

Terrace springs: habitat haven for macrobenthic fauna in the lower plain of the River Ticino (Lombardy, Northern Italy)

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Abstract - Springs are important environments between hypogean and epigeal habitats; the interaction between aquatic and terrestrial ecosystems is an important factor for their biotic communities. We investigated the ecology of the macrobenthic community of two lowland springs in the River Ticino valley, focusing on the autoecology of some relevant species and on the role of springs as hotspots of biodiversity in an area threatened by anthropogenic pressure.

We collected 26 *taxa* in total: diptera (8), trichoptera (6), gastropods (5), coleoptera (2), crustacea (2), lumbricidae (1), odonata (1), plathelminthes (1). Some of them are stenothermal and oligotrophic species thus quite unusual for a flood plain area characterized by intensive agricultural activities.

In conclusion, our study highlights the importance of lowland springs in conserving some habitat-selective macrobenthic species.

Key words: lowland springs, fluvial terrace, insular biotopes, crenic assemblages.

Riassunto - Sorgenti di terrazzo: un rifugio per la fauna macrobentonica nella bassa pianura del Ticino (Lombardia, Nord Italia).

Le sorgenti sono ambienti importanti tra habitat ipogei ed epigei; l'interazione tra ecosistemi acquatici e terrestri è un fattore importante per le loro comunità biotiche. Abbiamo studiato l'ecologia della comunità macrobentonica di due sorgenti di pianura nella valle del Ticino, concentrandosi sull'autoecologia di alcune specie rilevanti e sul ruolo delle sorgenti come punti caldi della biodiversità in un'area minacciata dalla pressione antropogenica.

Abbiamo raccolto 26 *taxa* in totale: ditteri (8), tricotteri (6), gasteropodi (5), coleotteri (2), crostacei (2), lumbricidi (1), odonata (1), platelminti (1). Alcuni di essi sono specie stenoterme e oligotrofe quindi piuttosto inusuali per una zona di pianura alluvionale caratterizzata da attività agricole intensive.

In conclusione, il nostro studio evidenzia l'importanza delle sorgenti di pianura nella conservazione di alcune specie macrobentoniche selettive dell'habitat.

Parole chiave: sorgenti di pianura, terrazza fluviale, biotopi insulari, assemblaggi crenici.

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INTRODUCTION

Springs are important environments between hypogean and epigeal habitats (Hanh, 2000; Smith *et al.*, 2003) and, despite their limited dimensions, they are composed of various microhabitats and are often characterized by high biodiversity (Weigand, 1998; Cantonati *et al.*, 2012). The interaction between aquatic and terrestrial ecosystems is important for the structure of spring habitat: shading by riparian vegetation can affect levels of autochthonous and allochthonous organic matter (Barquín & Scarsbrook, 2008) and it can also influence the temperature of spring water consequently extending habitats for cold stenothermic species (Roca & Baltanas, 1993).

In general, these biotopes are classified according to a method based on current velocity, which was developed by Steinmann (1915) and Thienemann (1922). Recently, Martin & Brunke (2012) suggested the term rheo-elocrene to classify an intermediate type of spring, which are often found on terraces, characterized by a thin film of water flowing constantly through canopy areas, with sandy fine gravel substrata (Gerecke, 1991). These habitats are also typical of forested areas with an elevated level of woody debris, leaves and mosses on the bottom (Cantonati & Bonnetini, 1995; Di Sabatino *et al.*, 2003).

Several studies have supported the importance of abiotic factors in determining the composition of the fauna and flora in springs (Erman & Erman, 1995; Hanh, 2000; Smith *et al.*, 2003). Nevertheless, one of the main characteristics that influence the compositions of spring communities is the cold stable water temperature (Illies, 1952; Erman & Erman, 1995).

