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Diet of *Dinocras cephalotes* and *Perla marginata* (Plecoptera, Perlidae) in an Apenninic stream (Orbarina, NW Italy)

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Abstract

Feeding habits of *Perla marginata* and *Dinocras cephalotes* (Plecoptera, Perlidae) nymphs have been investigated in the Rio Orbarina (NW Italy). These species are among the largest European carnivorous freshwater invertebrates and they play an important role in the trophic structure of little Apenninic fishless streams. In this study, we examined the gut contents of 60 *P. marginata* and 60 *D. cephalotes* nymphs. Aim of this study was to characterize the diet and to investigate the existence of feeding differences between the two species. The diet of both these predaceous stoneflies included vegetal detritus, mainly in the smaller instars. We detected evident trophic preferences in the diet of both species. In particular, few taxa of preys constituted the greater part of the item ingested, independently from their availability in the substratum. Interestingly, we also detected no clear differences in prey selection between nymphs of the two species.

Introduction

It is well known that aquatic insects predominate in the trophic structure of streams (Wallace et al. 1987; Allan 1995) and in the last decades there was a growing attention to the study of their feeding habits (Tierno de Figueroa and Sanchez-Ortega 1999; Monakov 2003). Plecoptera is one of the important and often dominant orders in stream ecosystems (Zwick 2000). Feeding habits of stonefly larvae is varied, as reflected by the great variation in the structure of the mouth parts of different Plecoptera species. Detritivorous-shredders Plecoptera are an interesting subject of study to investigate leaf break-down and allochtonous input process in lotic systems (Gessner et al. 1999; Fenoglio et al. 2005*a*), while predaceous stoneflies represent a good model for predatory-prey studies in streams (Tikkanen et al. 1997). In many small, fishless streams, Plecoptera belonging to the suborder Systellognatha are the dominant predator group, with an important role as top-down control elements of invertebrate communities (Wipfli and Gregovich 2002) and also as 'ecological engineers' (Zanetell and Peckarsky 1996). For this reason, in the last years many studies investigated the trophic roles of these organisms both in the field (Bo and Fenoglio 2005; Fenoglio et al. 2005*b*) and in laboratory conditions (Elliott 2004). In field studies, gut content analysis is the most used technique: this procedure is based on the assessment of undigested and sclerotized prey parts in the stomach of the predators and on the comparison between prey eaten and prey availability in the natural environment. The 'Optimal foraging theory' (Krebs 1978) states that predators include the most profitable prey in their diet on the basis of different elements, such as energy contents, encounter rate, prey density, handling time and others. Interestingly, among Perlidae, some species seems to be selective, feeding mainly on some selected items (Fenoglio and Bo 2004) while other seems to be more opportunistic (Dudgeon 2000). In this study we examined the diet of the immature stages of two Perlidae species: *Perla marginata* (Panzer 1799) and *Dinocras cephalotes* (Curtis, 1827). These two species attain a final size

that places them among the largest European carnivorous freshwater invertebrates and play an important role in the trophic structure of little Apenninic fishless streams. Aim of this study was to analyse the diet of *P. marginata* and *D. cephalotes* nymphs of different size, testing the hypothesis that these organisms have feeding preferences and investigating the existence of some differences in the diet of these species.

Materials and Methods

In the day 24-25 October 2005, *D. cephalotes* and *P. marginata* nymphs were collected in the Orbarina creek (570 m a.s.l.). This II° order stream is a tipical Apenninic lotic system (Regional Natural Park of Beigua), with good environmental quality, reaching First Class in the Italian Extended Biotic Index (I.B.E. - Ghetti 1997), corresponding to an environment without trace of human-inducted alteration.

