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Small-scale distribution of macroinvertebrates on Large Woody Debris and nearby sediment in a lowland river.

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1. INTRODUCTION

It's worldwide recognized that water is, and it will increasingly be, one of the biggest issues society must confront, and that freshwater quantity and quality in particular are already becoming primary concerns for local and national authorities. In 2015 will end the International Decade for Action – “Water for life”, which main topic is to halve the proportion of people lacking access to safe drinking water and sanitation. But the quantity is not the only concern, also the quality of our freshwater's sources is facing the worst crisis ever: pressures from human activities are degrading habitats all over the world, and surface waters could well be “the most endangered ecosystems in the world” (Dudgeon *et al.*, 2006).

Rivers and lakes in general are to be considered hotspots for biodiversity in themselves, since they are home to one third of the vertebrate species known, not talking about invertebrate and microorganisms, (Dudgeon *et al.*, 2006). Even if freshwater animals are among the least known and mapped species, it is clear that they are declining all over the world (Revenga *et al.*, 2005). The threats endangering freshwater ecosystems can be grouped in five main groups: overexploitation, water pollution, flow modification, destruction or degradation of habitat and invasive species (Revenga *et al.*, 2005; Dudgeon *et al.*, 2006; Allan and Flecker, 1993). Global environmental changes exasperate these main categories. Overexploitation hit vertebrates, mainly fishes. Water pollution is pandemic: not only from domestic and industrial chemicals, but also from nutrients run off from agricultural areas (Dudgeon *et al.*, 2006). In fact, while many states successfully reduced the firsts sources, the seconds remain a world spread problem. Habitat degradation can be direct (excavation of river sand) or, more often, indirect, when for example changes in the river basin trigger changes in the sediment discharge (Dudgeon *et al.*, 2006). Flow modification is also spread all over the world (Nilsson and Berggren, 2000), and with the objective of increasing the renewable energy supply, is likely to increase even more in the next future. Presence of invasive species, released intentionally or not, is also a widespread problem, and it's likely to increase, since physical and chemical modified freshwater habitats are more likely to be invaded (Bunn and Arthington, 2002).

During the last 13 years in Europe, after the adoption of the Water Frame Directive (WFD - 60/2000/EC) river ecology and functionality have gained more and more importance. The WFD showed an implicit recognition that the traditional management of water bodies, based on a physical and chemical approach, had to be changed. Particularly, it's recognized that the main objective of the WFD is the achievement, for all European water bodies of the Good Ecological Status, and every country should “protect and improve” his superficial water bodies, so developing reliable indicator indexes and efficient restoration methods is now a priority.

1.1. Importance of LWD

Definition: *Large Woody Debris* (referred here as **LWD** or **Log**) are usually defined as “ wood pieces greater than 10cm diameter and 1 m length” (Platts *et al.*, 1987 in Piégay and Gurnell, 1997)

Until few decades ago, river management has always focused its attention on the hydrological point of view. For centuries, rivers had been rectified, “cleaned” and in general, homogenized to achieve better hydraulic characteristics (Petts *et al.*, 1989; Gippel *et al.*). Nowadays, a different approach try to achieve the best result in flood defense with the smallest possible alteration of the river bed, reducing the impacts and the costs but improving the habitat quality (or at least not reducing it too much) (Gippel *et al.*). Not only the wood is a valuable habitat for several groups of benthic animals and the most productive area of lowland rivers (Benke *et al.*, 1985), but the wood accumulations in lowland, sand bedded rivers are “patch creators” adding diversity of habitat in an otherwise homogenous river (Gurnell *et al.*, 1995). From a “WFD” point of view, LWD can regulate minerals and nutrients distribution, storing and releasing material, thus providing temporal and spatial distributed food sources to the aquatic biota, helping to maintain an healthy river (Gurnell *et al.*, 1995). Also it can play an important role in sustaining refuge habitats for the biota, protecting it in case of disturbances and pollution events (Gurnell *et al.*, 2002; Hax and Golladay, 1998). However, the potential of restoration to deliver its anticipated benefits, the recovery process, and the identification of conditions that cause either success or failure all remain poorly understood (Lepori *et al.*, 2005).

In small lowland river, wood is of primary importance, first of all because the dimension of the pieces is normally too massive to be easily dislodged and transported by the flow (Warmke and Hering, 2000), thus they actually control more than respond to the hydrological characteristics of the river (Gurnell *et al.*, 2002). They are also much more stable than any other organic substrate (Webster and Benfield, 1986). (Benke *et al.*, 1985) demonstrated that, even if wood habitats were rare in a sand-mud bottom river, the logs sustained 60% of the total invertebrate biomass. The function of the wood is not only to provide stable substrate, but also of direct and indirect food source: some taxa are known to be able to directly eat woody debris e.g. (Anderson, 1989; Steedman and Anderson, 1985; Hynes, 1970) while others can benefit from the enhanced growth of biofilm (Sinsabaugh *et al.*, 1991).

