

Feeding habits of Perlodidae (Plecoptera) in the hyporheic habitats of Alpine streams (Trentino-NE Italy)

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Total of 75 nymphs of *Dictyogenus fontium*, *Perlodes intricatus* and *Isoperla* (cf. *rivulorum*) and 22 juvenile Perlodinae were collected in hyporheic habitats of the Noce Bianco watershed (Trentino, NE Italy). The identification of their life stages allowed considerations regarding their presence in the hyporheos, mainly used as a refuge habitat, particularly during vulnerable phases of their life cycle (neanid stage and pre-imaginal phase). This function is possible, given the more stable environmental conditions and the minor predatory pressures in this habitat. The gut content analyses of the three species assigned them to the engulfer-carnivorous functional feeding group, with slight resource partitions between the two largest species. *P. intricatus* appears to feed on less mobile and fleshy preys while *D. fontium* prefers more sclerotized and active preys. The results indicate that the hyporheos acts not only as a refuge area but also as a trophic habitat for some Perlodidae species.

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1. Introduction

The vertical expansion of the spatial limits of river ecosystems, to include subsurface zones, is crucial for a better understanding of their ecological functioning (Jones & Holmes 1996).

The hyporheic zone, i.e. the saturated interstitial areas beneath the stream bed and in the stream banks that contain some proportion of channel water (White 1993), is recognised as an integral

component of the stream (Fisher 1997). In fact streams can be viewed as perfusing the land and existing 'wherever water flows over or under the landscape'. This expanding view of lotic systems has emerged from several studies which show that streams continuously exchange water, nutrients, organic matter, and organisms with the underlying ground water (Williams 1984, Ward 1989, Harvey & Bencala 1993, Ward *et al.* 1998). Thus the hyporheic zone has become a subject of

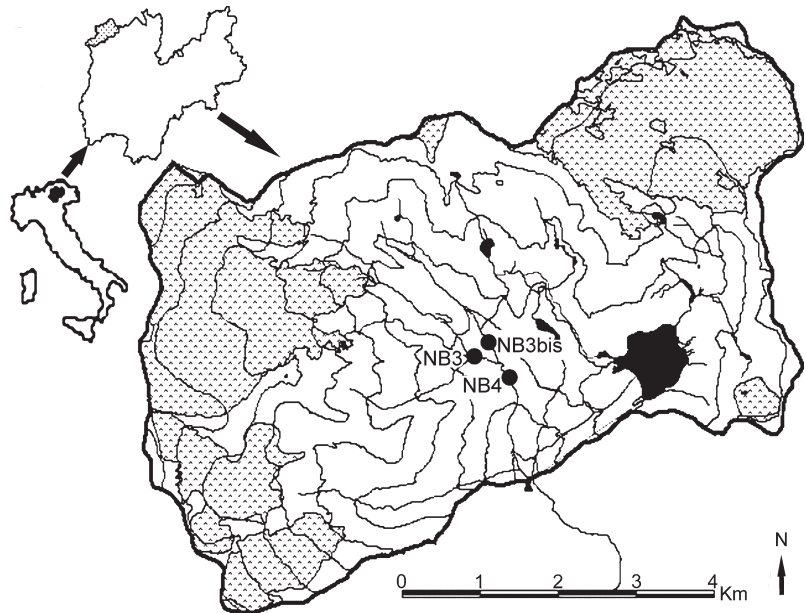


Fig. 1. Map of the study area in the Stelvio National Park (Trentino, N-E Italy).

general interest when limnologists recognize that exchange processes between surface water and ground water are key determinants of the river structure and function. Such processes are also an important element for the maintenance of high levels of diversity in macroinvertebrate cenosis, especially in high altitude streams. The importance of surface–subsurface exchanges on ecosystem processes and biodiversity in streams has been addressed recently in several papers (Boulton *et al.* 1998, Pusch *et al.* 1998, Dole-Olivier 1998, Bencala 2000, Jones & Mulholland 2000, Malard *et al.* 2000, Burgherr 2000).