We collected and examined 60 *D. cephalotes* and 60 *P. marginata* nymphs, collected in a 200-m riffle. All samplings were conducted early in the morning, because Systellognatha are considered to be chiefly nocturnal feeders (Vaught and Stewart 1974). Moreover, using a Surber net (20 x 20 cm; mesh 255 μm), we collected samples in the same reach to assess the presence and abundance of the taxa of the natural benthic invertebrate population. Samples were preserved in 95% ethanol. In the laboratory, all organisms were counted and identified to genus level, except for Lumbriculidae and early instars of some Diptera and Trichoptera, which were identified to family level. Total length of *P. marginata* and *D. cephalotes* nymphs was measured (0.1 mm accuracy). Nymphs were later processed to assess food consumption by means of gut contents analysis. Guts were removed and the contents of the alimentary canal were analysed by the transparency method for slides (Faure's fluid). Identification of prey was based on sclerotized body parts, particularly head capsules, mouthparts and leg

fragments. Stewart and Stark (2002) stated that the count of sclerotized fragments (i.e.: head capsules or legs) can give a reasonably accurate count of prey consumed. Each head was counted as an individual, so that was performed for every leg of the same type (for example first right leg of a mayfly). Gut contents were also compared with the natural composition and abundance of macroinvertebrate communities in the riverbed.

Moreover, to analyse the presence of dimensional shift in food preference, we separated nymphs in three length classes (*P. marginata*; smaller ones < 5.0 mm, n = 15, intermediate 5-15 mm, n = 21; larger ones >15 mm ; n = 24; *D. cephalotes*; smaller ones < 5.0 mm, n = 9, intermediate 5-15 mm, n = 30; larger ones >15 mm ; n = 21).

Feeding preferences were quantified using the trophic electivity index of Ivlev (1961):

$$E = (ri-pi)/(ri+pi).$$

where ri = the proportion of ingested species and pi = the relative abundance in the benthic community. This index ranges from −1 to 1. A value of −1 means total avoidance, 1 indicates preference and 0 indicates indifference.

Feeding preferences were also quantified using another electivity index (McCormick 1991):

$$E^* = (W_i-1 / N)/W_i+1 / N$$

where

$$W_i = (r_i / p_i)/\Sigma r_i / p_i$$

In this formula, r_i = the proportion of ingested species, p_i = the relative abundance in the benthic community, and N = the number of food items. Both indexes ranges from −1.0 to 1.0. A value of −1.0 means total avoidance, 1.0 indicates preference and 0 indicates indifference. The analysis of variance (ANOVA) was performed with Systat 8.0 (Wilkinson 1992).

Results

In total we collected 835 stream invertebrates, belonging to 44 taxa (see Table 1). We examined gut contents of 60 *D. cephalotes* and 60 *P. marginata* nymphs. Most of the food ingested consisted in insect larvae, but algae and vegetal fragments were found in a notable number of specimens of both species.

Concerning the overall gut analysis, we detected not significant differences in the number of elements ingested (ANOVA $F_{1,118} = 0.034$, $p = 0.56$.) and prey categories (ANOVA $F_{1,118} = 1.12$, $p = 0.29$) between the two species.

Considering the difficulties related to quantify and compare the detritus consumption, the statistical analysis was performed only on prey ingestion data. Considering the mean presence of prey categories in guts, we detected 1.13 ± 1.15 SD (min = 0, max = 5, n = 60) prey items in *D. cephalotes* and 0.92 ± 1.08 SD (min = 0, max = 4, n = 60) prey items in *P. marginata* guts. Comparing the amount of elements of each prey type found in the guts, no significant differences were found between the two stoneflies species considering the three size groups (ANOVA small nymphs $F_{1,22} = 0.18$, $p = 0.67$, intermediate nymphs $F_{1,49} = 0.065$, $p = 0.80$; large nymphs $F_{1,43} = 0.25$, $p = 0.62$).