1.2. LWD and flow patterns

As already stated, in lowland river with low energy, wood can be the main feature, modifying the surrounding environment. An interesting study about the modifications triggered by wood in this

particular environment can be found in Mutz (2000). In this study it is shown how logs hanging in mid-water creates high velocity flows that scour the sediment, thus creating patches of coarser sediment in rivers where the base sediment is shifting sand. On the other hand, wood resting directly on the streambed act as a roughness element, projecting a cone of low flowing water downstream, an area of accumulation for fine sand. It is also shown how, in these small rivers, influences of LWD diminish greatly between 1 to 3 meters downstream of the wood location.

1.3. The Micro - scale approach

Micro-scale habitats are largely defined as the smallest geomorphologic units in a river, with a scale of 1 meter (and 1 year) (see Harper and Everard, 1998). Habitats are important because they are the interface between organisms living in the river and physical processes controlling the river structure (Figure 1).

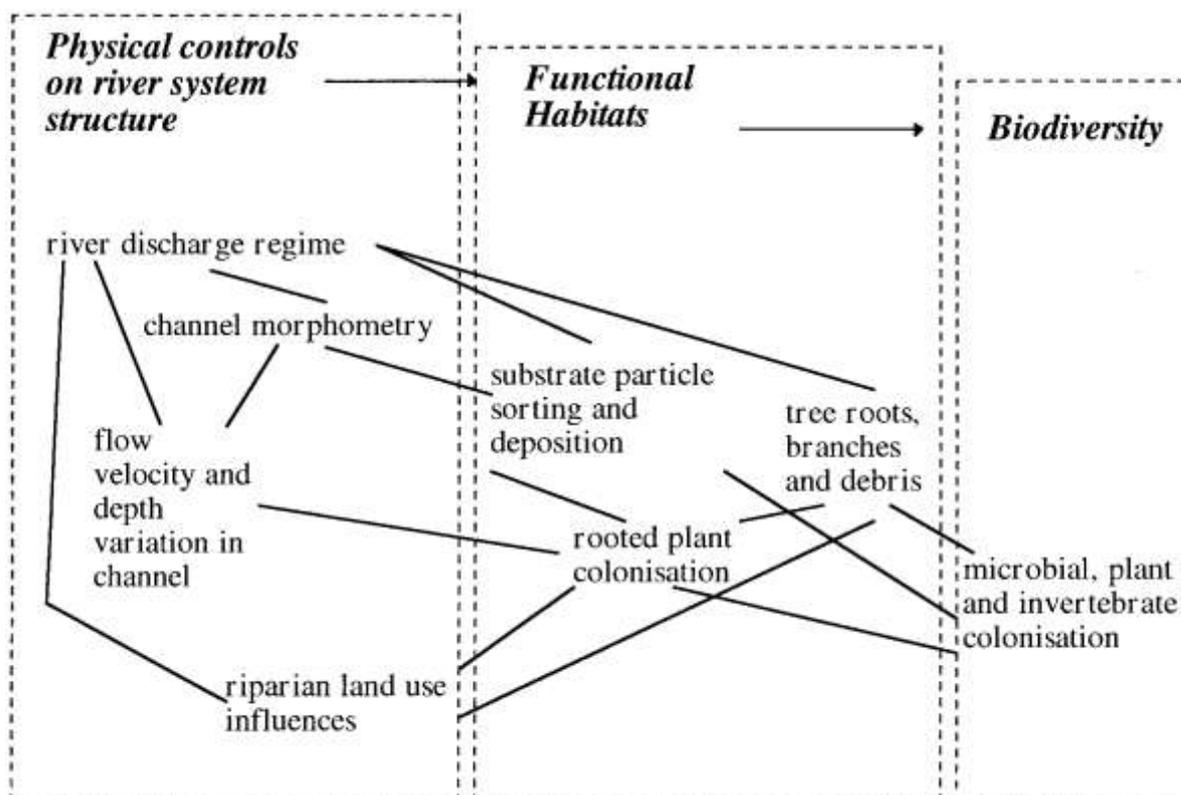


Figure 1 The importance of functional habitats at the interface between the geomorphological processes and human land uses in a river, and its in-stream biodiversity (from Harper and Everard, 1998).

The term “Functional Habitat” was then introduced because habitats are not a mere scale level, but the real basic physical functioning of a river ecology, thus they can be helpful in riverine management operations if considered (Harper *et al.*, 1992). Basic studies at a small-scale of the benthos

distribution should so be further developed, since they are the building – blocks of knowledge for wider assessments (Harper and Everard, 1998; Bond *et al.*, 2006).

In this study, we mostly used the term “Mesohabitat” or “Habitat”. The definition followed is: “Within a stream reach each mesohabitat type would be represented by multiple patches of habitat, which are physically similar to each other but distinct from other mesohabitats” (Pardo and Armitage, 1997). The assumption is that it could be identified, following general characteristics of flow velocity, depth and sediment size, different mesohabitats downstream LWDs.

1.4. Macroinvertebrates¹

Even if the term “Macroinvertebrates” is nowadays extensively used and recognized, the definition is somewhat variable, particularly for the lower boundary. In our study, we considered macroinvertebrates all those animals recognizable with a stereo microscope (>0.5mm). Also, we considered only benthic invertebrates, those animals living on or close to the surface of the sediment.

1.4.1. Benthic macroinvertebrates and humans

From a human approach, rivers and their components are valuable mainly for the direct services and resources that they can provide to mankind, and this has led, historically, to study fish and pollution. However, macroinvertebrates plays a leading role in these human based interests, mainly in three sectors: organic matter transformation, food source for the fish population and pollution.

