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PREY SELECTION OF *PERLA GRANDIS* (RAMBUR, 1841) NYMPHS (PLECOPTERA: PERLIDAE) IN BORBERA STREAM (NW, ITALY)

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Abstract: Feeding habits of *Perla grandis* nymphs have been investigated in the Borbera stream, an Apenninic water course of Northwestern Italy. In this study, we analyzed gut contents of 80 nymphs of this species, with the aim of detecting feeding preferences. Nymphs were collected from a single riffle, whose benthic coenosis was also determined. We detected the existence of an evident trophic selection: diet was almost entirely dominated by Chironomidae, independently from their availability on the substrate. This finding is discussed and compared with data previously obtained for this species from other populations.
Key words: Plecoptera, Perlidae, stoneflies, *Perla grandis*, gut contents, Ivlev's index, McCormick's index, North-west Italy, Apennine.

Selección de presa por parte de las ninfas de *Perla grandis* (Rambur, 1841) (Plecoptera: Perlidae) en el río Borbera (NO, Italia)
Resumen: Se investigaron los hábitos alimenticios de las ninfas de *Perla grandis* en el río Borbera, un curso fluvial de los Apeninos del norte de Italia. En este estudio, analizamos los contenidos digestivos de 80 ninfas de esta especie con la finalidad de detectar las preferencias alimenticias. Las ninfas fueron colectadas en un sólo tramo del río, cuya cenosis bentónica fue también determinada. Detectamos la existencia de una evidente selección trófica: la dieta estuvo casi totalmente dominada por Chironomidae, independientemente de su disponibilidad en el substrato. Se discuten estos resultados y se comparan con los previamente obtenidos para esta especie en otras poblaciones.
Palabras clave: Plecoptera, Perlidae, *Perla grandis*, contenidos digestivos, índice de Ivlev, índice de McCormick, Noroeste de Italia, Apeninos.

Introduction

Many recent papers studying stonefly feeding habits (e.g. Fenoglio & Bo, 2004; López-Rodríguez & Tierno de Figueroa, 2006) have supported the previously defended idea that diet of an unstudied species cannot be inferred or deduced from the placement of a genus or higher taxon in a generalized grouping based on studies of congeners (Stewart & Stark, 1993). Moreover, it is not enough to study the diet of a species in one particular place and time for getting a complete understanding of this aspect of its biology. Nevertheless, regarding feeding habits, only a few number of Plecoptera species has been analyzed in more than one stream. This is the case of *Perla grandis* (Rambur, 1841) (Fig. 1), studied in different parts of Europe (Berthélemy & Lahoud, 1981) and, particularly, in Northwestern Italy (Fenoglio *et al.,* 2007). *P. grandis*, previously known as *Perla maxima* (Scopoli, 1763), is widely distributed by Central and Southern Europe (Tierno de Figueroa *et al.,* 2003). In particular, it occupies a great part of continental and peninsular Italy, and can be assigned to the hyporrhitral habitat (Consiglio, 1980).

Thus, the aims of our study are: i) to describe the diet of this species in the studied stream, ii) to analyze the possible existence of prey selection, and iii) to compare our results with those previously known for other Italian population.

Materials and methods

In five dates during October and November 2006, 80 *P. grandis* nymphs (representing a wide size range) were collected in a 200 m uniform riffle of the Borbera stream (Borghetto Borbera, Fig. 2). This water course belongs to the Po basin. In the sampling station, Borbera stream is a typical Apennine lotic environment, characterized by moderate slope, width bigger than 100 m, gravel-riverbed, and fast-swallow flowing waters. In the sampling station, Borbera stream is characterized also by high environmental heterogeneity. Riparian vegetation is mainly constituted by *Salix alba* Linnaeus, 1753, *Populus nigra* Linnaeus, 1753, *Robinia pseudoacacia* Linnaeus, 1753, *Crataegus oxyacantha* Linnaeus, 1753 and *Sambucus nigra* Linnaeus, 1753.

