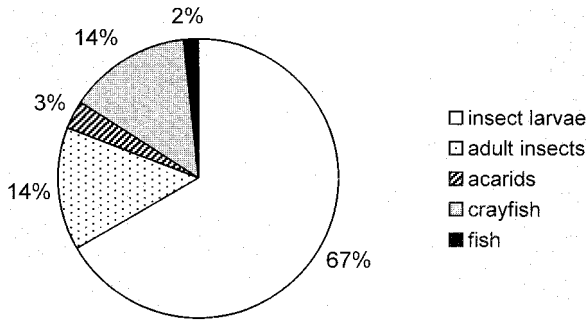


Fig. 2. Frequency of guts (%) where we recorded at least one piece of five animal categories. Because of the low occurrence of animal remains, guts found with at least one animal category throughout the year have been pooled together without any distinction of sexes and sizes. Sample size is 48.

Table 1. Mean + SE of the quantitative patterns of gut contents of all animals pooled per season.

	mean	SE	n
spring			
gut fullness (%)	33.12	6.87	16
dry weight (mg)	5.89	1.24	16
organic C (%)	35.32	1.52	15
organic N (%)	5.36	0.55	16
summer			
gut fullness (%)	56.35	5.37	26
dry weight (mg)	29.52	5.15	25
organic C (%)	36.26	1.48	25
organic N (%)	6.65	0.51	25
autumn			
gut fullness (%)	41.11	5.59	27
dry weight (mg)	14.10	5.40	27
organic C (%)	40.96	0.54	24
organic N (%)	4.55	0.30	24
winter			
gut fullness (%)	30	5.90	24
dry weight (mg)	8.59	2.26	24
organic C (%)	38.64	1.20	23
organic N (%)	6.69	0.34	23

those having other food categories in autumn and winter (autumn: $G = 76.861$, $df = 9$, $P < 0.001$; winter: $G = 37.253$, $df = 9$, $P < 0.001$); in contrast, guts with larger quantities of amorphous plant material were more frequent in spring than in the other seasons ($G = 29.934$, $df = 9$, $P < 0.001$) and no difference among food items was found in summer ($G = 7.900$, $df = 6$, $P > 0.05$). The

Table 2. Quantitative patterns of gut content: comparison among seasons (one-way ANOVA after arcsine square root transformation for percentages, and Tukey test). sp = spring, su = summer, au = autumn, wi = winter. * $P < 0.05$.

	F	df	
gut fullness (%)	3.10*	3 & 89	su > au = wi = sp
dry weight (mg)	5.61*	3 & 88	su > au > wi = sp
organic C (%)	3.97*	3 & 83	au = wi > sp = su
organic N (%)	5.75*	3 & 84	su = wi > sp = au

Table 3. Mean + SE and comparison of the quantitative patterns of gut contents between sexes for each season (Student's t-test after arcsine square root transformation for percentages). * $P < 0.05$.

	Males			Females			t	df
	mean	SE	n	mean	SE	n		
spring								
gut fullness (%)	25.42	7.17	12	56.25	11.97	4	2.189*	14
dry weight (mg)	5.15	1.31	12	8.10	3.07	4	1.675	14
organic C (%)	35.98	1.92	11	33.49	2.25	4	0.695	13
organic N (%)	5.82	0.67	12	3.98	0.42	4	1.523	14
summer								
gut fullness (%)	58.67	6.08	15	53.18	3.91	11	0.689	24
dry weight (mg)	31.67	7.09	15	26.30	7.57	10	0.503	23
organic C (%)	35.94	2.03	15	36.77	3.96	10	0.278	23
organic N (%)	6.77	0.68	15	6.47	0.83	10	0.291	23
autumn								
gut fullness (%)	44.41	6.84	17	35.50	9.82	10	0.927	25
dry weight (mg)	18.07	8.13	17	7.33	4.43	10	0.959	25
organic C (%)	40.78	0.68	16	41.33	0.84	8	0.463	22
organic N (%)	4.78	0.34	16	4.47	0.56	8	1.186	22
winter								
gut fullness (%)	29.00	7.73	15	31.67	9.61	9	0.479	22
dry weight (mg)	8.88	2.94	15	8.10	3.95	9	0.342	22
organic C (%)	38.08	1.32	15	39.68	2.44	8	0.590	21
organic N (%)	7.00	0.43	15	6.09	0.54	8	0.217	21

analysis of each volume category separately (Fig. 1) showed that more numerous guts contained larger quantities (category 3) of: detritus during autumn ($G = 16.132$, $df = 9$, P ca. 0.05), moss during summer ($G = 21.132$, $df = 9$, $P < 0.05$), amorphous plant material in both spring and winter ($G = 21.132$, $df = 9$, $P < 0.02$), and animal remains in both spring and summer ($G = 19.043$, $df = 9$, $P < 0.05$).

Male guts contained a relatively larger volume of animal material than female guts in summer ($G = 10.791$, $df = 3$, $P < 0.02$), while the opposite was found in the case of detritus in autumn ($G = 6.642$, $df = 2$, P ca. 0.05). Imma-

Table 4. Mean + SE and comparison of the quantitative patterns of gut contents between stages for each season (Student's t-test after arcsine square root transformation for percentages). * $P < 0.05$.