In general, stonefly communities are not very diversified in Alpine streams, in response to the harsh environmental conditions there (Brittain *et al.* 2000, Ward 1994). However, the hyporheic stonefly communities in high altitude streams maintain a very good level of diversity in relation to the corresponding benthic ones (Malard *et al.* 2002). This is probably due to the use of the hyporheos as a refuge from the adverse conditions in surface water, as nursery, and as feeding area as suggested by Varricchione (1999) and Boulton (2000).

Eleven species of Perlodidae stoneflies have been cited in Italian Alpine streams (Fochetti 2004, Tierno de Figueroa & Fochetti 2001, Knispel *et al.* 2002): two species of the genus *Dictyo-*

genus, two of *Perlodes* and seven of *Isoperla*. It is usually accepted that Perlodidae nymphs are carnivorous, although some of them can eat vegetal matter during their first nymphal instars (Hynes 1976, Tachet *et al.* 2000, Monakov 2003).

Studies regarding carnivorous species allow investigation of the feeding nets that regulate the structure of stream communities (Allan 1995). Moreover, an interesting research field is the resource partition among the predaceous stonefly species (Sheldon 1980, Peckarsky 1984, Elliott 2003, Bo *et al.* 2007). In the last years the study of the feeding habits of carnivorous Plecoptera has been implemented particularly in Northern Italian streams (Bo & Fenoglio 2005, Bo *et al.* 2007, Fenoglio *et al.* 2005, 2007a, 2007b, Maiolini & Silveri 2005). This interest is due to both the important role of top down control of predatory stoneflies on the macroinvertebrate assemblages and their role as bioindicators of environmental changes, because of their top position in the food chain. Despite this, only few studies have dealt with the feeding habits of predatory species in the hyporheic habitats, especially in high altitude streams.

The aim of this work is (i) to study which species of predatory stoneflies are present in the hyporheic zone of high altitude streams, (ii) to define the age-class composition of Perlodidae liv-

Table 1. Characteristics of the sampling sites.

Station	Altitude (m a.s.l.)	Typology	Substrate	Stream
NB3	2,270	Glacial	Pebble-gravel	Noce Bianco Stream
NB3bis	2,270	Rhithral	Cobble	Larcher Stream
NB4	2,260	Glacio-rhithral	Cobble-gravel	Confluence

ing in this habitat, as indicator of the role of the hyporheic zone for each species, (iii) to analyze feeding habits of hyporheic Perlodidae and the possible existence of resource partition.

2. Material and methods

The sampling stations are located on the Noce Bianco (3rd order stream), a main tributary of the Adige River (the second longest Italian river). The Noce Bianco watershed is in De la Mare Valley (Stelvio National Park, Trentino, NE Italy, 46°N, 10°E). The three sampling sites are located in the Venezia subvalley (Fig. 1) and their characteristics are presented in Table 1.

Macroinvertebrates were collected seasonally (2003–2005) with various sampling methods: a Bou-Rouch pump inserted into the riverbed to a depth of 40 cm; bottle traps inserted in the riverbed substratum to a depth of 30 cm, and hyporheic artificial substrates inserted in the sediment to a depth of 15 cm.

Samples were preserved in 75% ethanol or in 4% formalin in the field and transported to laboratory, where nymphs were identified to the species level. For each of the three studied species, *Perlodes intricatus* (Pictet, 1841), *Dictyogenus fontium* (Ris, 1896) and *Isoperla rivulorum* (Pictet, 1842), we used measurements from specimens collected in the hyporheos and from individuals captured in the benthic layer (from a complementary study in the same area). Thus, we choose a more complete information regarding the real size range for each species. Measurements of body length (from the head to the last urite) and head dimension (length and width) were taken with an ocular micrometer microscope. All three measures were highly correlated (for *D. fontium*: Spearman $r = 0.79$ for body length vs. head length; Spearman $r = 0.82$ for body length vs. head width; Spearman $r = 0.92$ for