Borbera stream shows a good environmental quality, without trace of human-inducted alteration (I class in the Italian Extended Biotic Index, *sensu* Ghetti, 1997). Main environmental characteristics and abiotic parameters (measured by Eijkelkamp 18.28 portable instruments) are reported in Table I.

Samplings were carried out early in the morning, because Systellognatha are considered to be chiefly nocturnal feeders (Vaught & Stewart, 1974). Using a Surber net (25 x 25 cm; mesh 250 μm), we collected a total amount of 25 samples to assess the presence and abundance of the taxa of the natural benthic invertebrate population. For getting a higher number of *P. grandis* nymphs, qualitative samples were taken with a Kick net (mesh 250 μm). Samples were preserved in 90% ethanol. In the laboratory, all organisms were counted and identified to the highest possible taxonomical level.

Table I: Some environmental characteristics and abiotic parameters of stream reach during field sampling

Stream order	3rd
Altitude (m a.s.l.)	280
Width (m)	100 ± 30
Depth (cm)	15 ± 5
Water temperature (°C)	16.4 ± 1.5
pH	7.3 ± 0.53
Conductivity (µS/cm)	279.6 ± 7.3
O_2 (mg/l)	9.9 ± 0.78
O_2 (% sat.)	106.1 ± 5.21

P. grandis nymphs were processed to assess food consumption by means of gut content analysis. The most common method to measure patterns of differential predation by aquatic insects is the examination of relative numbers of prey remains in predator stomach contents, and the comparison of those numbers to prey density estimated from samples from the predator's habitat (Peckarsky, 1984; Peckarsky & Penton, 1989).

For the analysis of small specimens, gut content of nymphs was studied using a transparency technique described by Bello & Cabrera (1999) which has also usually been used to study feeding in stoneflies (e.g. Bo & Fenoglio, 2005; López-Rodríguez & Tierno de Figueroa, 2006). We used Hertwig's liquid, a modification of Hoyer's liquid, which clears the body wall, thus allowing direct examination of the gut content without dissection. To examine larger specimens we removed guts and the contents of the alimentary canal were analysed by using the transparency method for slides. Fragments of animal prey were identified to the lowest taxonomic level possible. Identification of prey was based on sclerotized body parts, particularly head capsules, mouthparts and leg fragments. We also differentiated three fractions: algae, FPOM (fine particulate organic matter) and CPOM (coarse particulate organic matter).

During the laboratory phase of the study we use a NIKON SMZ 1500 light microscope (60-100 x) with JVC TK-C701EG videocamera.

To investigate the existence of feeding preferences, gut contents were compared with the natural composition and abundance of macroinvertebrate communities in the riverbed using the trophic electivity index of Ivlev (1961):

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

where ri = relative abundance of a particular taxon in the diet and pi = relative abundance of the same taxon in the benthic community. The formula considers the number of taxa (i) found in the diet. The 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):

$$E^* = (W_i\text{-}1 / N)/W_i+1 / N$$

where

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

In this formula, r_i = proportion of ingested species, p_i = relative abundance in the benthic community, and N = number of food items. This index also ranges from −1 to 1. Again, a value of −1 means total avoidance, 1 indicates preference and 0 indicates indifference.