	Immature crayfish			Mature crayfish			t	df
	mean	SE	n	mean	SE	n		
spring								
gut fullness (%)	35.83	14.28	6	31.5	7.56	10	0.159	14
dry weight (mg)	4.41	1.16	6	6.77	1.84	10	0.921	14
organic C (%)	32.88	1.98	6	36.61	2.28	9	1.338	13
organic N (%)	4.52	0.39	6	5.86	0.82	10	1.075	14
summer								
gut fullness (%)	54.29	8.30	14	58.75	6.80	12	0.536	24
dry weight (mg)	14.54	2.97	13	45.75	7.99	12	4.67*	23
organic C (%)	34.59	1.73	13	38.09	2.36	12	1.153	23
organic N (%)	5.81	0.73	13	7.57	0.75	12	1.634	23
autumn								
gut fullness (%)	33.67	6.18	15	50.42	9.54	12	1.057	25
dry weight (mg)	5.23	1.86	15	25.18	11.38	12	1.929	25
organic C (%)	40.69	0.76	14	41.34	0.72	10	0.581	22
organic N (%)	5.14	0.39	14	3.72	0.31	10	2.545*	22
winter								
gut fullness (%)	29.23	9.05	13	30.91	7.65	11	0.375	22
dry weight (mg)	5.93	1.95	13	14.95	4.97	11	1.269	22
organic C (%)	34.6	3.24	13	38.70	1.57	10	0.970	21
organic N (%)	6.12	0.66	13	6.74	0.55	10	0.138	21

ture individuals seemed to consume relatively larger volumes of both detritus ($G = 11.116$, $df = 3$, $P < 0.02$) and amorphous plant material ($G = 6.226$, $df = 2$, $P < 0.05$) than mature specimens during the winter season.

Because of the low occurrence of animal remains, data recorded throughout the year were pooled without any distinction of sexes and age classes. These included insect larvae (Ephemeroptera, Trichoptera, Plecoptera, and Odonata), adult insects (among others, mostly ants and aquatic Coleoptera), other arthropods such as acarids, crayfish (mostly pieces of exoskeleton), and small fish. There was a significant difference among the ingested items ($G = 75.394$, $df = 4$, $P < 0.001$), with insect larvae being the most frequent prey (67% of occurrence), followed by both adult insects and crayfish remains. Acarids and fish were the least represented categories (Fig. 2).

Data on relative gut fullness (in %), dry weight (in mg) and organic carbon and nitrogen percentages of gut contents of all animals pooled per season are shown in Table 1. Based on a one-way ANOVA (Table 2), guts displayed a higher relative fullness and the dry weight of their content was heavier in summer. Gut contents were richer in organic carbon in autumn and winter but poorer in organic nitrogen in autumn and spring.

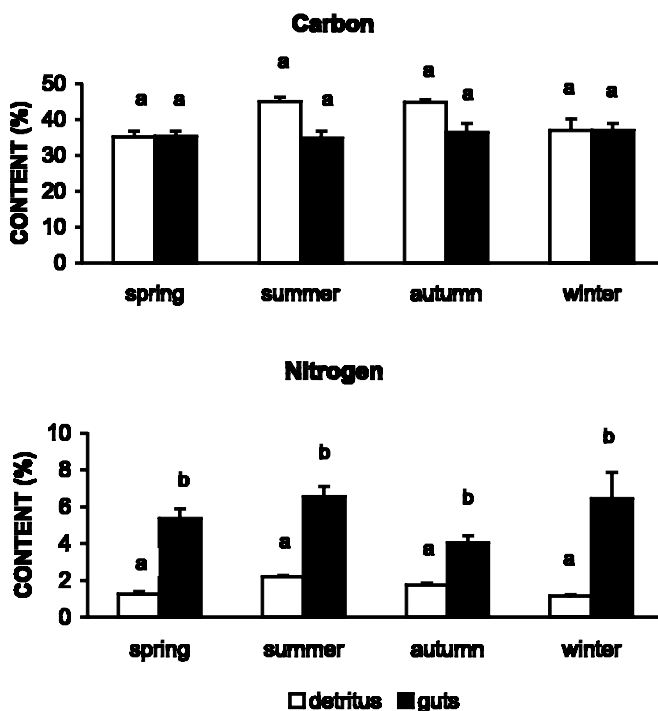


Fig. 3. Organic carbon (top) and nitrogen (bottom) content (in %, mean + SE) in vascular plant detritus collected from the environment and crayfish guts: a comparison among seasons. Different letters indicate significant differences between detritus and guts ($P < 0.05$). In spring, summer, autumn, and winter, sample sizes were, respectively: 3, 5, 9, and 5 for detritus and 16, 26, 27, and 24 for guts.

Female guts had a higher relative fullness in spring than male guts (Table 3), while gut contents of immature individuals were heavier in summer and richer in nitrogen content in autumn than those of mature specimens (Table 4).