Comparing the diet with the composition of bottom community, the application of Ivlev's and McCormick's electivity indexes revealed some interesting elements. Chironomidae were the most selected prey for both species: larvae of this family were found in the 28.3 % of *P. marginata* and in 25.0 % of *D. cephalotes* nymphs and both indexes indicated an high preference (Figs. 1 and Table 2). The second most abundant item in the guts were Ephemeroptera parts: undetermined mayflies, Baetidae and Leptophlebiidae were found in the 33.3 % of *P. marginata* and in 26.7 % of *D. cephalotes* nymphs. Interestingly, some taxa were abundant and widespread on the natural environment but they were less represented in the diet

of the two stoneflies nymphs: in particular, large organisms such as Rhyacophilidae, Philopotamidae and Odonata or organisms living in particular microhabitats, such as aquatic Hemiptera. Vegetal detritus was found in 35 % of *Perla* and in 40 % of *Dinocras* nymphs. We detected not significant difference in the presence of detritus between the two species (ANOVA $F_{1,118} = 0.034$, $p = 0.61$). Considering the dimension groups, smaller nymphs showed larger amounts of this element than larger ones, also if the presence of vegetal detritus was not significantly different among nymphs of different size, both in *Perla* (ANOVA $F_{2,57} = 2.39$, $p = 0.06$) and in *Dinocras* (ANOVA $F_{2,57} = 0.14$, $p = 0.29$) nymphs.

Discussion

Recent studies demonstrated that many Perlidae are not strictly carnivorous but, especially in their first instars, they integrate their diet with vegetal detritus and algae (Gray and Ward 1979; Fenoglio 2003). According to these findings, we detected that in the Rio Orbarina, *D. cephalotes* and *P. marginata* feed on vegetal detritus: interestingly, also if this item is most abundant in younger nymphs, also larger ones consume significant amount of detritus.

Also if related to a single sampling campaign, we think that this results could be of general interest, because of the investigated species are semivoltine and analysing different size groups we collected data covering the entire post-embrionic development of the two species. Comparing the diet with the composition of the natural community, our study indicates that carnivorous stoneflies prefer small to large prey and sedentary to mobile prey (Allan and Flecker 1988). Different elements can play an important role in this contest: for example, dimensions (e.g. in the case of Trichoptera Philopotamidae) and/or predaceous habits ability (e.g in the case of Odonata and Diptera Athericidae) maybe increase the handling time and discourage attacks and, moreover, mobile preys can easily avoid attacks. On the other hand, it

is well known that encounter rate is one of the most important elements influencing predator-prey interactions in aquatic invertebrates (Sih 1993; Tikkanen et al. 1997): the more common insects are not consumed at a greater rate, but some organisms, also if well represented in the environment, inhabit particular microhabitats and are almost absent in the diet of the two species studies.

Elliott (2000, 2003), comparing gut contents of four large Systellognatha (*D. cephalotes*, *P. bipunctata*, *Perlodes microcephalus* and *Isoperla grammatica*), stated that these species are active night predators, with no clear differences in prey selection among them. This can be particularly evident in environments with rich and abundant prey communities.

Our study confirms this finding, providing evidence that large carnivorous stoneflies could have common characteristics in prey election: they seem to prefer some preys, such as Chironomidae and different families of Ephemeroptera. This overlapping in the diet could be related to the adoption of similar hunting strategies (sit-and-wait ambush strategy, Elliott 2000, 2004) or to phylogenetic constraints, related for example to the semivoltinism of these Perlidae. In the Orbarina creek *P. marginata* and *D. cephalotes* are the larger predators, whose activity could have a top-down effect on the invertebrate community: future works could investigate if the presence of fish could change diet and feeding habits of these Systellognatha.

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Caption to Figures and Table:

- Fig. 1. Percent of guts with prey fragments, with detritus and with preys and detritus for each size class of the two species (P = *P. marginata*, D = *D. cephalotes*).

- Fig. 2. McCormick's Electivity index for the macroinvertebrate taxa in the *P. marginata* and *D. cehalotes* nymphs diet of Rio Orbarina. A value of −1.0 means total avoidance, 1.0 indicates preference and 0 indicates indifference.

- Table 1: Percent relative abundance (% value in the community) for macroinvertebrates collected in the natural riverbed in the Orbarina Stream (NW Italy). (*) FFG: functional feeding groups (Cg=collectors-gatherers; F=filterers; P=predators; Sc=scrapers; Sh=shredders. See Merritt and Cummins, 1996).

- Table 2: Ivlev's Index values for *P. marginata* and *D. cephalotes* nymphs.