The first point is related to the food chain in the rivers. Rivers are mainly heterotrophic environments, sustained by alloctonous inputs of energy (leaves and other vegetal detritus). Thus degradation of these food sources is of paramount importance, leading to the fact that all the ecology of rivers is based on this (Vannote *et al.*, 1980). Macroinvertebrates break down leaves and other materials, enhancing bacterial and fungal degradation.

For the fish ecology, it is easily said: macroinvertebrates are often a huge part in fish diet, both for the juvenile and the adult stages.

The relation with the pollution it is nowadays the most highlighted. Macroinvertebrates, like all living organisms, are sensible to pollution, either organic or chemical, with a differential sensibility to different types and level of pollution. The answer of macroinvertebrates to the pollution is typically a reduction of the diversity and structure of the assemblages, with a disappearance of the taxa most

¹Information in this chapter, if not otherwise stated, are taken from Tachet *et al.* (impr. 2010)

pollution – sensible. There are several reasons why macroinvertebrates are so pollution-related. One is their life cycle, often annual, and their locomotion, that it is normally weak. This means that a population can show the consequences of an acute pollution event even several months after the event, when the pollutant maybe has already disappeared. In the best case, a population will recover at the start of the next cycle. On the other hand, fishes and bacteria, although pollution sensible, are more difficult to use in the case of acute pollution: they both can recover, one due to their high mobility, while the others for their really short life cycle.

For all these reasons, having a deep knowledge of the ecology of macroinvertebrates, their preferences and distribution among the different habitats in pristine environments it is important. These kind of studies provide a solid base on which to construct indexes and evaluation methods to monitor pollution and evaluate success in the restoration measures.

1.4.2. Invertebrates present in this study

It is here reported, in few words, some characteristics of the principal taxa considered in this study. A list of the species found can be found on Annex 1.

Turbellaria

The only order of Turbellaria considered it is the Tricladida. These flat worms crawl along the river bottom and feed on other invertebrates. Their life span is normally no more than few months, the reproduction can be sexual or asexual. Their life cycle it is completely aquatic. Turbellaria bigger than 5mm have been identified to the genus or species level.

Oligochaeta

Oligochaeta are Anellida characterized by the presence of two pairs of setae's bands. They have a segmented body. Their life cycle it's very variable, from few weeks to some years. These worms are for the vast majority detritivores , feeding on organic matter highly decomposed. Oligochaeta have not been identified, due to the high numbers, difficult identification and the often bad state of the specimens. To avoid overestimation, only the "heads" were counted (Figure 13).

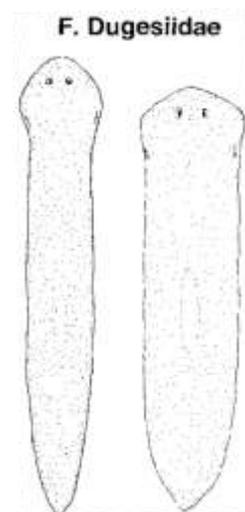


Figure 2 Two different forms of *Dugesia* sp. (Tachet et al., impr. 2010)

Hirudinea

Few Hirudinea have been found. They are also Anellida, with a segmented body and two suckers, one on the front and one on the end of the body. Their life span it is in the order of the years. Leeches are normally predators of invertebrates or parasites hematophagous of vertebrates. Hirudinea have normally been identified to the species level.

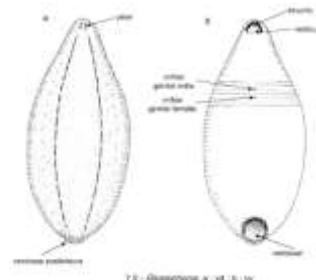
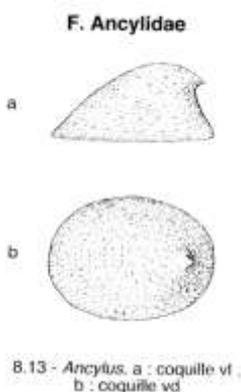


Figure 3 *Glossiphonia sp.* dorsal (left) and ventral vision (right)(Tachet *et al.*, impr. 2010)



Gastropoda

Specimens belonging to both the groups of Prosobranchia (evolved from marine species) and the Pulmonata (terrestrial origin) have been found. Both the groups present normally a shell, but while the first present an operculum, the second group never. Their life cycle can be mono or bivoltinuous. Snails are basically herbivorous and detritivores, even if occasionally they can feed on dead animals. All the Gastropoda have normally been identified to the species level.

Figure 4 Shell of *Ancylus fluviatilis*, Pulmonata. Example of grazer of hard substrates. (Tachet *et al.*, impr. 2010)

Bivalvia

Bivalves are Mollusca that present an outer shell formed by two valves. Two main families have been found: Unionidae and Sphaeriidae. The Unionidae can live up to 30 years and reach several centimeter in length on the main axis of the shell, while the Sphaeriidae live normally few years and are much smaller. All the mussels are filtrators of phytoplankton, bacteria or Fine Organic Matter. Mature stages of Unionidae have been identified to species level, while Sphaeriidae have been identified to genus (*Sphaerium sp.* and *Pisidium sp.*).