Organic carbon and nitrogen contents of detritus differed among seasons, being significantly higher in both summer and autumn (carbon: $F_{3,17} = 6.780$, $P < 0.01$; nitrogen: $F_{3,17} = 16.740$, $P < 0.001$). When a comparison was done between the organic content of detritus and guts, the latter proved to be richer in nitrogen than the former (after a two-way ANOVA, $F_{1,107} = 39.29$, $P < 0.01$) in the four seasons ($F_{3,107} = 4.34$, $P < 0.01$), whereas no significant difference was ever found in carbon content ($F_{1,105} = 0.001$, $P > 0.05$; $F_{3,105} = 0.001$, $P > 0.05$) (Fig. 3).

Food preferences

In the absence of crayfish, the six food items here tested were subject to slight changes in their weight ($P > 0.05$) that did not affect the outcome of the food preference experiment.

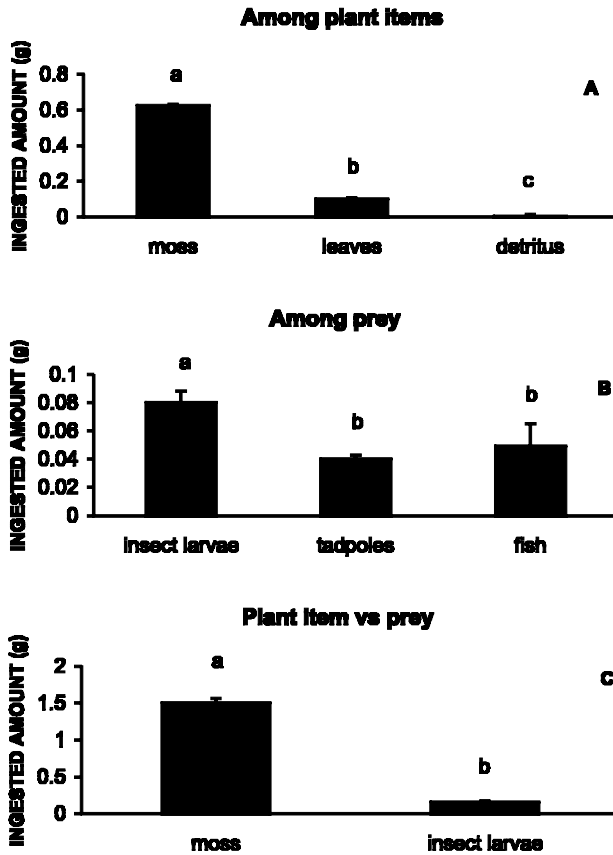


Fig. 4. Results of the food preference experiment (mean dry weight amount ingested for each food category in g + SE): choice among plant items (A), among prey (B), and between the preferred plant item and prey (C). Different letters indicate significant differences among choices. The number of tested crayfish in each trial was 20.

When presented with the three plant materials, crayfish chose differently ($F_r = 34.962$, $df = 2$, $P < 0.01$) and showed a preference for moss with respect both to leaves ($P < 0.05$) and to detritus ($P < 0.05$) and for leaves with respect to detritus ($P < 0.05$) (Fig. 4 A). When crayfish were tested with prey, a clear preference was revealed for insect larvae ($F_r = 11.2$, $df = 2$, $P < 0.01$) rather than for toad tadpoles and trout fry ($P < 0.05$) (Fig. 4 B). Crayfish chose moss when this was offered together with insect larvae (Wilcoxon signed ranks test, z for large samples: $z = 1.96$, $P < 0.01$) (Fig. 4 C).

Organic content

Moss had a lower content of both carbon and nitrogen than fresh leaves and detritus (Table 5; $F_{2,14} = 106.24$ and 8.03 , $P < 0.01$), while fish were richer than

Table 5. Organic carbon and nitrogen content (% based on dry weight, mean + SE) for six food categories used in the food preference experiment.

	moss	leaves	detritus	insect larvae	tadpoles	fish
Carbon						
n	5	5	7	5	5	5
mean	27	46.38	45.15	41.75	17.45	51.83
SE	1.48	0.77	0.61	0.21	0.2	0.82
Nitrogen						
n	5	5	7	5	5	5
mean	1.17	2.12	1.8	8.23	2.04	11.75
SE	0.16	0.19	0.14	0.13	0.17	0.44

Table 6. Dry matter assimilation efficiency (AE, mean + SE) of six food categories used in the food preference experiment.

	moss	leaves	detritus	insect larvae	tadpoles	fish
n	7	5	6	5	4	5
mean	90.25	87.79	81.01	73.17	80.25	93.88
SE	2.59	4.26	5.45	3.22	2.61	1.41

the other prey (carbon: $F_{2,12} = 494.18$, $P < 0.01$; nitrogen: $F_{2,12} = 417.62$, $P < 0.01$). Both organic carbon ($t = 9.894$, $df = 8$, $P < 0.01$) and nitrogen ($t = 34.88$, $df = 8$, $P < 0.01$) were higher in insect larvae than in the moss.