In Europe, lowland eucrenic habitats were often subject of various studies (Zollhöfer *et al.*, 2000; Gerecke & Franz, 2006; Von Fumetti *et al.*, 2006; Ilmonen *et al.*, 2009; Von Fumetti & Nagen, 2012). In Italy, instead, researchers have mainly studied north-eastern Alpine and pre-Alpine springs (Crema *et al.*, 1996; Gerecke *et al.*, 1998; Stoch & Tomasin, 2002; Mezzanotte & Sambugar, 2004; Cantonati *et al.*, 2006; Sambugar *et al.*, 2006) or central and southern Apennine areas (Galassi, 1997; Cianficconi *et al.*, 1998; Di Sabatino *et al.*, 2003; Bottazzi *et al.*, 2008) while lowland springs have been neglected, apart from a few studies on plain-springs and resurgence springs (Girard, 1965; Cotta-Ramusino *et al.*, 1991; Cianficconi *et al.*, 1998; Crosa & Buffagni, 2002) and qualitative studies on

river springs of the Ticino valley (Sconfiatti *et al.*, 2009a; Pesci, 2010). These types of microhabitats were mainly investigated for water chemistry in relation to agricultural impact and meiofauna (Gerecke *et al.*, 1998; Laini *et al.*, 2011).

Therefore, the present study addresses the benthic community of two springs of the River Ticino valley (River Po plain, North of Italy), focusing on the autoecology of some relevant species and on the role of springs as hotspots of biodiversity in an area threatened by anthropogenic pressure.

METHODS

Study area

The River Ticino is one of the most important tributaries of the River Po and its Italian course is recognized as a regional park and is protected by UNESCO. The lower flood plain is characterized by intensive agricultural activities and high anthropogenic pressure, however numerous semi-natural areas are present (Sconfiatti *et al.*, 2009b).

The study area is situated near the city of Pavia (South-East Lombardy, Northern Italy), a few kilometres upstream from the River Po confluence (Fig. 1). It is located on the left side of the floodplain, along the scarp of the fluvial terrace covered by a shady forested zone, characterized by the dominance of the allochthonous species *Robinia pseudoacacia* L. and a thick underwood with *Robus caesius* L. and highly hygrophilous species, such as *Equisetum telmateia* L., *E. arvensis* L. and mosses.

Where the slope of the terrace shows a marked stair step, many permanent hydric emergences seep out from a perched aquifer, about 8 metres below the upper level of the alluvial plain, almost at the same level as the lower level of the floodplain. These water streaks converge in a few small streams, which slowly flow towards a narrow water course called Vernavola Stream. The springs are 50-100 cm wide and 20-25 m long and they have a weak discharge of less than 10 l m⁻¹ and a water speed of 20 cm s⁻¹. Their substrate is mainly composed of fine gravel,

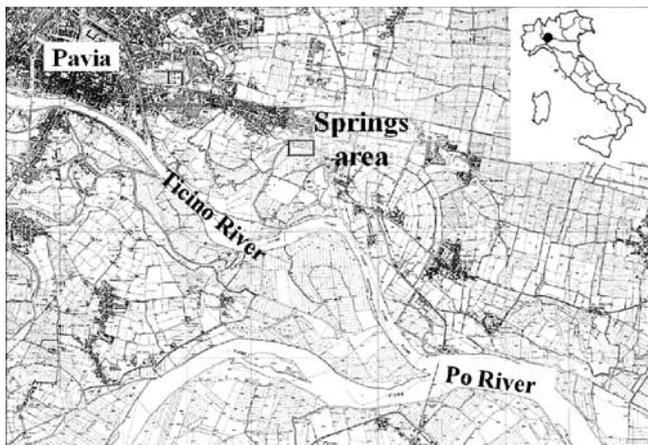


Fig. 1 - Study area.

sand and clay; the bottom is also rich in vegetal detritus. Nitrate concentration in their waters is known to range from 10 mg l⁻¹ to 13 mg l⁻¹ (Pesci, 2010).

A few species of aquatic or semi-aquatic plants were present in the spring brooks: *Lemna minor* L., *Ranunculus ficaria* L. and *Nasturtium officinale* L. We also found a large population of *Leucojum aestivum* L., a threatened protected species by the Habitat Directive 92/43 CEE.