Table II. Macroinvertebrate community of the sampled station

Taxa		N	Density (ind/m²)
Plecoptera			
Perlidae	*Perla grandis*	5	3,20
Perlodidae	*Isoperla grammatica*	28	17,92
Chloroperlidae	*Chloroperla susemicheli*	15	9,60
Leuctridae	*Leuctra* sp.	3	1,92
Ephemeroptera			
Baetidae	*Baetis* sp.	69	445,44
	Centroptilum luteolum	17	10,88
Caenidae	*Caenis* sp.	9	5,76
Leptophlebiidae	*Habroleptoides* sp.	9	5,76
Heptagennidae	*Ecdyonurus* sp.	26	169,6
	Rhythrogena sp.	13	88,96
Trichoptera			
Hydropsychidae	*Hydropsyche* sp.	11	72,32
	Cheumatopsyche lepida	54	34,56
Diptera			
Anthomyidae		1	0,64
Ceratopogonidae		1	0,64
Chironomidae		18	120,32
Tanypodinae		6	3,84
Simuliidae		11	7,04
Tipulidae	*Tipula* sp.	1	0,64
Athericidae	*Atherix* sp.	11	7,04
Limoniidae		15	9,60
Tabanidae		1	0,64
Coleoptera			
Elmidae	larvae	45	28,80
Elmidae	adults	25	16,00
Dytiscidae		3	1,92
Dryopidae	*Helichus substriatus*	6	3,84
Hydraenidae		9	5,76
Helodidae		35	22,40
Hemiptera			
Corixidae	*Micronecta* sp.	1	0,64
Odonata			
Gomphidae	*Onychogomphus forcipatus*	8	5,12
Calopterygidae	*Calopteryx splendens*	1	0,64
	Calopteryx virgo	1	0,64
Annellida			
Lumbriculidae		5	3,20
Naididae		1	0,64
Lumbricidae		3	1,92
Crustacea			
Niphargidae		68	43,52
Arachnida			
Hydracarina		10	6,40

Results

A total of 1809 macroinvertebrates belonging to 36 taxa were collected and identified (Table II), with a mean of 1400 ind/m^2 , coinciding with previous data for Apennine Alessandria area (Fenoglio *et al*., 2005; Bo *et al*., 2006).

Sixty five of the total analysed *P. grandis* nymphs had some prey in the gut (Table III). Moreover animal matter, 6.25% had CPOM (coarse particulate organic matter), 3.75% algae (mainly diatoms) and 18.75% FPOM (fine particulate organic matter). The main preys in this species diet are Diptera Chironomidae, followed by Ephemeroptera *Baetis* sp., and in a lower percentage Ephemeroptera Heptageniidae and Trichoptera Hydropsychidae.

According with obtained values of Ivlev's and McCormick's indexes (Fig 3), *P. grandis* shows a positive selection for Chironomidae. Some other invertebrate groups, despite of being present in benthos, are not consumed.

1

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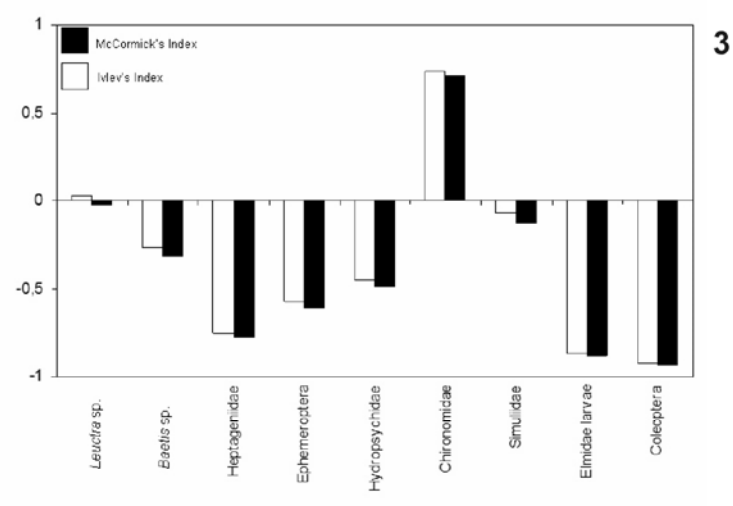
Fig. 1. *Perla grandis* nymph.

Fig. 2. Study area.



3

Fig. 3. Ivlev's and McCormick's electivity indexes for the macroinvertebrate taxa in the guts of *Perla grandis*.