Dry matter assimilation efficiency

AE did not differ among plant items ($F_{2,15} = 1.44$, $P > 0.05$), but AE of fish fry was significantly higher ($F_{2,11} = 16.85$, $P < 0.01$) than the AE of both insect larvae (after Tukey test, 8.12 vs. 3.82) and toad tadpoles (4.91 vs. 3.82) (Table 6). Moreover, moss showed a higher AE than insect larvae ($t = 4.171$, $df = 10$, $P < 0.01$).

Organic assimilation efficiency

The assimilation efficiency of organic carbon and nitrogen (Table 7) did not differ among the three plant items here tested ($F_{2,15} = 0.72$, $P > 0.05$; nitrogen: $F_{2,15} = 0.99$, $P > 0.05$). While no difference was observed in the case of nitrogen when the three prey were compared ($F_{2,11} = 3.65$, $P > 0.05$), a higher carbon assimilation efficiency was recorded in fish ($F_{2,11} = 9.35$, $P < 0.01$) than in insect larvae (after Tukey test, 5.5 vs. 3.82) and tadpoles (4.95 vs. 3.82). Moreover, carbon was more assimilated when the food was moss compared with insect larvae ($t = 3.329$, $df = 10$, $P < 0.01$).

Table 7. Organic carbon and nitrogen assimilation efficiency (% based on dry weight, mean + SE) for six food categories used in the food preference experiment.

	moss	leaves	detritus	insect larvae	tadpoles	fish
Carbon						
n	7	5	6	5	4	5
mean	89.88	88.56	84.42	78.36	79.53	96.49
SE	2.56	3.49	4.45	1.91	6.15	0.73
Nitrogen						
n	7	5	6	5	4	5
mean	90.14	84.13	81.83	88.03	84.75	98.68
SE	2.7	5.18	5.77	0.82	7.34	0.28

Discussion

Until now, few direct tests on food selection by freshwater omnivores have been conducted. For generalist herbivores, LODGE et al. (1998) suggested a conceptual model based on the rationale that we need to understand which plants are mostly affected by herbivores and how macrophyte communities change under the influence of herbivory. To be preferentially consumed – these authors stated – a macrophyte must, first, have a structure that makes it possible for a herbivore to take a bite; second, lack chemical deterrents; and, third, be nutritious. These three elements of the model follow the issue that decision rules for gathering food are to be faced by a wide variety of predators, regardless of their feeding tactics (KREBS 1978). It is a prerequisite for a model of optimal prey choice that predators should be able to distinguish between items of different ratio net food value: handling time and to select the more profitable types (KREBS 1978).

From the “energy maximisation theory” (SCHOENER 1971) viewpoint, it seems like a paradox that crayfish in natural habitats frequently have plants and detritus in their guts, but in laboratory experiments often behave as energy maximisers and prefer macro-invertebrates (NYSTRÖM 2002). In order to determine crayfish food preference, we analysed gut contents, a method that, although criticised (NYSTRÖM 2002), furnishes good information on recent crayfish feeding activity, especially if integrated with direct observations of foraging (GHERARDI et al. 2001). These data showed that *A. pallipes* (1) is mostly detritivorous, but (2) behaves as a polytrophic and opportunistic consumer and (3) modifies its feeding activity throughout its life cycle. On the other hand, experimental studies revealed (4) that this species is selective and that food is chosen on the basis of factors other than its higher nutritional quality and assimilation efficiency.

1. Detritivory

Proportions of food types in *A. pallipes*' gut contents were similar to those of many other stream- and lake-dwelling crayfish, which also consumed large amounts of vascular plant detritus and lesser quantity of other plant items and animal matter (MOMOT et al. 1978, WHITLEDGE & RABENI 1997, GUTIÉRREZ-YURRITA et al. 1998). Previous field studies (GHERARDI et al. 2001) showed that the occupation by male *A. pallipes* of detritus patches was not related simply to its availability on the habitat, but seemingly to a choice operated by crayfish.

Several authors (e. g., POLIS & STRONG 1996) have stated that detritivory is a common form of omnivory. Detritus originates differently throughout the trophic spectrum and does not form one homogeneous food source, epiphytic microflora and protozoan being associated with decaying vegetation. Plant detritus is also colonised by invertebrate browsers, collector-gatherers, shredders and predators (USIO 2000) and serves as a refuge for a large community of both insects and soft-bodied metazoans (MOMOT 1995).

The nutritional value of detritus has been the object of opposing views. On the one hand, up to 50 % of the nitrogen found in aged detritus appeared to consist of non-proteinaceous compounds, i. e. relatively refractory complexes with lignin and lignin-like substances formed in diagenesis (e. g., RICE 1982). On the other, a number of authors considered detritus to be highly nutritional (D'ABRAMO & ROBINSON 1989), because of the association with bacteria and fungi that alter its composition (GODDARD 1988).