Sample collection and data analyses

From April 2008 to March 2009, we monthly collected the macrobenthic community in two permanent terrace springs (hereafter called S1 and S2). In order to perform quantitative sampling and due to the small dimension of the springs, we used a small hand-net (950 μ mesh) with an additional aluminium frame (19.2x8.7 cm), placing the hand-net on the ground and gently moving the substrate within the aluminium frame. For all samples, we collected three replicates to cover an area of 500 cm².

Temperature was recorded using a digital thermometer DELTA OHM HD 8705, and dissolved oxygen was measured using an Idronaut Oxyliquid Oximeter probe.

Macrobenthos was sorted, identified and counted in the laboratory using a stereo microscope at 10-40x magnification, and preserved in 70 % ethanol. We identified to species level crustacea, platyhelminthes, trichoptera, odonata, lumbricidae and coleoptera; diptera and all other taxa were identified to genus or family level.

In order to compare the macrobenthic communities of the two springs and seasonal effects on the biotic assemblages, we performed an ANOSIM test on log transformed data and moreover, a SIMPER analysis was performed in order to define the most dominant species in each spring. Finally, a cluster analysis (using species as response variable) was carried out with the aim of highlighting the ecological affinity between macrobenthic species.

All the statistical analyses were performed using PRIMER 5.0 software package (Clarke and Warwick, 1994).

RESULTS

In S1, the water temperature varied from 11°C to 18.5°C, while in S2 it varied from 18.7°C to 9.8°C. Moreover, the oxygen saturation reached 90% in S1 and 94% in S2.

Considering the biotic data, we collected 26 taxa in total: 15 were identified to species level while 6 to genus level. In total, we identified: diptera (8), trichoptera (6), gastropods (5), coleoptera (2), crustacea (2), lumbricidae (1), odonata (1), plathelminthes (1) (Tab. 1).

The community in S1 reached a density of about 4000 ind. m⁻² while in S2 it had density of 2375 ind. m⁻². This difference was confirmed by the ANOSIM test (R=0.38, p<0.05); at the contrary, no statistical difference was recorded between seasons (S1, R=-0.019, p>0.05; S2, R=-0.019, p>0.05).

Furthermore, SIMPER analysis showed that the community of S1 was mainly characterized by the presence of the amphipod *Echinogammarus stammeri*, the planarian *Polycelis felina* and the caddisfly *Crunoecia irrorata*.

Tab. 1 - Taxa collected in spring S1 and S2.

Group	Taxon	Author's name	S1	S2
Crustacea	<i>Echinogammarus stammeri</i>	(Karaman, 1931)	X	X
	<i>Androniscus dentiger</i>	Verhoeff, 1908	X	X
Gastropoda	<i>Bythinella schmidtii</i>	(Küster, 1852)	X	X
	<i>Gyraulus crista</i>	(Linnaeus, 1758)	X	X
	<i>Gyraulus laevis</i>	(Alder, 1838)	X	X
	<i>Lymnaea</i> sp.		X	X
	<i>Sadleriana</i> sp.	(Küster, 1852)	X	
Coleoptera	<i>Macronychus quadrituberculatus</i>	Ph. Müller, 1806	X	X
	Helodidae		X	X
Diptera	<i>Dixa</i> sp.		X	X
	Ceratopogonidae		X	
	<i>Hexatoma</i> sp.			X
	<i>Odontomia</i> sp.		X	X
	Psycodidae		X	X
	Diamesinae		X	X
	Tanypodinae		X	X
	<i>Tipula</i> sp.			X
Oligochaeta	<i>Eiseniella tetraedra</i>	(Savigny, 1826)	X	X
Odonata	<i>Cordulegaster boltonii</i>	(Donovan, 1807)		X
Trichoptera	<i>Crunoecia irrorata</i>	(Curtis, 1834)	X	X
	<i>Ernodes articularis</i>	(Pictet, 1834)	X	X
	<i>Helicopsyche sperata</i>	McLachlan, 1876	X	X
	<i>Philopotamus montanus</i>	(Donovan, 1813)		X
	<i>Silo nigricornis</i>	(Pictet, 1834)	X	X
	<i>Wormaldia occipitalis</i>	(Pictet, 1834)	X	X
Planaria	<i>Polycelis felina</i>	(Dalyell, 1814)	X	X

Similarly, in S2, the most abundant species was *E. stammeri*, even though its density was lower compared to S1. Even here, *C. irrorata* was very abundant, as the dragonfly *Cordulegaster boltonii*, which at the contrary was absent in S1; finally, here *P. felina* was less frequent and abundant compare to S1.

