Macroinvertebrate colonization of two different tree species leaf packs (native vs. introduced) in a Mediterranean stream

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Impacts of a micro-sewage effluent on the biota of a small Apennine creek

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Organic pollution of domestic origin represents the most important cause of water quality deterioration in rural and mountainous areas of the northern Apennines. In this study, the ecological consequences of a small sewage dump in the Caramagna Creek (northwestern Italy) were analyzed. The addition of organic matter and nutrients led to a dramatic change in the taxonomic richness and density of the macrobenthic community. Also functional, biological, and ecological composition of the invertebrate assemblages changed downstream of the effluent. Interestingly, benthic chlorophyll *a* showed only a weak increase in the downstream section, despite the increased levels of nutrients. This work emphasizes the importance of better management of sewage treatment also in remote areas.

2 **Keywords:** ■■■

Introduction

Organic pollution represents one of the most common causes of degradation of water quality in stream ecosystems (Paul and Meyer 2001). This kind of pollution is usually categorized as derived from point sources (Goudie 2006). Industrial and farm effluents, and urban run-off are surely important in this context, but sewage pollutants of domestic origin represent the greatest source of organic materials discharged into fresh waters (Mason 2002). In much of the developed world, the greatest part of the population is served by public sewers, and approximately the 80% of sewage receives at least secondary treatment, but the release of crude sewage into watercourses still remains a great ecological problem. Sewage from villages and towns is usually treated and then sent into river systems of medium-high order, but this practice is not so diffuse when we consider wastewaters from isolated houses and small housing assemblages. The impacts of these small point sources attain special relevance in circum-Mediterranean ecosystems, where water-level is scarce and temperature is elevated (Ortiz et al. 2005; López-Rodríguez et al. 2009).

The northern Apennines area, situated between the Alps and the Mediterranean, represents an important biodiversity hotspot, with peculiar climatic, geomorphologic, and biologic characteristics. This area has a low human population density; the economy is generally based on non-intensive agricultural and silvicultural practices,

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while medium and large industrial plants are lacking. Northern Apennine rivers are characterized by the presence of discharge peaks produced by autumn–winter rains, after which flows decline to baseflow in early summer. The hydrographic network of the area, because of the complex morphology of the Apennine mountains, is characterized by the presence of a few medium-sized rivers but a myriad of small to very small streams and creeks. These lotic systems host rich and diversified biological communities (Bo et al. 2009, 2010), with many rare and interesting taxa (Tierno de Figueroa et al. 2009; Fenoglio et al. 2010a). In the recent years, these environments have been increasingly threatened by human activities. Recent studies have emphasized the role of morphological alterations in damaging lotic systems of the northern Apennines (Hering et al. 2001), but little information is available about the effects of sewage micro-effluents on small Apennine creeks. The aim of this study was to analyze the effects of a micro-point source of organic pollution on the biota of a small Apennine creek, investigating the impacts of the effluent on the macroinvertebrate community structure and composition and on the benthic chlorophyll *a* abundance.

Methods and materials

This study was conducted in the Caramagna Creek, a small tributary of the Bormida River, northwestern Italy (44°36′ N–8°32′ E; 280 m a.s.l.). Dense woodlands with small scattered urban areas cover the entire catchment. Riparian vegetation is abundant and mainly composed by *Alnus glutinosa*, *Carpinus betulus*, and *Robinia pseudoacacia*, and the stream flows through a narrow, sinuous channel, characterized by a moderate slope. Riverbed width is approximately 2.0–2.5 m, and in this area substrate showed the following particle composition: 10% sand, 30% gravel, 50% pebbles, and 10% boulders. Moreover, there is a natural series of riffles alternating with shallow pools, with a generally moderate current velocity. At the study site, no morphological alterations are present, but the creek receives an effluent sewer from a small cluster of houses. To evaluate the impact of this point source of pollution, the creek was divided into two parts, and samplings were performed at one reach located 50 m upstream and one 50 m downstream of the sewer pipe. Main chemical and microbiological parameters were measured in six occasions (Table 1). Main physicochemical parameters were measured in the field

Table 1. Main chemical-microbiological parameters upstream and downstream of the sewage outfall in Caramagna Creek (mean ± SD).