This second viewpoint seems to be confirmed by our results. The assimilation efficiency of detritus by *A. pallipes* was 81 %. This relatively high value can be explained by the cellulolytic activity revealed in this species' hindgut (GHERARDI et al. 1999 a) and agrees with estimates by ÍLHÉU & BERNARDO (1995) for *Procambarus clarkii* (62 and 73 % for, respectively, old and fresh detritus). However, it is decidedly higher than the percentage provided by WHITLEDGE & RABENI (1997) for *Orconectes luteus* (14 %). ÍLHÉU & BERNARDO (1995) showed that detritus was more digestible than fresh macrophytes, but from our data crayfish assimilation efficiencies did not significantly differ among plant items.

Organic substances of detritus, particularly nitrogen, varied in their concentration throughout the year. The highest nitrogen content recorded in summer may be related to both the increased metabolic activity of micro-organisms and the hatching of insects and other arthropods in detritus (HYNES 1970).

According to MOMOT (1995), detritus may serve to provide some energy for crayfish maintenance but may be quite incapable of sustaining growth, confirming results provided by HILL et al. (1993). Our data showed that, although plant detritus is rich in organic substances and is efficiently assim-

ilated, *A. pallipes*' guts always displayed a higher nitrogen content than the detritus. As a consequence, the latter seems not to be the only source of proteins for crayfish, but may be integrated by animal prey.

2. Polytrophic and opportunistic behaviour

Validating field observations on foraging (GHERARDI et al. 2001), the moss, *Fontinalis antipyretica*, was the green plant most frequently found in *A. pallipes*' guts, supporting observations by BROWN & BOWLER (1977) in Northumbria, England. This is somewhat surprising in that crayfish are known to feed mostly on algae and macrophytes in other systems (REYNOLDS 1978, MATTHEWS et al. 1993, NYSTRÖM 1999). In our study, moss was digested by crayfish to a similar extent as detritus and its organic carbon and nitrogen contents were equally assimilated. Its structural complexity may favour the protection to invertebrates; both periphyton growing on it and fine particles trapped in these plants are food sources for many invertebrates (GILLER & MALMQVIST 1998).

Since crayfish are unable to float and can swim only backwards, their ability to capture more motile aquatic animals is low and they normally feed on passive materials (D'ABRAMO & ROBINSON 1989). Thus, the most usual prey are benthic invertebrates, especially insect larvae, as shown in other species (ILHÉU & BERNARDO 1993).

A percentage, although low (2%), of animal remains originated from fish prey. Predation on fish has been reported previously (see, e. g., GUTIÉRREZ-YURRITA et al. 1998). In laboratory aquaria in the presence of trout fry, adult *A. pallipes* and *Procambarus clarkii* displayed a 'sit-and-wait' preying strategy (RENAI & GHERARDI 2004). Since fry are abundant in shallow waters, fish may be an important animal food source for large individuals. Other animal food sources such as molluscs, especially snails, annelid worms, leeches and frogs were mentioned as crayfish prey (DINIS 1978, NYSTRÖM 1999).

Cannibalism was considered as a supplementary dietary requirement (CAPPELLI 1980), especially when the metabolic demand for calcium is great. Rather, field and laboratory observations suggested that in this species cannibalism is uncommon, even though small crayfish would seem to be vulnerable to predation by larger individuals; on the contrary, consumption of the own post-moult exuviae occurs frequently (LODGE & HILL 1994, F. GHERARDI, pers. observ.) and possibly accounts for exoskeleton fragments found in some *A. pallipes*' guts.

The observed changes throughout the year in the relative amount of ingested items reflect the opportunistic behaviour of this crayfish. Detritus was more represented in guts during autumn, which coincided with the period of

peak litter accumulation. Whereas moss was mostly found in summer, the ingestion of prey as insect larvae increased during spring and summer seasons, when their biomass reaches its highest level.

3. Feeding and life cycle

Austropotamobius pallipes modifies the intensity of its feeding behaviour throughout the year as an obvious consequence of its activity cycle (GHERARDI et al. 1997). In winter, crayfish showed reduced activity, taking refuge inside shelters or hiding under banks. Because activity reaches maximum in summer (BARBARESI & GHERARDI 2001), gut contents had a higher relative fullness in this than in the other seasons.

Orconectes propinquus (CAPELLI 1980) and *Procambarus clarkii* (GUTIÉRREZ-YURRITA et al. 1998) show no differences in diet between sexes. In the present study, the higher relative fullness of female guts in spring, the large amount of animal material in male guts in summer and of detritus in female guts in autumn might be related to this species' breeding cycle (VILLANELLI & GHERARDI 1998).

Several authors underlined that animal material is much more important in the diets of rapidly growing juveniles than of adults (ILHÉU & BERNARDO 1993, O'BRIEN 1995). A shift in food habits seems to accompany an increase in length as mentioned for several crayfish species (e.g., ILHÉU & BERNARDO 1993, WHITLEGE & RABENI 1997). Our findings showed that immature *A. pallipes* had gut contents relatively heavier than mature individuals in summer when the growth rate of juveniles is likely to be more intense. Moreover, the diet of immature specimens was characterized by a higher energetic and proteinaceous input in autumn, when their gut contents were richer in nitrogen than those of mature ones.