Moreover, the performed analyses highlighted that the slight dissimilarity between these two communities was mainly due to the abundance of *P. felina* in S1 and the presence of *C. boltonii* in S2.

After this analysis, we compared the biotic data by means of a cluster analysis: in S1, twelve *taxa* (more than half of the total) have over 50% affinity and nine out of twelve were clustered with over 70% of similarity: *Helicopsyche sperata*, *Ernodes articularis*, Helodidae, *Silo nigricornis*, *Crunoecia irrorata*, Tanypodinae, Diamesinae, *Polycelis felina* and *Echinogammarus stammeri* (Fig. 2). In the dendrogram of S2, six out of nine *taxa* were clustered with almost 70% affinity; when a seventh taxon, *Polycelis felina*, was added to the cluster, the affinity decreased to 45% (Fig. 3).

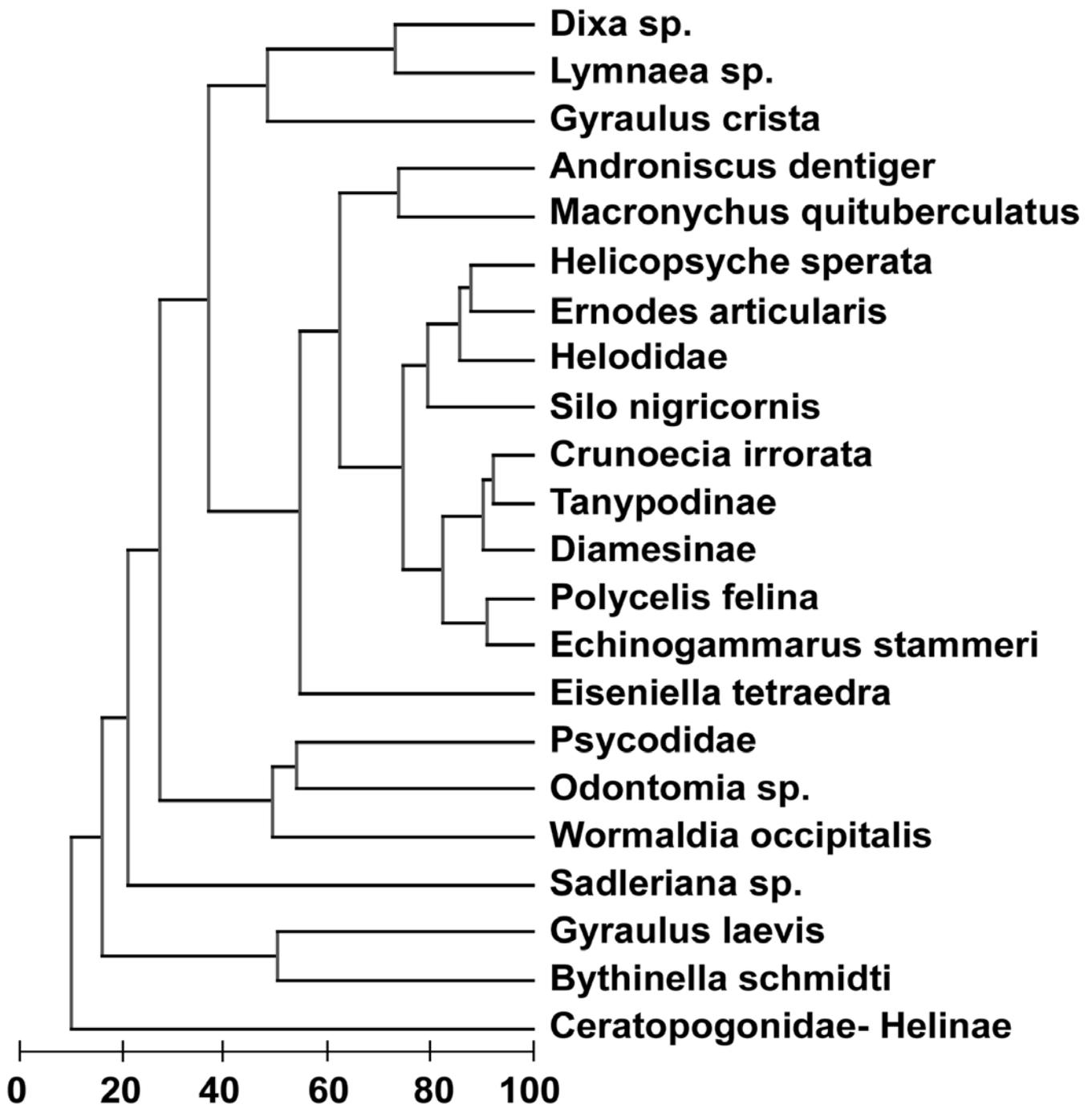


Fig. 2 - Cluster analysis of the macrobenthic community collected in Spring 1.

DISCUSSION

The annual investigation on two plain-springs of Northern Italy showed the absence of clear seasonality for the macroinvertebrate community, despite a marked seasonality of the water temperature. The temperature recorded in the investigated springs in the morning reached almost 20°C in the Summer, and presumably higher in the afternoon. At the same time, in Winter it never fell below 9°C. The relatively high Summer temperature is due to the shallowness of the perched aquifer, which is only about 8 m under the main level of the plain.

Water temperature is undoubtedly the most influential factor in crenal life, but the composition of spring communities also depends on a variety of other factors including habitat structures, microclimate, food and competition (Fisher, 1996). As well known, the presence of riparian vegetation is another important factor which governs the structure of the invertebrate communities in spring brooks: it is the principal resource of allochthonous matter and it has an important role in the trophic chain (Vannote *et al.*, 1980).

In addition, the presence of bordering wet areas that never become dry between spring water is important