Taxa	Relative abundance %	FFG (*)
Plecoptera		
Leuctra sp.	5,75	Sh
Nemoura sp.	1,56	Sh
Amphinemura sp.	1,20	Sh
Protonemura sp.	0,36	Sh
Isoperla sp.	2,51	P
Perla marginata	7,19	P
Dinocras cephalotes	7,19	P
Ephemeroptera		
Ecdyonurus sp.	4,31	Sc
Torleya major	2,75	Cg
Paraleptophlebia sp.	3,95	Cg
Epeorus sylvicola	8,26	Sc
Baetis sp.	8,38	Cg
Ephemera danica	0,12	Cg
Trichoptera		
Tinodes sp.	0,12	Sh
Sericostoma sp.	7,31	Sh
Hydropsyche sp.	8,02	F
Philopotamus sp.	6,71	F
Hyporhyacophila sp.	0,96	P
Rhyacophila sp.	0,36	P
Odontocerum albicorne	0,48	Sh
Photamophilax cingulatus	0,36	Sh
Limnephilidae	0,12	Sh
Diptera		
Chironomidae	4,07	Cg
Atherix sp.	3,71	P
Tipula sp.	0,60	Sh
Limoniidae	0,24	P
Simuliidae	0,24	F
Odonata		
Onychogomphus forcipatus	0,48	P
Boyeria irene	0,36	P
Coleoptera		
Gyrinidae (larvae)	0,12	P
Elmidae (adults)	0,48	Cg
Haenydra truncata	2,16	Sc
Scirtidae (larvae)	2,16	Sh
Pomatinus substriatum	1,32	Sh
Heteroptera		
Hydrometra stagnorum	0,12	P
Micronecta sp.	0,48	P
Velia sp.	0,12	P
Gerridae	1,32	P
Annelida		
Lumbricidae	0,12	Cg
Lumbriculidae	0,48	Cg
Tricladida		
Dugesia sp.	2,16	P
Gastropoda		
Ancylus fluviatilis	0,24	Sc
Nematomorpha		
Gordius sp.	0,36	P
Arachnida		
Hydracarina	0,72	P

	P. marginata	*D. cephalotes*
Hydropsychidae	-0,82	-0,40
Philopotamidae	-0,79	0,20
other Plecoptera	-0,62	-0,49
Leptophlebiidae	-0,59	0,00
other Trichoptera	-0,35	0,01
Leuctra sp.	0,00	-0,67
Heptageniidae	0,00	-0,83
Limnephilidae	0,00	0,40
Rhyacophilidae	0,00	-0,07
Scirtidae	0,00	-0,31
Hydraenidae	0,00	-0,31
Coleoptera	0,00	-0,25
Baetis sp.	0,16	-0,11
other Ephemeroptera	0,39	0,55
Chironomidae	0,82	0,72