4. Selectivity

In the presence of different food sources, mature *A. pallipes* showed distinct feeding preferences. Selectivity has been reported for other crayfish in laboratory feeding experiments (CHAMBERS et al. 1991, SAFFRAN & BARTON 1993, ILHÉU & BERNARDO 1995, NYSTRÖM & STRAND 1996). Also, selective foraging by crayfish has been observed in controlled mesocosm studies (CHAMBERS et al. 1990, HANSON et al. 1990) and under natural conditions (LODGE & LORMAN 1987). *Austropotamobius pallipes* juveniles showed a higher level of carnivory than the adults, when subject to laboratory experiments (REYNOLDS 1978).

For macroconsumers such as crayfish, preferences may be related to differences in food nutritional quality and assimilation efficiency. For example, *A. pallipes*' choice of moss over insect larvae (on the opposite, see, ILHÉU &

BERNARDO 1993 for *Procambarus clarkii*) was possibly due to the higher efficiency of carbon assimilation from the former.

On the other hand, preference for moss over plant material may be explained by diverse palatability, chemical deterrents (e. g., alkaloids, CHAMBERS et al. 1991, and phenolics, LODGE 1991), structural defences (BRYANT & KUROPAT 1980), and ease of handling. Similarly, LODGE & LORMAN (1987) observed that *Orconectes rusticus* favoured single-stemmed species over rosulate or highly branched forms. The macroalga *Chara* and the duckweed species *Lemna* were chosen by *Orconectes virilis* possibly because the number of stalks or individual plants per unit weight is high (CHAMBERS et al. 1991). Moss morphology, occurring in 'bite-size' pieces, could be selected by *A. pallipes* over other plant items that need to be manipulated into smaller pieces.

Among prey, insect larvae were preferred over trout fry that however were more efficiently assimilated. One hypothesis that awaits to be tested is that *A. pallipes*' choice mostly depends on a 'searching image' (DAWKINS 1971) of insect larvae. In the white-clawed crayfish as a predator, the development of a searching image may be the result of visual and/or chemical perceptual processes, allowing a faster detection and an easier capability to manipulate the species they are more familiar with.

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References

- APHA, AWWA, and WPCF (1976): Standard methods for the examination of water and wastewater. – 14th edition. American Public Health Association, Washington, DC.
- BARBARESI, S. & GHERARDI, F. (2001): Daily activity of the white-clawed crayfish, *Austropotamobius pallipes* (Lereboullet): a comparison between field and laboratory studies. – *J. Nat. Hist.* **35**: 1861–1871.
- BROWN, D. J. & BOWLER, K. (1977): A population study of the British freshwater crayfish *Austropotamobius pallipes* (Lereboullet). – *Freshwat. Crayfish* **3**: 33–50.
- BRYANT, J. P. & KUROPAT, P. J. (1980): Selection of winter forage by subarctic browsing vertebrates: the role of plant chemistry. – *Ann. Rev. Ecol. Syst.* **11**: 261–285.
- CAPELLI, G. M. (1980): Seasonal variation in the food habits of the crayfish *Orconectes propinquus* (Girard) in Trout Lake, Vila County, Wisconsin, U. S. A. (Decapoda: Astacidea, Cambaridae). – *Crustaceana* **38**: 82–86.
- CHAMBERS, P. A., HANSON, J. M., BURKE, J. M. & PREPAS, E. E. (1990): The impact of the crayfish *Orconectes virilis* on aquatic macrophytes. – *Freshwat. Biol.* **24**: 81–91.