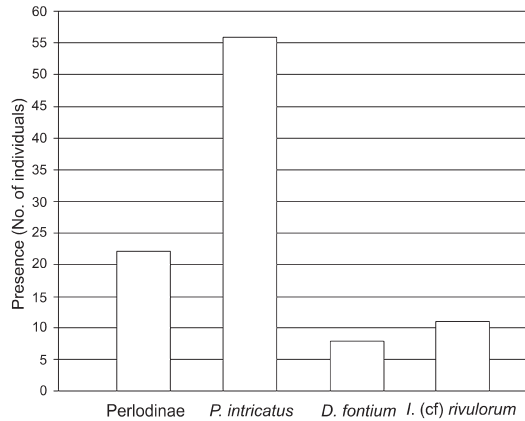


Fig. 2. Number of individuals of Perlodidae species found in the hyporheic habitat.

head length vs. head width, $N = 67$; for *I. (cf) rivulorum*: Spearman $r = 0.79$ for body length vs. head length; Spearman $r = 0.81$ for body length vs. head width; Spearman $r = 0.91$ for head length vs. head width $N = 7$ and for *P. intricatus*: Spearman $r = 0.97$ for body length vs. head length; Spearman $r = 0.96$ for body length vs. head width; Spearman $r = 0.85$ for head length vs. head width $N = 89$; all with $P < 0.05$). Thus, we choose to use only head width to define five size ranges for each species. We also considered the size of the wing pads and the wing development level to define these classes, corresponding to different stages of nymphal development as follows: I class (newly-emerged neanids), II class (neanids), III class (neanids with traces of wing pads), IV class (nymphs with well-developed wing pads) and V class (nymphs ready to emerge).

To analyze the feeding habits of the different Perlodidae nymphs, we extracted their gut contents. Preys were identified using undigested chitinous parts, especially head capsules.

3. Results and discussion

3.1. Species found in the hyporheic zone

Only three species, *D. fontium*, *P. intricatus* and *I. (cf) rivulorum*, of the eleven Perlodidae known in the Italian Alps (Fochetti 2004, Tierno de Figueroa & Fochetti 2001, Knispel *et al.* 2002) were identified in the hyporheic zone of our sampling stations (Fig. 2). This scarce biodiversity was probably due to the severe environmental conditions of high elevation freshwater ecosystems.

D. fontium is widespread in the Italian Alps, where it populates rhithral streams even at high elevations. In Trentino it has been recorded up to 2700 m.a.s.l. (Lencioni *et al.* 2002). Adults emerge from June to September (Consiglio 1980).

P. intricatus is another predatory species present in the Trentino Alps that can be classified as an orophile species of fast flowing waters (Consiglio 1979) and inhabiting the hyporhithral and rhithral zones (Consiglio 1980). The adults present a summer flight period, from June to July (Consiglio 1980).

I. rivulorum is an orophile species distributed mainly in Central Europe. It lives in rhithral habitats and its flight period extends from June to September (Consiglio 1980).

Only one other species belonging to Perloidea superfamily was found in the same habitat: *Chloroperla susemicheli* Zwick, 1967 (family Chloroperlidae) (Silveri *et al.* 2007). This is the most abundant predatory stonefly species collected in the hyporheic habitat of our stations (49%). The three species of Perlodidae, plus some non-identified juvenile Perlodidae nymphs, constitute together the remaining 51%.

No Perlidae species were found, but this fact could be related to the total absence of Perlidae in the benthos of high altitude streams (over 1800 m.a.s.l.) of Trentino (Fochetti 2004).

3.2. Relationship between nymphal size and use of hyporheos as a habitat

In Figure 3, the Perlodidae species collected in the hyporheic habitat and their classification according to the five developmental stages are

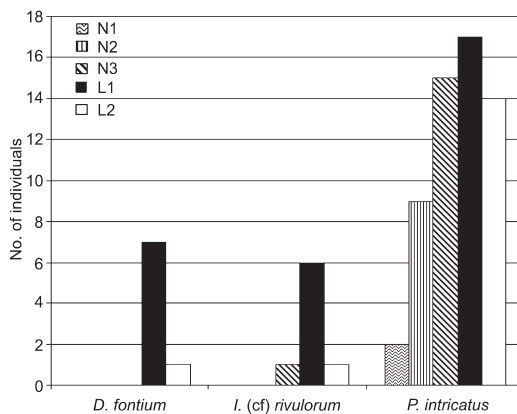


Fig. 3. Distribution of the five size classes of the nymphs of three species.

shown.