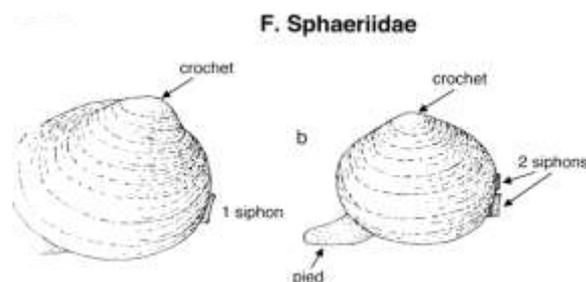


Figure 5 *Pisidium sp.* (left) and *Sphaerium sp.* (right). Typical active filter feeders. (Tachet *et al.*, impr. 2010)

Crustacea

Two orders of Crustacea were found: Amphipoda and Isopoda. Only one specimen belonging to the order Isopoda was sampled while among the Amphipoda, only Gammaridae (Figure 12). Life span is normally one or two years and they are mainly detritivores. All the specimens bigger than 5mm have been identified to species level.

Insecta

Insecta are by far the most diverse group of aquatic invertebrates living in freshwater environments. From an evolutionary point of view, insecta are originally terrestrial animals, that colonized the freshwater environment. This adaptation is seldom complete: their life cycle normally includes at least one terrestrial passage (often the adult form). This lead to define Of particular interest are the so called EPT taxa (Ephemeroptera, Plecoptera, Trichoptera), that include most of the species considered as good quality indicators. EPT relative or absolute values are sometimes used as direct indication of water quality status (Bonada *et al.*, 2006; Álvarez-Cabria *et al.*, 2011; Lester *et al.*, 2007; Li *et al.*, 2001; Hrodey *et al.*, 2008; AQEM consortium, 2002).

Ephemeroptera

Ephemeroptera is an order of insects hemimetabolous, which larvae are exclusively aquatic. They are characterized by a life cycle spanning from three to six months (2 generations per year), but with several exceptions. The life of the Imago seldom surpasses three days. These insects are widespread around the globe, with most of the feeding types represented. Anyway, most ephemeroptera are either collectors or grazers. All the specimens older than first larval stage have been identified at least at genus level, possibly at species level.

Plecoptera

Plecoptera is an order of insects hemimetabolous, which larvae are exclusively aquatic. Some species are monovoltinous (6 months to one year) while others require more than two years to complete the development. The adult stage survive normally longer than Ephemeroptera (few weeks). Plecoptera are mostly collectors and scrapers. Some genus are predators. The identification to the species level is only for the last larval stages.

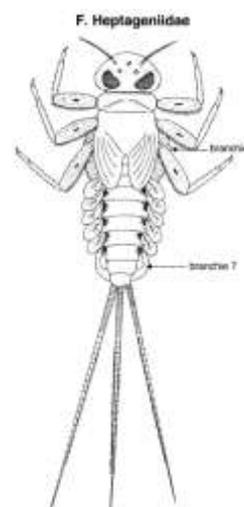


Figure 6 *Heptagenia sp.* larval stage. This is a grazer form of ephemeroptera. (Tachet *et al.*, impr. 2010)

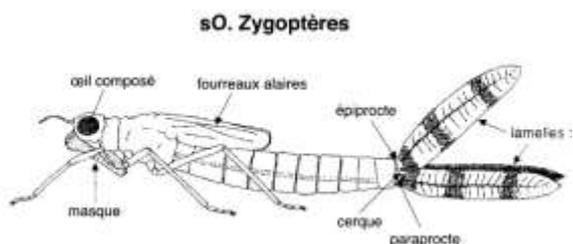


Figure 7 Generic example of damselfly larvae (Zygoptera). (Tachet *et al.*, impr. 2010)

Odonata

Odonata is an order of insects hemimetabolous, which larvae are exclusively aquatic. Dragonflies and Damselflies are exclusively predators. Larval life last between one and five years, while the adult life several weeks. All the specimens have been identified either at the genus or the specie level.

Heteroptera

This group is an under-order of Hemiptera, the only one in Europe with some representatives in the water related environment. They are insects paurometabolous, with coexistence of adults and larval stages. The taxa that were found, Corixidae and Aphelocheiridae (Figure 21), are respectively collectors and predators. They were all identified to the specie level.

Neuroptera

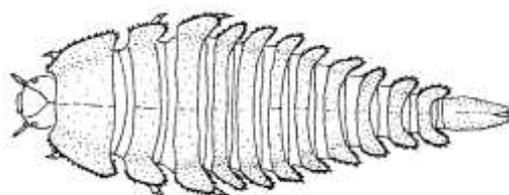
Most of these primitive insects are terrestrial, only some species are aquatics. The group that was found (Sisyridae) are parasites of freshwater sponges, on which they feed. The few specimens found were identified to specie level (Figure 16).

Lepidoptera

Of this order mainly terrestrial, only one family has aquatic larvae. As terrestrial caterpillars, they feed exclusively on plants. Some larvae construct a case made by leaves fragments. They are monovoltinuous. The identification was carried to the genus level.