- CHAMBERS, P. A., HANSON, J. M. & PREPAS, E. E. (1991): The effect of aquatic plant chemistry and morphology on feeding selectivity by the crayfish, *Orconectes virilis*. – *Freshwat. Biol.* **25**: 339–348.
- D'ABRAMO, L. R. & ROBINSON, E. H. (1989): Nutrition of crayfish. – *Rev. Aquat. Sci.* **1**: 711–728.
- DAWKINS, M. (1971): Perceptual changes in chicks, another look at the 'search image' concept. – *Anim. Behav.* **19**: 566–574.
- DIEHL, S. (1992): Fish predation and benthic community structure: the role of omnivory and habitat complexity. – *Ecology* **73**: 1646–1661.
- (1993): Relative consumer sizes and the strengths of direct and indirect interactions in omnivorous feeding relationships. – *Oikos* **68**: 151–157.
- (1995): Direct and indirect effects of omnivory in a littoral lake community. – *Ecology* **76**: 1727–1740.
- DINIS, M. T. (1978): Notas sobre uma experiencia de estabelacao do lagostim de agua doces *Astacus pallipes* (Lereboullet), no Aquario Vasco de Gama. Redatorios de Actividades do Aquario Vasco de Gama, Portugal.
- FOSTER, J. & SLATER, F. M. (1995): A global review of crayfish predation with observations on the possible loss of *Austropotamobius pallipes* in the Welsh Wye due to crayfish plague. – *Freshwat. Crayfish* **8**: 589–613.
- FRATINI, S., ZACCARA, S., BARBARESI, S., GRANDJEAN, F., SOUTY-GROSSET, C., CROSA, G. & GHERARDI, F. (submitted): Phylogeography of the threatened crayfish (genus *Austropotamobius*) in Italy: implications for its taxonomy and conservation. – *Heredity*.
- GHERARDI, F., VILLANELLI, F. & DARDI, P. (1997): Behavioral ecology of the white-clawed crayfish *Austropotamobius pallipes* in a Tuscan stream: preliminary results. – *Freshwat. Crayfish* **11**: 182–193.
- GHERARDI, F., ACQUISTAPACE, P., SALVI, G. & MASTROMEI, G. (1999 a): Cellulolytic activity in the hindgut of crayfish (Decapoda, Astacoidea). – *Crustaceana* **72**: 534–536.
- GHERARDI, F., BALDACCINI, G. N., ERCOLINI, P., BARBARESI, S., DE LUISE, G., MAZZONI, D. & MORI, M. (1999 b): The situation in Italy. – In: GHERARDI, F. & HOLDICH, D. M. (eds): *Crayfish in Europe as alien species*. – A. A. Balkema, Rotterdam, Brookfield, pp. 107–128.
- GHERARDI, F., ACQUISTAPACE, P. & SANTINI, G. (2001): Foraging by a threatened species – the white-clawed crayfish, *Austropotamobius pallipes*. – *Arch. Hydrobiol.* **152**: 339–351.
- GILLER, P. S. & MALMQVIST, B. (1998): *The biology of streams and rivers*. – Oxford University Press, Oxford, U. K.
- GODDARD, J. S. (1988): Food and feeding. – In: HOLDICH, D. M. & LOWERY, R. S. (eds): *Freshwater crayfish: biology, management and exploitation*. – Chapman & Hall (Croom Helm), London, U. K., pp. 145–166.
- GRANDJEAN, F., FRELON-RAIMOND, M. & SOUTY-GROSSET, C. (2002): Compilation of molecular data for the phylogeny of the genus *Austropotamobius*: one species or several? – *Bull. Fr. PêchePiscic* **367**: 671–680.
- GUTIÉRREZ-YURRITA, P. J., SANCHO, G., BRAVO, M. Á., BALTANÀS, Á. & MONTES, C. (1998): Diet of the red swamp crayfish *Procambarus clarkii* in natural ecosystems of the Doñana National Park temporary fresh-water marsh (Spain). – *J. Crustacean Biol.* **18**: 120–127.

- HANSON, J. M., CHAMBERS, P. A. & PREPAS, E. E. (1990): Selective foraging by the crayfish *Orconectes virilis* and its impact on macroinvertebrates. – *Freshwat. Biol.* **24**: 69–80.
- HILL, A. M. & LODGE, D. M. (1994): Diel changes in resource demand: competition and predation in species replacement among crayfishes. – *Ecology* **75**: 2118–2126.
- HILL, A. M., SINARS, D. M. & LODGE, D. M. (1993): Invasion of an occupied niche by the crayfish *Orconectes rusticus*: potential importance of growth and mortality. – *Oecologia* **94**: 303–306.
- HOLDICH, D. M. (1991): The native crayfish and threats to its existence. – *Br. Wildl.* **2**: 141–161.
- HUXLEY, T. H. (1880): The crayfish. An introduction to the study of zoology. – Kegan Paul, London, U. K.
- HYNES, H. B. N. (1970): The ecology of running waters. – Liverpool University Press, Liverpool, U. K.
- ILHÉU, M. & BERNARDO, J. M. (1993): Experimental evaluation of food preferences of red swamp crayfish, *Procambarus clarkii*: vegetal versus animal. – *Freshwat. Crayfish* **9**: 359–364.
- (1995): Trophic ecology of red swamp crayfish *Procambarus clarkii* (Girard) – preferences and digestibility of plant foods. – *Freshwat. Crayfish* **10**: 132–139.
- KREBS, J. R. (1978): Optimal foraging: decision rules for predators. – In: KREBS, J. R. & DAVIES, N. B. (eds): Behavioural ecology. – Blackwell, Oxford, U. K., pp. 23–63.
- LAURENT, P. J. (1988): *Austropotamobius pallipes* and *A. torrentium*, with observations on their interactions with other species in Europe. – In: HOLDICH, D. M. & LOWERY, R. S. (eds): Freshwater crayfish: biology, management and exploitation. – Chapman & Hall (Croom Helm), London, U. K., pp. 341–364.
- LAWTON, J. H. (1989): Food webs. – In: CHERRET, J. M. (ed.): Ecological concepts. – Blackwell, Oxford, U. K., pp. 43–78.
- LOCKWOOD III, J. R. (1998): On the statistical analysis of multiple-choice feeding preference experiments. – *Oecologia* **116**: 475–481.
- LODGE, D. M. (1991): Herbivory on freshwater macrophytes. – *Aquat. Bot.* **41**: 195–224.
- LODGE, D. M., CRONIN, G., VAN DONK, E. & FROELICH, A. J. (1998): Impact of herbivory on plant standing crop: comparisons among biomes, between vascular and nonvascular plants, and among freshwater herbivore taxa. – In: JEPPESEN, E., SØNDERGAAED, M. & CHRISTOFFERSON, K. (eds.): The structuring role of submerged macrophytes in lakes. – Springer-Verlag, New York, USA, pp. 149–174.
- LODGE, D. M. & HILL, A. M. (1994): Factors governing species composition, population size, and productivity of cool-water crayfishes. – *Nordic J. Freshwat. Res.* **69**: 111–136.
- LODGE, D. M., KERSHNER, M. W., ALOI, J. E. & COVICH, A. P. (1994): Effects of omnivorous crayfish (*Orconectes rusticus*) on a freshwater littoral food web. – *Ecology* **75**: 1265–1281.
- LODGE, D. M. & LORMAN, J. G. (1987): Reductions in submerged macrophyte biomass and species richness by crayfish *Orconectes rusticus*. – *Can. J. Fish Aquat. Sci.* **44**: 591–597.
- MATTHEWS, M. A., REYNOLDS, J. D. & KEATINGE, M. J. (1993): Macrophyte reduction and benthic community alteration by the crayfish *Austropotamobius pallipes* (Lereboullet). – *Freshwat. Crayfish* **9**: 289–299.