Parameters	Upstream	Downstream
Conductivity (μS/cm)	469.0 ± 41.8	510.8 ± 37.1
DO (mg/L)	11.30 ± 0.99	9.04 ± 0.69
T (°C)	8.30 ± 5.28	8.53 ± 5.84
Total P (mg/L)	0.05 ± 0.00	0.82 ± 0.68
COD (mg/L)	7.51 ± 1.40	24.0 ± 30.7
NH_4^+ (mg/L)	0.07 ± 0.07	2.88 ± 3.84
NO_3^- (mg/L)	0.71 ± 0.34	0.69 ± 0.40
pH	7.91 ± 0.19	6.78 ± 2.85
Anionic tensioactives (mg/L)	0.05 ± 0.00	0.06 ± 0.01
E. coli (cfu/mL)	0.00 ± 0.00	1983.3 ± 3504.5

with Eijkelkamp 13.14 and 18.28 portable instruments. Water samples were also collected from the sub-surface in acid-washed polythene bottles or sterile glass bottles. In laboratory, some other chemical and bacteriological properties of the water were assessed by using A.P.A.T. - I.R.S.A. (2003) methods (total P = M.598, COD = M.014, NH_4^+ = M.589, NO_3^- = M.020, anionic tensioactives = M.268, and *Escherichia coli* = M.001).

Macroinvertebrate community composition and structure were evaluated using a 20 × 20 cm^2 Surber sampler (255 μm mesh). Samples were collected at each station monthly from January 2005 to March 2006. In the laboratory, all organisms were counted and identified to the genus level, except for Annelida and early instars of some Trichoptera and Diptera, which were identified to the family level. Each taxon was also assigned to one of the following functional feeding groups: scrapers (Sc), shredders (Sh), collector-gatherers (Cg), filterers (F), or predators (P) according to Merritt and Cummins (1996). Moreover, a classification of taxa into seven biological and seven ecological groups was conducted according to the Usseglio-Polatera et al. (2000) species traits approach. In the same period, benthic chlorophyll *a* was assessed on six occasions by positioning 84 ceramic tiles in the stream reach. Tiles were left in place for 1 month and then brushed to collect attached material. We determined chlorophyll *a* concentrations by spectrophotometry following extraction in 90% acetone according to Steinman and Lamberti (1996). We used the Mann–Whitney U-test to evaluate differences in biological measures between the upstream and the downstream sites.

Statistical analyses were performed with Systat 8.0 (Wilkinson 2000).

Results and discussion

Totally, we collected 156 samples and identified 13,981 organisms belonging to 92 taxa in the section upstream of the sewage outfall and 48,549 organisms belonging to

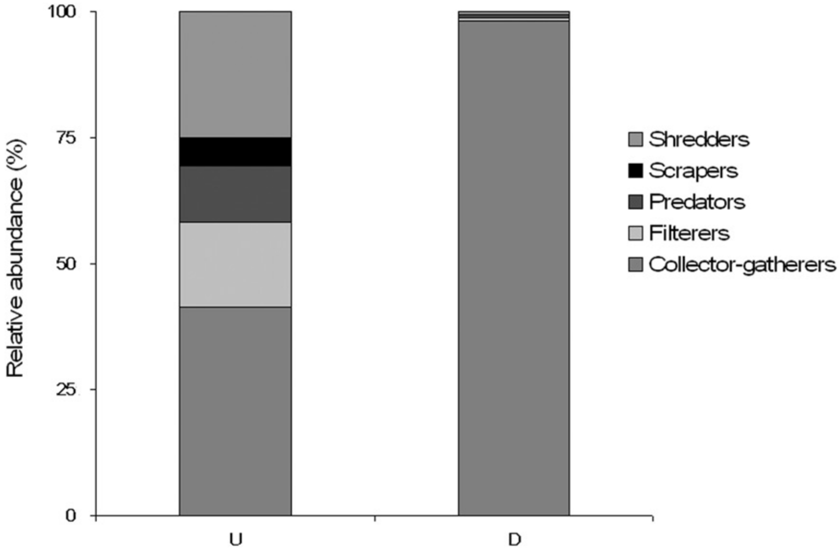

Figure 1. Relative abundance of functional feeding groups upstream (U) and downstream (D) of the sewage outfall in Caramagna Creek.