Coleoptera

Coleoptera are the main order of insects, as number of species. Several groups live in the aquatic environment, some only as larvae, some only as adults while in others both stages coexists (but in this case at least the nymph is terrestrial). Among coleoptera, almost all feeding types are present. Among the groups found, there were grazers, predators, xylophagous and shredders. The life span it is normally around 1 year. All the larval specimens have been identified to the specie or genus level. All the adults to the specie level.



Elmis

Figure 8 *Elmis sp.* larvae as example of a xylophagous coleopteran specie. (Tachet *et al.*, impr. 2010)

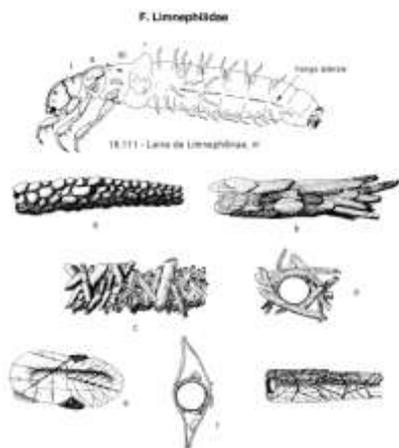


Figure 9 Limnephilidae larvae with case examples. (Tachet *et al.*, impr. 2010)

Trichoptera

Trichoptera is an order of insects with larvae exclusively aquatic (except one specie). Life span it is very variable among different taxa. Stoneflies are characteristic because in many families larvae create a case around their bodies. These cases can be made out of many materials (pebbles, sand, leaves, etc.) and are often useful for the identification. There are a lot of different feeding types among trichoptera: filters, grazers, predators and collectors are the most common. All the specimens have been identified to specie level, except for the Limnephilidae family, which was not identified further (identification possible only in the last larval instars).

Diptera

Several different families of this big order of insects have larval adaptations to live underwater. Life cycle it is variable (from few weeks to several years). Diet of diptera is extremely variable. Among the groups found there were collectors(e.g. Tipuliidae), gatherers (e.g. some Chironomidae), filter feeders (e.g. Simuliidae,

Chironomidae) (Figure 15), predators (e.g. Tanypodinae, Athericidae, Empididae, Tabanidae) (Figure 11) but also parasites and xylophagous are present. Among these families, some are very important for humans because they include pest species, particularly hematophagous flies. Most of the diptera have been identified to family or tribu level. Some (e.g. Athericidae) to specie level.

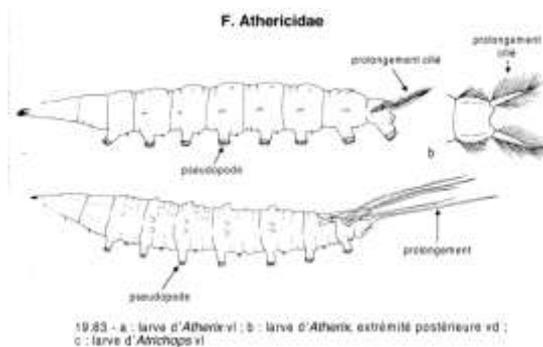


Figure 11 *Atherix ibis* larvae (top), *Atrichops crassipes* larvae(bottom). Examples of predatory diptera. (Tachet *et al.*, impr. 2010)

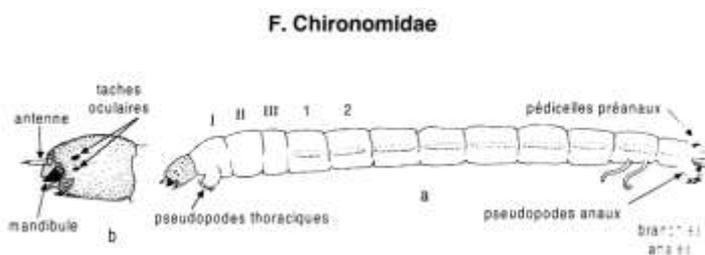


Figure 10 Chironomidae larvae (a) and detail of the head capsule (b) (Tachet *et al.*, impr. 2010)

1.4.3. Feeding groups

Stream Invertebrates are usefully classified among feeding guilds. These guilds are normally not based on the type of food, but on the adaptation the animals present for food collection (see Cummins and Klug, 1979). It is reported here a small description of the feeding types as considered in this study. All the example species are from the taxa list in Annex 1.

Shredders

This group plays a prominent role in many small, high energy streams (Vannote *et al.*, 1980). They can be considered the “first stage” of the river food chain, since they are specialized in attacking big organic structures (CPOM, particularly leaves) thus reducing their dimensions and augmenting the surface available for bacterial and fungal action that will transform CPOM in FPOM (Cummins and Klug, 1979). Examples: *Gammarus sp.* (Crustacea), *Nemoura avicularis* (Plecoptera), Psychomyiidae (Trichoptera), Lepidoptera and Tipuliidae (Diptera).