Although we found a low number of specimens of *D. fontium* and *I. (cf) rivulorum*, our results suggest that these species do not use the hyporheic habitat during the intermediate stadium of their development but only during their last nymphal instars (Fig. 3). Probably the hyporheos acts as a refuge area where the nymphs can complete their development and growth.

P. intricatus is the most abundant Perlodidae found in our sampling area (Fig. 2). Our results highlight that this species uses the hyporheic zone mainly during the central and the final phases of its life cycle (Fig. 3). However, some first instar *P. intricatus* nymphs also seem to use the hyporheic habitat (Fig. 3).

The presence of some juvenile Perlodidae (Fig. 2) together with the presence of first instar nymphs of *P. intricatus*, suggest the nursery role of hyporheic habitat.

These results support the idea that the use of the hyporheos is an important refuge habitat for some amphibiotic stonefly species (Williams 1984, Varrichione 1999, Boulton 2000, Bencala 2000), especially during the most vulnerable phases of their life cycle. This important function is due to the more stable environmental conditions and the minor predator pressure in the hyporheic zone, especially in high altitude streams. So, the hyporheic buffer seems to be an ideal area to spend the most sensitive nymphal stages (neanid stage and pre-imaginal phase) for these Perlodidae species. On the contrary, it ap-

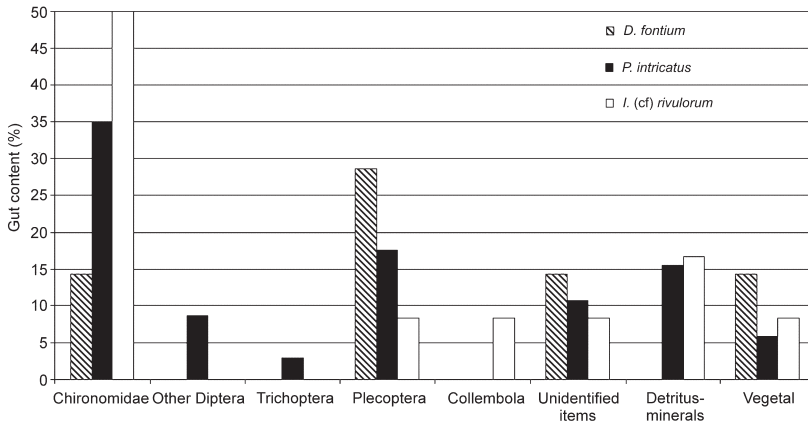


Fig. 4. Percentage of items found in the gut contents of the three studied Perlodidae species.

pears that, in the same area, *C. susemicheli* spends all its life cycle in the hyporheic area suggesting that it is a typical hyporheic species (Silveri *et al.* 2007).

3.3. Feeding habits

The gut contents of the three analyzed stonefly species are shown in Figure 4 and Tables 2, 3 and 4.

Seven of the eight studied *D. fontium* had gut contents (Table 2). Our results seem to suggest preference for preying on other stonefly species (probably belonging to Nemouridae and Perlodidae), despite they are not the most common prey in the hyporheos of high altitude streams (Milner & Petts 1994, Maiolini & Lencioni 2000), and vegetable matter. Moreover, they also prey on Chironomid larvae (the most abundant prey in the hyporheos) and other unidentified insects (mayflies or stoneflies).

D. fontium is one of the largest predatory stonefly present in Alpine streams (mature nymphs reach lengths from 15 to 25 mm, according to Consiglio, 1980), and their feeding habits have been previously studied in benthic populations, showing some differences between localities. Thus, in a study in Trentino, nymphs showed an opportunistic character preying on the more numerous taxa in the community (Maiolini & Silveri 2005), while in a study in Piemonte, Fenoglio *et al.* (2007b) reported a preference for Chironomidae and the presence of vegetal detritus and algae in the preimaginal diet.

Of the 56 studied *P. intricatus*, 52 had gut contents (Table 3). This species feeds mainly on Chironomid larvae, but also on Plecoptera (probably belonging to Perlodidae, Leuctridae and Nemouridae), other Dipterans (Limoniidae and Simuliidae) and Trichoptera. Large quantities of digested chitinous parts were found inside their guts, but their identification was not possible. Detritus and vegetal matter were also found in a large quantity. *P. intricatus* is also a large predator species (mature nymphs can grow from 15 to 25 mm long, according to Consiglio, 1980). Biological data of this species are scarce compared to those of other species of the genus, such as *Perlodes microcephalus* (Pictet, 1933), which feeds also mainly on Chironomidae and other aquatic macroinvertebrates in benthic Pyrenean populations (Berthélemy & Lahoud 1981).