Tab. 2

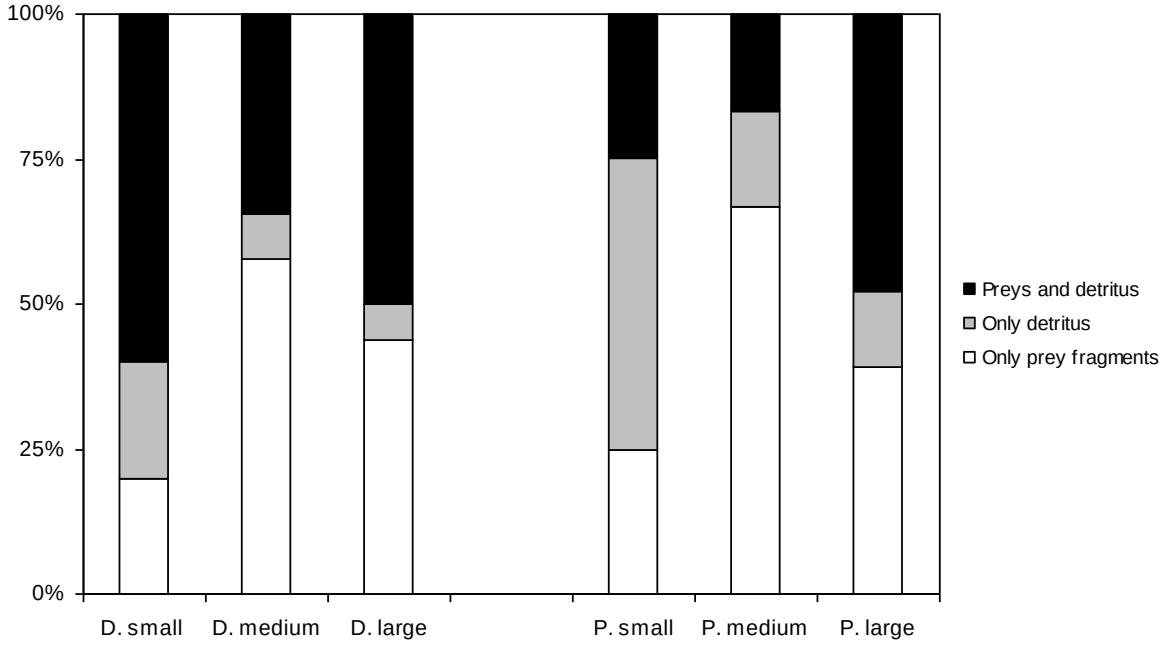
100%
75%
50%
25%
0%
D. small
D. medium
D. large
P. small
P. medium
P. large
Preys and detritus
Only detritus
Only prey fragments

Fig. 1

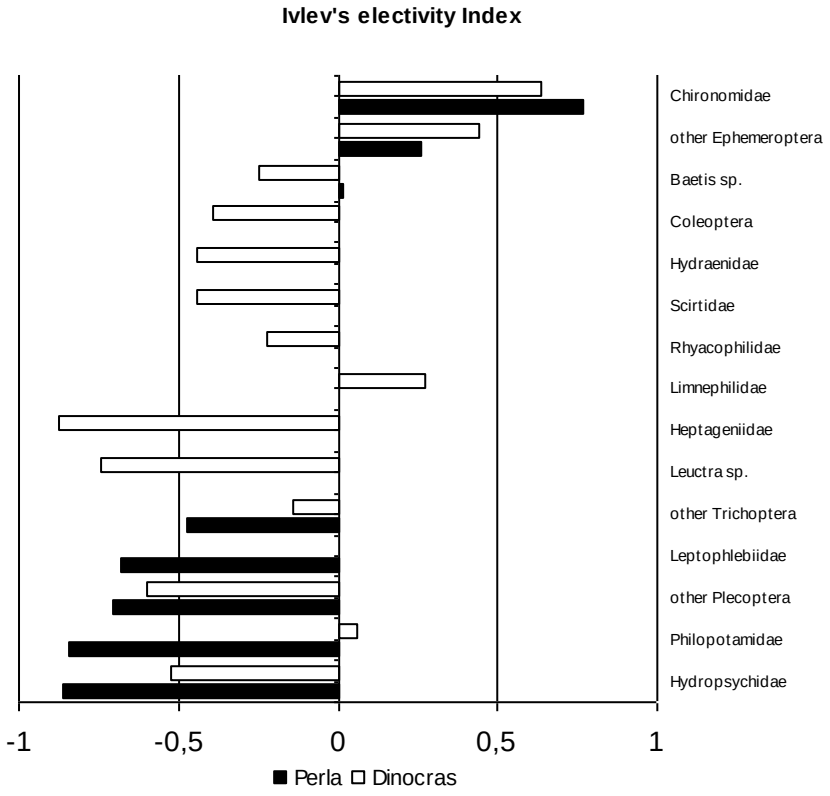
Ivlev's electivity Index
Chironomidae
other Ephemeroptera
Baetis sp.
Coleoptera
Hydraenidae
Scirtidae
Rhyacophilidae
Limnephilidae
Heptageniidae
Leuctra sp.
other Trichoptera
Leptophlebiidae
other Plecoptera
Philopotamidae
Hydropsychidae
-1
-0,5
0
0,5
1
Perla
Dinocras

Fig. 2