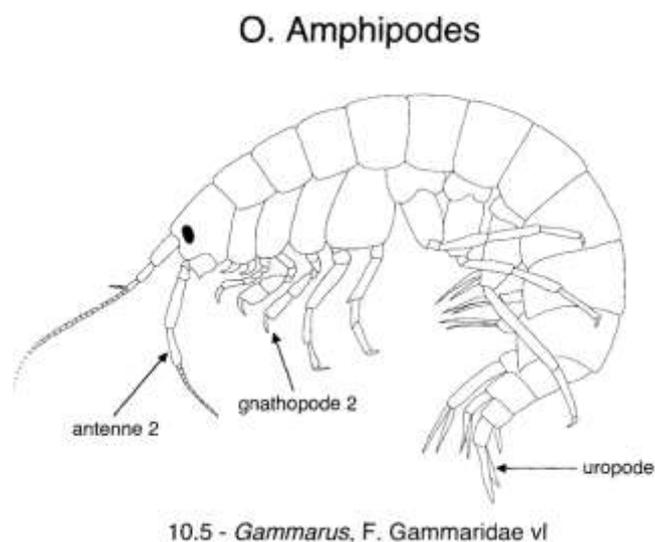


Figure 12 *Gammarus sp.* (Tachet *et al.*, impr. 2010)

Gatherer Collectors

This important group is dominant in bigger and slower streams, gradually supplanting the Shredder dominance to become by far the most common group. The collectors feed on FPOM lying on every underwater surface, and are often living on the bottom of the stream, or burrowed close to the sediment surface (Cummins and Klug, 1979). Examples: Oligochaeta, *Brachycercus harrisella* (Trichoptera), *Micronecta sp.* (Heteroptera), *Taeniopteryx nebulosa* (Plecoptera) (Figure 22), Limoniidae (Diptera).

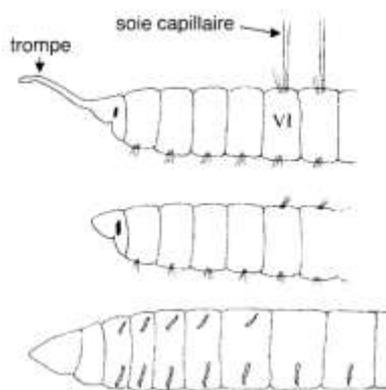


Figure 13 Oligochaeta: different head morphologies. (Tachet *et al.*, impr. 2010)

Grazers & Scrapers

This group include invertebrates with morpho – behavioral adaptation to graze upon food that adhere to surfaces (e.g. periphyton). They normally have adaptation to resist fast flowing water over

exposed surfaces (Cummins and Klug, 1979). Examples: *Ancylus fluvialis* (Gastropoda), *Elmis sp.* (Coleoptera) (Figure 8), *Ithytrichia lamellaris* (Trichoptera).

Miners

Animals that feed from leaves of aquatic plants, algae and cells of aquatic plants .

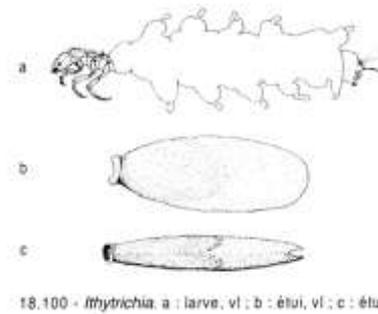
Example: Hidroptilidae (Trichoptera), Lepidoptera

Xilophagous

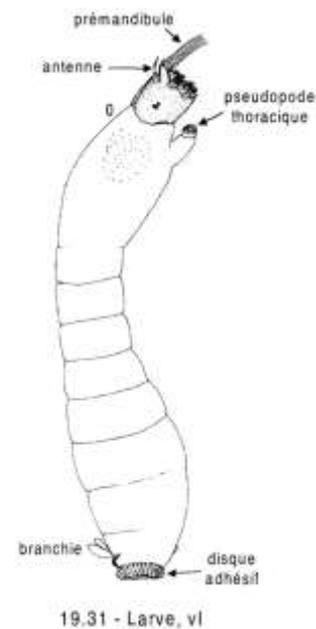
Xilophagy it is not a feeding behaviour but an actual food source. Xilophagous could be either united with shredders or miners, but it underpin so many particular adaptation (physiological and of habitat selection), that since few years it is often considered a separated feeding guild (see Benke and Wallace, 2003). Seldom xylophagy is the only food source of invertebrates. Often, it is a more or less consistent addition to other food behaviour (like grazing). Example: *Macronichus quadrituberculatus* (Coleoptera), *Lype sp.* (Trichoptera)

Filter feeders(Active, Passive Filter feeders)

Filter feeders are often associated with collectors. They feed on fine organic matter (FPOM) suspended in the water (Cummins and Klug, 1979). Filters can be divided in two main groups: active and passive filters, based on their feeding adaptation. As a general rule, passive filters prefer fast flowing areas, while active focus on more stagnant areas (AQEM consortium, 2002). Examples: Active – *Anodonta Anatina* (Bivalvia), *Bithynia tentaculata* (Gastropoda), *Ephemera sp.* (Ephemeroptera); Passive – *Brachycentrus sp.*(Figure 20) , *Hydropsyche sp.* (Trichoptera), Simuliidae (Diptera).