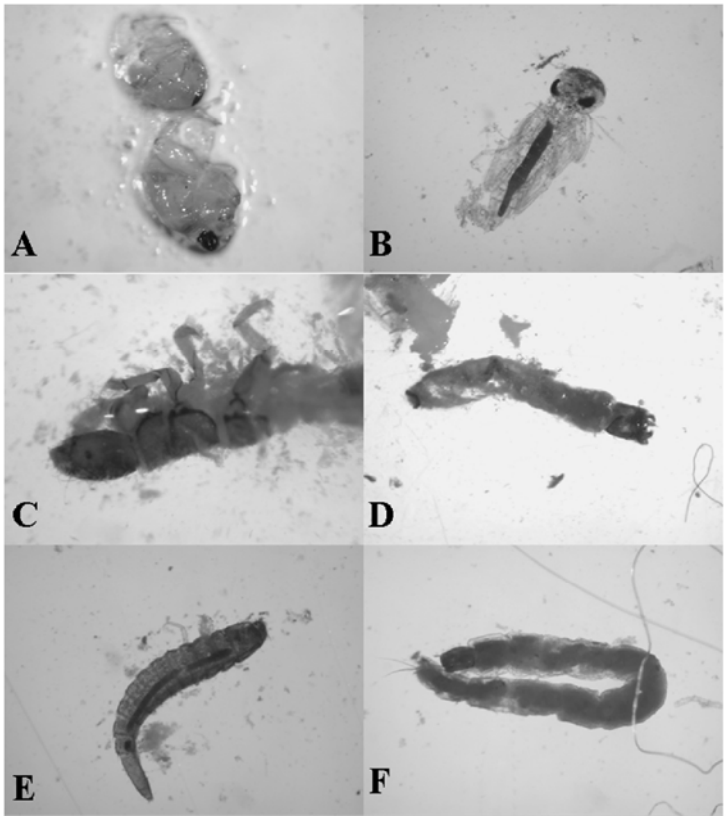


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Fig. 4. Preys found in *Perla grandis* gut:
A) *Baetis* sp.,
B) Heptageniidae,
C) Hydropsychidae,
D) Simuliidae,
E) Elmidae larva,
F) Chironomidae.

Table III. Percentages of prey found in *P. grandis* Gutsand in benthic community

Taxa	Gut contents %	Benthic community %
Plecoptera		
Leuctra sp.	0.18	0.16
Ephemeroptera		
Baetis sp.	22.61	39.51
Heptageniidae	3.18	22.38
Trichoptera		
Hydropsychidae	3.18	9.25
Diptera		
Chironomidae	69.08	10.41
Simuliidae	0.53	0.60
Coleoptera		
Elmidae larvae	0.18	2.49

Discussion

Presence of a certain quantity of vegetal matter and detritus in the guts of the studied species coincides with the general fact that Perloidea stoneflies, in spite of being mainly carnivorous, complete their diet with those kinds of food (Stewart & Stark, 1993).

It is known that big Perlidae and Perlodidae are active nocturnal predators that can feed on a wide variety of taxa (Spellman & Drinan, 2001). The optimal foraging theory (Krebs, 1978) assumes that a predator include in its diet the prey offering greater advantages, such as energetic content, density, capture facility, manipulation time, etc. As shown above, *P. grandis* is a selective predator that attacks with higher frequency some types of prey independently of its availability on substrate (Fig. 4). This is the case of Chironomidae that only suppose 10 % of the community but 70% of the consumed prey. Chironomidae are for *P. grandis*, as for other Perlidae (e.g. Berthélemy & Lahoud, 1981; Bo & Fenoglio, 2005), the main prey. These Diptera have many characteristics that probably make them particularly attractive: intermediate size, preference for places with no very fast flow, soft body without developed protection structures, and scarce movement capacity. Furthermore, although Ephemeroptera Heptageniidae (*Ecdyonurus* sp. and *Rhithrogena* sp.) are an important component of the benthic community in terms of number of individuals in Borbera stream, they are rarely predated. That could be a consequence of being rheophile organisms living in particular microhabitat characterized by very fast water flow and usually shallow, while *P. grandis* prefers, according with its size, spaces among riverbed stones.

Finally, comparing the diet of the studied population of *P. grandis* in Bordera stream with that of the same species in a close first order stream (Rio Berga) (Fenoglio *et al*., 2007), and despite of the different characteristic of the fluvial course, we found the same preference for preying on Chironomidae, being the electivity index value for this taxa higher than 0.7 in both cases.

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