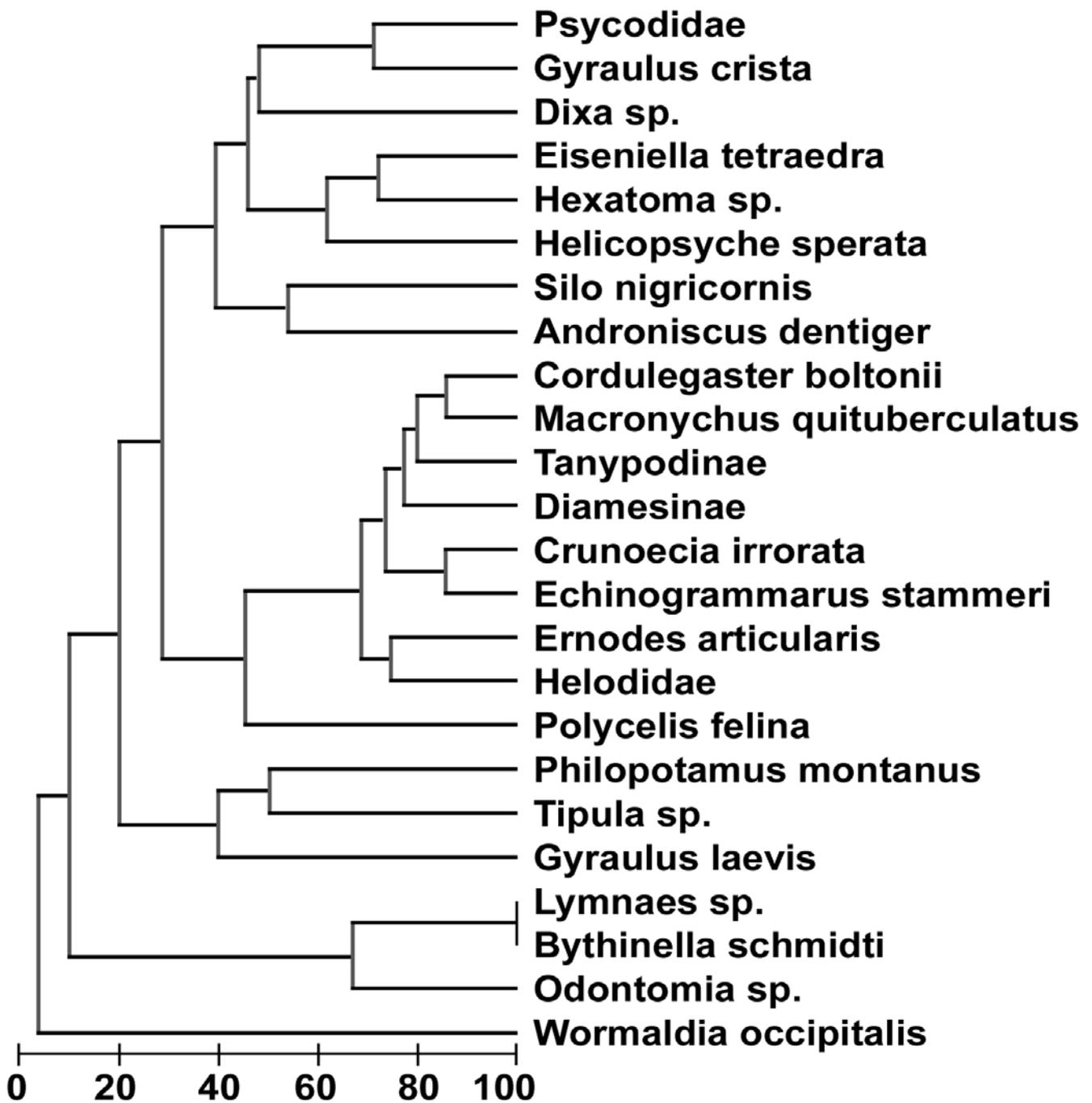


Fig. 3 - Cluster analysis of the macrobenthic community collected in Spring 2.

for the crenic community as also stated by Wagner *et al.* (1998).

The slight differences in species composition observed between the two springs can be attributed to two possible ecological explanations: the first hypothesis was related to a different granulometric composition of the bottom of the two studied springs while the second one could be related to a difference between the life-cycle of the taxa.

According to us, the first hypothesis should be the most relevant because, during our survey, we noticed that the bottom of S1 was mainly composed of gravels,

a most suitable substrate for various species such as *Polycelis felina*, *Silo nigricornis* and *Helicopsyche sperata*; on the contrary, the bottom of S2 was dominated by sand and clay, which instead facilitate the colonization of taxa typical of lentic water, such as the odonata *Cordulegaster boltonii*.

In the two studied springs, 10 out of 26 taxa collected showed a marked preference for spring specific habitat and they are known in literature as cold stenothermic species, associated to mountain springs with an annual temperature range from 3°C to 12°C.

Furthermore, we collected a high number of flies and caddisflies: high abundance of these groups is very common in lowland springs characterized by madicolous microhabitats (Cantonati *et al.*, 2006).

Noteworthy is the constant presence and high abundance of the caddisflies *Crunoecia irrorata*, *Philopotamus montanus*, *Ernodes articularis*, *Helicopsyche sperata* and species belonging to the genera *Wormaldia* and *Silo*. As reported in literature, these species are less common in lowlands springs and their autoecology is strictly linked to springs and spring brooks surrounded by riparian forest, typical of crenic habitats (Moretti & Viganò, 1959; Andree & Timm, 1993; Laukötter, 1993; Cianficconi *et al.*, 1998; Fischer *et al.*, 1998; Hoffsten & Malmqvist, 2000; Ilmonen & Paasivirta, 2005; Smith *et al.*, 2003; Buffagni *et al.*, 2007).