18.100 - *Ithytrichia*. a : larve, vl ; b : étui, vl ; c : étui, vd.
Figure 14 *Ithytrichia lamellaris*, body (top) and case (bottom). (Tachet et al., impr. 2010)



19.31 - Larve, vl
Figure 15 Typical passive filter (Simuliidae) (Tachet et al., impr. 2010)

Predators

This group includes all those invertebrates specifically adapted for the capture of living preys (Cummins and Klug, 1979). Predators in the River Continuum Concept are considered to be fairly constant in all the different river habitats (Vannote *et al.*, 1980). Examples: *Aphelocheirus aestivalis* (Heteroptera) (Figure 21), *Atherix ibis* (Diptera) (Figure 11), *Dugesia sp.* (Hirudinea) (Figure 2), *Gomphus sp.* (Odonata) (Figure 23), *Polycentropus sp.* (Trichoptera)

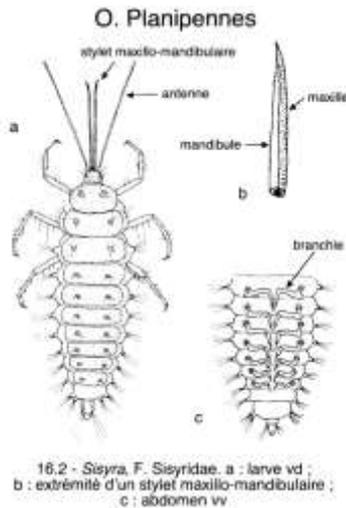


Figure 16 *Sysira nigra*. Full larval body (a), mouth detail (b), abdominal gills (c) (Tachet *et al.*, impr. 2010)

Parasites

Parasites are invertebrates living at the expenses of other animals. Ticks (Hydracnida) are a common example. The only “pure” parasite taxa found is a Planipennes, living at the expenses of freshwater sponges.

Example: *Sysira nigra* (Planipennes).

Others

Invertebrates for which the feeding habit it is not clear or not specialized.

Examples: *Potamopyrgus antipodarum* (Gastropoda), *Gammarus sp.* (Crustacea) (Figure 12).

2. Research question

The working hypothesis is that since underwater wood changes the hydrology of his surrounding, the sediment texture of the river bed will be affected (Abbe and Montgomery, 1996), creating different mesohabitats downstream and directly upstream of the wood log. The increased in stream habitat heterogeneity, creating a wider niche availability (Lemly and Hilderbrand, 2000), should enhance a higher invertebrate biodiversity, or at least a detectable different composition than the river background.

This should lead also to a detectable difference in the community abundance and composition (Harper and Everard, 1998) based on the local physical characteristics, so that communities at the same distance from the LWD but inhabiting different mesohabitats, should be significantly different. It is also expected to be found a gradient, with differences among communities leveling out with increasing distance from the log – related effects.

3. METHODS

3.1. Sampling Site

The sampling site chosen is the river Pliszka, an affluent on the Polish side of the river Oder. The samplings were carried out in a reach not far from the mouth of the river (Lat. 52°14'59.24"N, Lon. 14°43'57.11"E), in an area easily reachable by car, but with no direct influences from human activities or structures.

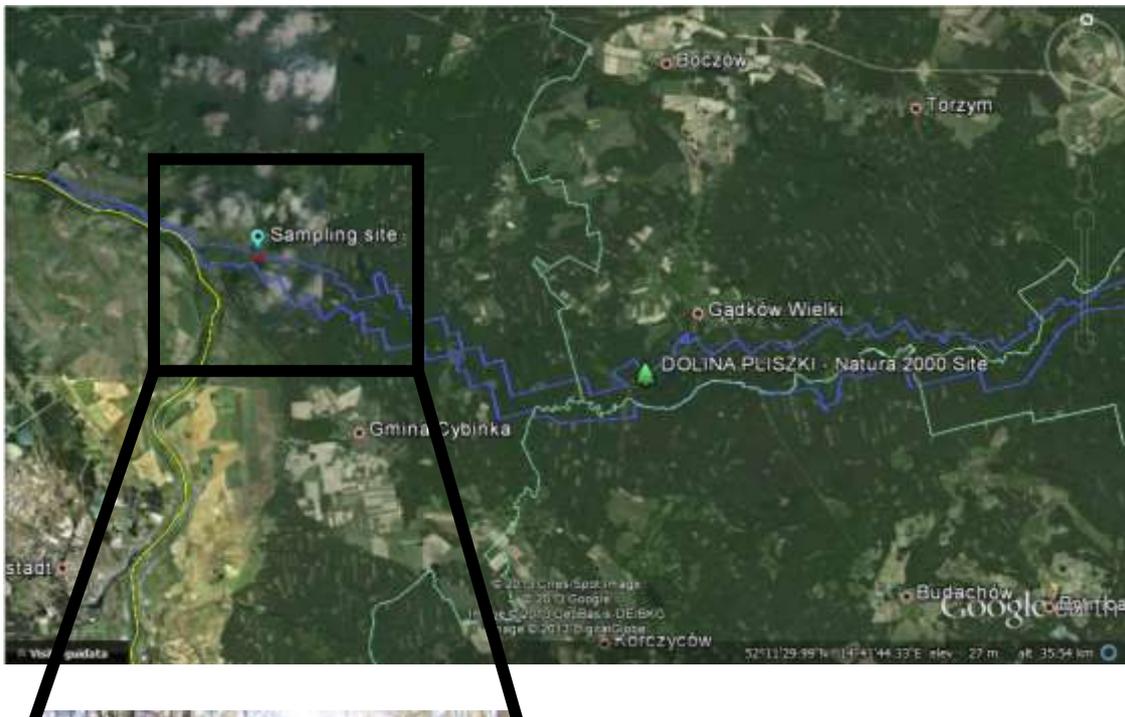


Figure 17 The sampling site. Top picture: In blue the boundaries of the Dolina Pliszki Natura 2000 Site. Lower picture: The sampling site at the level of the third LWD.