All seven analyzed individuals of *I. (cf) rivulorum* had gut contents but four were severely damaged and thus gut analyses were not possible (Table 4). This species appears to feed mainly on Chironomidae larvae. Vegetable and detritus matter were also frequent in the gut. There was also one Collembola specimen among the prey, probably ingested incidentally. This species is considerably smaller than the other studied Perlodidae (mature nymphs grow from 10 to 12 mm long, according to Consiglio, 1980). There are no data on feeding habits of this species in Italy, but it is usually considered that *Isoperla* species, at least in the benthos, are mainly predators, although some species feed entirely on vegetation (Hynes 1976).

The presence of vegetal matter and detritus in

Table 2. Gut content of *D. fontium* (N=8).

Items	Mean	Min–Max	SD	Presence
Chironomidae	0.25	0.00–1.00	0.46	2 (25%)
Plecoptera	0.50	0.00–1.00	0.53	4 (50%)
Unid. prey	0.25	0.00–1.00	0.46	2 (25%)
Veg. Mat. (%)	12.50	0.00–60.00	23.75	2 (25%)

Table 3. Gut content of *P. intricatus* (N=56).

Items	Mean	Min–Max	SD	Presence
Simuliidae	0.04	0–1	0.19	2 (3.6%)
Limoniidae	0.12	0–2	0.38	6 (10.7%)
Chironomidae	4.36	0–35	6.92	36 (64.3%)
Other Diptera	0.02	0–1	0.13	1 (1.8%)
Trichoptera	0.05	0–1	0.23	3 (5.4%)
Plecoptera	0.46	0–4	0.87	18 (32.1%)
Unid. prey	0.20	0–1	0.40	11 (19.6%)
Detritus (%)	11.96	0–100	25.68	16 (28.6%)
Veg. Mat. (%)	2.41	0–65	9.53	6 (10.7%)

Table 4. Gut content of *I. (cf) rivulorum* (N=7).

Items	Mean	Min–Max	SD	Presence
Chironomidae	2.00	0–4	1.41	6 (85.7%)
Plecoptera	0.29	0–2	0.76	1 (14.3%)
Collembola	0.14	0–1	0.38	1 (14.3%)
Unid. prey	0.14	0–1	0.38	1 (14.3%)
Detritus (%)	13.75	0–6	24.27	2 (28.6%)
Veg. mat. (%)	2.86	0–20	7.56	1 (14.3%)

high quantities in the guts of these predaceous stonefly species could be related to their feeding mechanism. In fact, they can be classified as engulfers-carnivorous *sensu* Cummins & Merritt (1996) and, as pointed out by Hynes (1976), in the carnivorous stoneflies food is very often accompanied by algae and detritus.

Despite the low numbers of individuals of two of the studied species, the results indicate a slight resource partition at least between the two largest species. Thus, *P. intricatus* seems to feed on less mobile and fleshy preys (such as Chironomidae, Limoniidae, Simuliidae.) while *D. fontium* feeds mainly on more sclerotized and active preys (such as other Plecoptera). The preference of *Perlodes* (at least *P. microcephalus*) for sedentary preys has also been pointed out in a work in Great

Britain (Elliott 2000). The sharing of food resources among coexisting Perlodidae species has been previously demonstrated in benthos (e.g. Sheldon 1972). Our results suggest that the hyporheos could act not only as a refuge area but also as an important food resource for some Perlodidae species.

From a more general point of view, and regarding all the stonefly community present in the studied hyporheic area, Perlodidae species share their role as predators with the Chloroperlid species *C. susemicheli* (considerably smaller, 8 mm, according to Fochetti, 2004), which spends all its life cycle in this habitat and feeds mainly on the most common preys (Chironomidae) and more scarcely on other more active preys (Silveri *et al.* 2007).

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