Table 2. Relative abundances (%) for macroinvertebrates collected upstream (U) and downstream (D) of the sewage outfall.

Taxon	U	D	Taxon	U	D
Plecoptera			*Calopteryx* sp.	0.036	0.004
Capnia bifrons	0.336	0.006	*Onychogomphus forcipatus*	0.129	0.002
Leuctra sp.	2.525	0.035	*Orthetrum* sp.	0.029	0.000
Nemoura sp.	1.280	0.189	Coleoptera		
Protonemura sp.	0.072	0.004	Haliplidae	0.007	0.000
Isoperla sp.	1.001	0.027	Dytiscidae		
Brachyptera sp.	13.140	0.272	*Deronectes delarouzei*	0.000	0.000
Ephemeroptera			*Deronectes moestus*	0.007	0.000
Electrogena sp.	0.143	0.000	*Hydroglyphus pusillus*	0.007	0.000
Serratella ignita	0.837	0.132	Unidentified larvae	0.043	0.004
Epeorus sylvicola	0.036	0.000	Hydrophilidae	0.014	0.000
Siphlonurus lacustris	0.486	0.132	Dryopidae		
Caenis sp.	0.694	0.014	Unidentified larvae	0.014	0.000
Baetis sp.	14.700	0.692	*Helichus substriatus*	1.688	0.027
Torleya major	0.014	0.004	Hydraenidae	0.000	0.000
Centroptilum luteolum	1.016	0.000	*Hydraena similis*	0.000	0.002
Ecdyonurus sp.	2.775	0.066	*Haenydra solarii*	0.000	0.000
Ephemera danica	0.229	0.004	*Hydraena andreinii*	0.658	0.023
Paraleptophlebia sp.	0.315	0.000	*Hydraena assimilis*	0.093	0.000
Habroleptoides sp.	0.608	0.021	*Haenydra heterogina*	0.021	0.000
Habrophlebia sp.	3.169	0.043	*Haenydra truncata*	0.086	0.000
Trichoptera			*Hydraena subimpressa*	0.286	0.000
Polycentropodidae	0.000	0.002	*Haenydra devillei*	0.021	0.000
Hydroptilidae	0.007	0.000	*Limnebius mucronatus*	0.007	0.000
Beraeidae	0.043	0.000	Gyrinidae	0.322	0.006
Hydropsyche sp.	1.395	0.031	Elmidae		
Wormaldia sp.	0.486	0.004	Unidentified larvae	0.479	0.016
Philopotamus sp.	0.229	0.002	Unidentified adults	0.615	0.014
Leptoceridae	0.501	0.000	*Esolus* sp.	0.021	0.000
Goeridae	0.043	0.000	Scirtidae	0.622	0.002

Glossosomatidae	1.473	0.016	Hemiptera		
Psychomyidae	0.315	0.006	*Velia* sp.	0.007	0.000
Limnephilidae	1.209	0.008	*Hydrometra stagnorum*	0.007	0.000
Rhyacophila sp.	0.000	0.002	Gerridae	0.000	0.002
Hyporhyacophila sp.	0.036	0.002	*Micronecta* sp.	2.589	0.004
Anabolia lombarda	0.057	0.000	Hirudinea		
Potamophylax cingulatus	0.050	0.000	*Dina* sp.	0.007	0.000
Odontocerum albicorne	0.336	0.000	*Helobdella stagnalis*	0.007	0.000
Sericostoma sp.	0.136	0.000	Annelida		
Diptera			*Eiseniella tetraedra*	2.296	0.035
Culicidae	0.007	0.000	Lumbricidae	0.858	0.014
Anopheles sp.	0.007	0.000	Lumbriculidae	0.479	0.091
Atherix sp.	0.136	0.002	Naididae	1.802	79.730
Chironomidae	11.780	16.980	Tubificidae	0.093	0.187
Ceratopogonidae	0.966	0.181	Tricladida		
Dixa sp.	0.043	0.000	*Dugesia* sp.	5.443	0.051
Tabanidae	0.143	0.000	Bivalvia		
Tipula sp.	0.944	0.045	*Pisidium* sp.	0.029	0.000
Stratiomyidae	0.107	0.004	Gastropoda		
Limoniidae	0.272	0.004	*Ancylus fluviatilis*	0.007	0.000
Simuliidae	14.650	0.661	*Lymnaea peregra*	0.100	0.004
Empididae	0.043	0.006	Arachnida		
Psychodidae	0.107	0.004	Hydracarina	1.874	0.037
Sciomyzidae	0.000	0.002	Nematomorpha		
Rhagionidae	0.007	0.000	*Gordius* sp.	0.043	0.000
Megaloptera			Nematoda		
Sialis fuliginosa	0.007	0.000	Unidentified species	0.072	0.117
Odonata			Mermithidae	0.021	0.019
Cordulegaster boltoni	0.093	0.000	Hymenoptera		
Boyeria irene	0.021	0.000	*Agriotypus armatus* (in Goeridae)	0.014	0.000