The pool of the nine *taxa* highlighted by the similarity matrix and its cluster analysis was completed by *Polycelis felina*, one of the commonest freshwater planarian in cold and fast-water streams (Stoch, 2001). It is very frequent in rheocrenes springs that are characteristic of highly oxygenated or even oversaturated high-flow springs (Roca *et al.*, 1992; Hanh, 2000).

The presence of stenothermal species is quite unusual for a flood plain area, and their presence highlights the role of springs as a refuge: the environmental characteristics of springs support a relatively rare fauna which can find refuge from disturbing events or extreme seasonal conditions (Di Sabatino *et al.*, 2003; Maiolini *et al.*, 2011).

Although less important from a quantitative point of view, other caddisflies (*Philopotamus montanus*, *Wormaldia occipitalis*) and few gastropods (*Bythinella schmidti* and *Sadleriana* sp.) show a marked habitat-specific preference for crenic conditions (Stoch, 2001; Wiberg-Larsen *et al.*, 2000).

Other frequent species were the coleoptera *Macronychus quadrituberculatus* and the flies *Odontomia* sp.: both species that usually colonize rotten trees near small rivers or wetlands (Olmi, 1976; Campaioli *et al.*, 1994).

Finally, it is also important to highlight the presence of two crustacean: the isopod *Androniscus dentiger* and the amphipod *Echinogammarus stammeri*. *A. dentiger* is mainly considered to be a troglophilous species, but it is also frequently found in the moss in ecotonal margins of springs and spring brooks (Harding & Sutton, 1985; Latella & Stoch, 2001); moreover, the high density of *E. stammeri* could be interpreted as a piece of evidence of a strong stability of the hydric emergence fed by the perched aquifer.

As indicated by Vannote *et al.* (1980), macrobenthic assemblages in springs are different from those in streams in the same geographic area. In accordance with this state and even though the studied terrace springs were very close to a semi-natural stream called Vernavola, between these two habitats there was not a direct connection, and this was also confirmed by the different composition of the two macrobenthic community. In the Vernavola Stream, the community was dominated by *taxa* very common in the lotic environments of the southern River Po plain with moderate/high anthropogenic impact such as the mayflies *Baetis* and *Caenis*, the caddisflies *Hydrop-*

sychidae, the flies *Chironomidae* and *Simuliidae*, the dragonflies *Calopteryx* and *Onychogomphus* (<http://www.comune.pv.it/on/Home/Canalitematici/Ambienteeterritorio/C.R.E.A.html>).

CONCLUSIONS

Springs are important microhabitats in the riverine landscape (Ward & Tockner, 2001; Lamberti *et al.*, 2010; Ilmonen *et al.*, 2012). According to Werum (2001), they can be defined as “water island” because they often show a marked disjunction from neighbouring streams. Furthermore, their community could be considered as a metacommunity because its scarce possibility to overcome barriers (Mac Arthur & Wilson, 1963; 1967; Whittaker *et al.*, 2001; Cantonati *et al.*, 2012).

Our results confirm the important role of lowland springs as haven for some macrobenthic species: the community found in this study is composed of a set of species never been reported before in the nearby freshwater or wetland environments.

As described in the literature, the benthic assemblages of springs (mainly from mountain springs) are typically dominated by species which strictly colonize these particular types of aquatic environments. However, our study demonstrates that some of these stenoeccious species can also colonise springs located in a flood plain composing a “mixed community” with other species commonly found in lowland freshwater environments.

Therefore, our data supports the hypothesis that these “spring species” are not as stenothermic and oligotrophic as usually reported in literature. In fact, our springs are characterised by high winter and summer temperatures and nitrate concentrations, due to the intensive agriculture activities and the superficial level of the aquifer.

In conclusion, considering this data, we hypothesize that the preference for springs of some species is probably more influenced by morpho-physical habitats, namely a thin layer of water, than by low temperature and low organic charge.

Furthermore, our study suggests how the spring habitat is peculiar and conservative, despite its heterogeneity in terms of ecological factors, including altitude, depth of the aquifer and, consequently, temperature.

The scarceness of studies on these habitats could be an incentive to continue investigations, in order to better define the auto-ecology of some species and to enrich the ecological database of these microhabitats.

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