- MOMOT, W. T. (1995): Redefining the role of crayfish in aquatic ecosystems. – *Rev. Fish Sci.* **3**: 33–63.
- MOMOT, W. T., GOWING, H. & JONES, P. D. (1978): The dynamics of crayfish and their role in ecosystems. – *Amer. Midl. Nat.* **99**: 10–35.
- NYSTRÖM, P. (1999): Ecological impact of introduced and native crayfish on freshwater community: European perspectives. – In: GHERARDI, F. & HOLDICH, D. M. (eds): Crayfish in Europe as alien species. – A. A. BALKEMA, Rotterdam, Brookfield, pp. 63–85.
- (2002): Ecology. – In: HOLDICH, D. M. (ed.): Biology of freshwater crayfish. – Blackwell, Oxford, U. K., pp. 192–235.
- NYSTRÖM, P., BRÖNMARK, C. & GRANÉLI, W. (1996): Patterns in benthic food webs: a role for omnivorous crayfish? – *Freshwat. Biol.* **36**: 631–646.
- NYSTRÖM, P. & STRAND, J. A. (1996): Grazing by a native and an exotic crayfish on aquatic macrophytes. – *Freshwat. Biol.* **36**: 673–682.
- O'BRIEN, B. G. (1995): The natural diet of the freshwater crayfish *Cherax tenuimanus* (Smith, 1912) (Decapoda: Parastacidae) as determined by gut content analysis. – *Freshwat. Crayfish* **10**: 151–162.
- PAINE, R. T. (1966): Food web complexity and species diversity. – *Amer. Nat.* **100**: 65–75.
- PIMM, S. L. (1982): Food webs. – Chapman and Hall, London, U. K.
- POLIS, G. A. (1991): Complex trophic interactions in deserts: an empirical critique of food web theory. – *Amer. Nat.* **138**: 123–155.
- POLIS, G. A. & STRONG, D. R. (1996): Food web complexity and community dynamics. – *Amer. Nat.* **147**: 813–846.
- RENAI, B. & GHERARDI, F. (2004): Predatory efficiency of crayfish: comparison between indigenous and non-indigenous species. – *Biol. Invasions* **6**: 89–99.
- REYNOLDS, J. D. (1978): Crayfish ecology in Ireland. – *Freshwat. Crayfish* **4**: 215–220.
- RICE, D. L. (1982): The detritus nitrogen problem: new observations and perspectives from organic chemistry. – *Mar. Ecol. Prog. Ser.* **9**: 153–162.
- SAFFRAN, K. A. & BARTON, D. R. (1993): Trophic ecology of *Orconectes propinquus* (Girard) in Georgian Bay (Ontario, Canada). – *Freshwat. Crayfish* **9**: 350–354.
- SCHOENER, T. W. (1971): Theory of feeding strategies. – *Ann. Rev. Ecol. Syst.* **11**: 369–404.
- SIEGEL, S. & CASTELLAN, N. J., Jr. (1988): Nonparametric statistics for the behavioral sciences. – McGraw-Hill International Editions, Singapore.
- USIO, N. (2000): Effects of crayfish on leaf processing and invertebrate colonisation of leaves in a headwater stream: decoupling of a trophic cascade. – *Oecologia* **124**: 608–614.
- VILLANELLI, F. & GHERARDI, F. (1998): Breeding in the crayfish, *Austropotamobius pallipes*: mating patterns, mate choice and intermale competition. – *Freshwat. Biol.* **40**: 305–315.
- WHITLEDGE, G. W. & RABENI, C. F. (1997): Energy sources and ecological role of crayfishes in an Ozark stream: insights from stable isotopes and gut analysis. – *Can. J. Fish. Aquat. Sci.* **54**: 2555–2563.
- ZAR, J. H. (1984): Biostatistical Analysis. – Prentice-Hall Inc., Englewood Cliffs.