56 taxa in the downstream section of Caramagna Creek. The upstream and downstream reaches were different from each other in many biological aspects.

The upstream section of Caramagna Creek is a typical Apennine low-order watercourse. Richness and density of the macroinvertebrate community were high and in the range reported for other streams in this area (Fenoglio et al. 2005, 2010b). However, taxonomic richness was significantly ($p < 0.001$) different between upstream ($N = 17.7 \pm 5.76$ SD) and downstream ($N = 8.70 \pm 6.18$ SD) sites. Likewise, organism density was significantly ($p < 0.001$) different, being 128.2 ± 106.6 SD organisms/m^2 upstream but 1032.9 ± 1495.6 SD organisms/m^2 downstream.

Furthermore, in the upstream section, the functional composition of the benthic biocenosis seemed well structured. Collector-gatherers comprised the most abundant functional feeding group, followed by shredders, filterers, predators, and scrapers. Downstream of the sewage effluent, the functional composition was completely altered, and collector-gatherers represented the dominant and almost exclusive group. Considering the relative importance of the functional feeding groups in the upstream and downstream communities, significant ($p < 0.001$) differences were also present for all functional groups except the collector-gatherers (Figure 1). Ephemeroptera/Plecoptera/Trichoptera taxa were present, with 34 taxa (representing 49.7% of the total number of organisms) in the upstream reach and 24 taxa (1.7% of the total number) in the downstream reach (Table 2).

Considering the relative importance of the biological traits group in the two communities, significant ($p < 0.001$) differences were evident in all trait groups except group ‘b’ (Figure 2). The most represented biological trait group was the ‘e’ group (small- or medium-sized organisms, uni-or plurivoltine, with aquatic respiration, crawlers), followed by the ‘f’ group (medium-sized or large monovoltine organisms, with aquatic respiration, crawlers), while the ‘b’ group was the rarest (medium or large crawlers or burrowers, mostly ovoviviparous).

Considering the relative importance of the ecological traits groups, significant ($p < 0.001$) differences were also apparent between the two communities for all groups except the ‘E’ group (i.e., eurythermic or thermophilous mesosaprobic, living in lentic riverine microhabitats) (Figure 3). The most important group was the ‘B’ group (organisms adapted to rhithronic and oligotrophic environments with coarse substrate), followed by the ‘C’ group (those living in rhithronic or epipotamic oligotrophic environments with slow-medium current velocities). The ‘F’ and ‘D’ groups (organisms avoiding the main channel and living in semi-lentic habitats, oligo- or mesotrophic) were also abundant.

Downstream of the sewage outfall the biological community changes dramatically. Density of invertebrates increased eightfold, while the taxonomic richness of the community collapsed, with the complete loss of most taxa. Oligochaeta Naididae and Diptera Chironomidae represented, respectively, 79.7% and 17.0% of the total invertebrates collected in the downstream samples. These groups are usually well known for their tolerance to organic pollution and, as collector-gatherers, are advantaged by the increased presence of fine particulate organic matter downstream. Naididae are multivoltine, burrowers/interstitial, microphagous deposit-feeders (biological trait group ‘F’), while Chironomidae are small/medium-sized organisms, plurivoltine, and short-lived crawlers with varied feeding habits (group ‘E’).

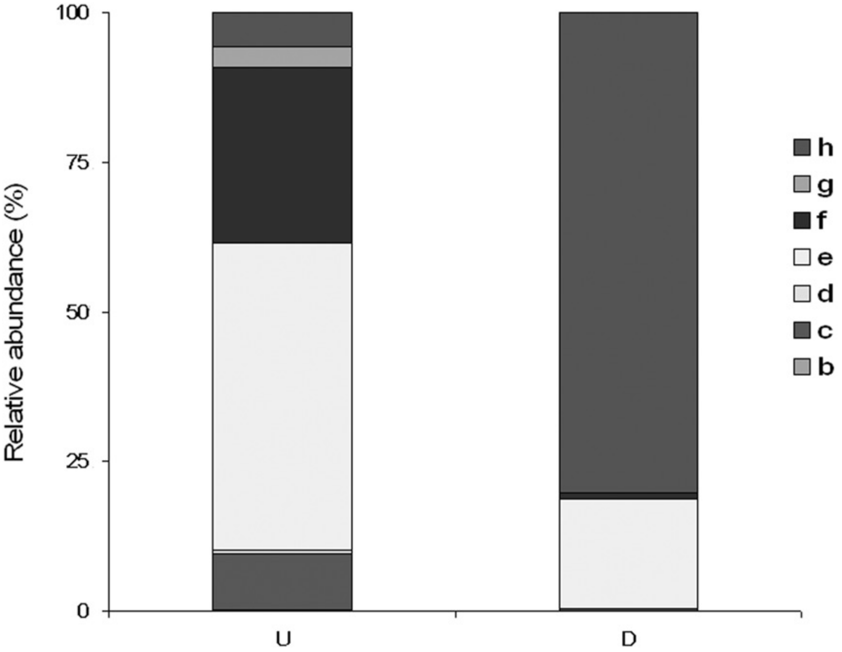

Figure 2. Relative abundance of biological traits groups (Usseglio-Polatera et al. 2000) in the functional feeding group presence in the upstream (U) and downstream (D) reaches of Caramagna Creek.

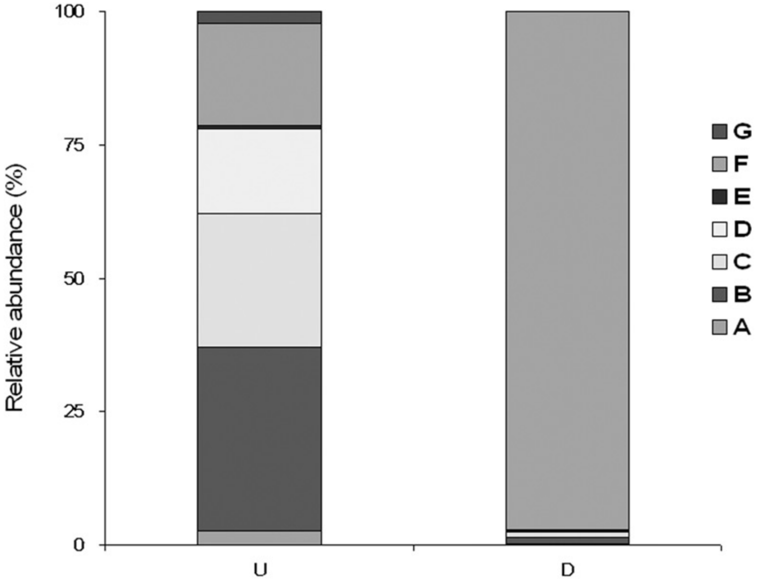

Figure 3. Relative abundance of ecological traits groups (Usseglio-Polatera et al. 2000) in the functional feeding group presence in the upstream (U) and downstream (D) reaches of Caramagna Creek.

Ecologically, both Naididae and Chironomidae are organisms adapted to the life in slow flowing or semi-lentic, mesosaprobic environments with fine and organic substrata (ecological group 'F').

The increased concentrations of nutrients downstream versus upstream would suggest an increase in the instream autotrophic component. However, although the amount of chlorophyll *a* found on artificial substrates was slightly higher downstream of the sewage outlet (0.44 ± 0.50 SD μg/cm^2) than upstream (0.32 ± 0.19 SD μg/cm^2), the difference was not significant.

The impact of small-not-regulated sewage sources on high quality aquatic environments is important. The protection of lotic environments should not be entrusted only to an efficient management of large sewage treatment plants, but it must assume an accurate control of the many, small organic waste dumps that are present also in isolated and rural areas.

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