This river was chosen because is a good example of small² lowland river, ecologically well preserved. The river is situated between 70 and 28 m of altitude (European Environmental Agency (EEA), 2008), while the length is of about 65.7Km (Ziemia Lubuska Region, 2013), with a catchment area of 241.8 km² and a geology principally siliceous (Szozkiewicz *et al.*, 2004). The human activities in the catchment are mainly woodland plantations (Non-native Forests, almost 50%), and croplands (18%), but anyway Native Deciduous Forests are still almost the 30% and urban areas are less than 5%. In the floodplain properly, forest is by far dominant (Szozkiewicz *et al.*, 2004).

The river is the core of a Natura2000 site (PLH080011 – Dolina Pliszka), protecting several endangered species of fishes, invertebrates, plants and mammals (European Environmental Agency (EEA), 2008). In particular, beavers are present that have a clear heavy impact on the supply of Logs and other big wood pieces to the riverbed.

Other criteria for the selection of this particular reach were the requirements of this study: high numbers of logs lying in the river, dominant sandy substrate and high amount of in river wood logs. In the studied reach, the Pliszka present a forested shoreline, width of around 10 m and a low energy. This allow fallen logs not to hang above the water level and, in connection with the low flow energy, not to be easily dislocated from the perpendicular to the flow. Its reduced dimension allow also the wood to produce a strong influence on the morphology of the river, unlike wider lowland rivers where the influence of logs is limited to the riverbanks (Piégay and Gurnell, 1997).

Three LWD were selected within the 100 m reach. They had similar characteristics: oriented perpendicular to the flow, at least partially lying on the river bottom, not in close proximity to other morphological features that could influence flow and bed morphology (e.g. other logs, debris dams and meanders).

² A “small” river is defined a river where most of the LWD are longer than the channel width Gurnell and Petts (2002)).

3.2. Sampling methods

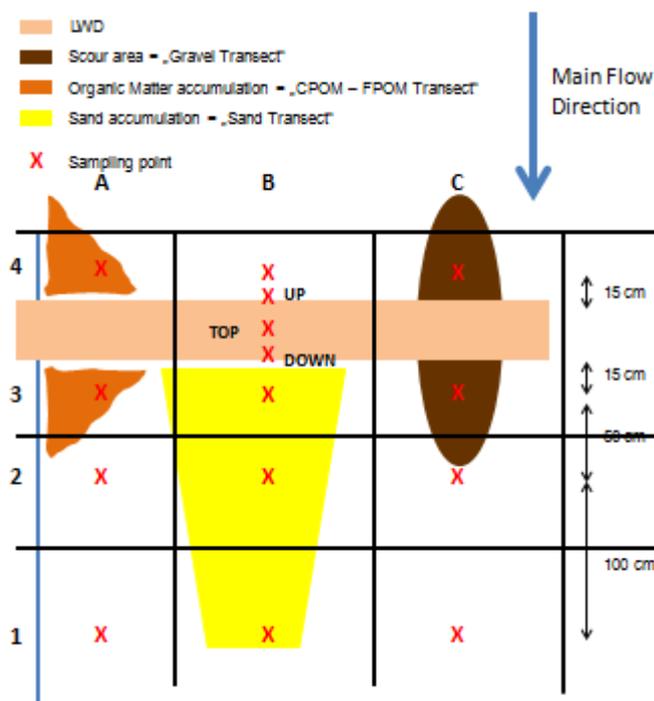


Figure 18 Sampling design.

Between November and December 2012 we collected 45 samples of invertebrate and sediment from the three LWD.

The sampling scheme devised was a grid of points around every chosen log (Figure 18). After having identified the three main mesohabitats created by the log (Sand, Gravel, CPOM-FPOM), three transects (A, B, C) were identified parallel to the flow (Figure 18). From now on, these transects will be referred to with the mesohabitat name to which they belong (transect “Gravel”, “Sand” and “CPOM – FPOM”). The distance relative to the LWD was fixed for every sampling point, thus identifying also 4 transects perpendicular to the flow (Table 1). In every sampling point was also measured the water depth.

Transect 1	160 cm downstream
Transect 2	60 cm downstream
Transect 3	10 cm downstream
Transect 4	10 cm upstream

Table 1 Distance of the sampling points from the wood. Every transect is composed by three samples, one for every habitat transect.

With this scheme, to every sampled point could be assigned an “Habitat” (Gravel, CPOM-FPOM, Sand, Log) and a number representing the distance from the wood. The result was so 3 replica for every sampling point relative to the wood (e.g. three Gravel1, one from every LWD sampled). In

addition to these, also 3 sample for every LWD were taken from the wood surface (Figure 18) and 3 samples were taken in an area as far away as possible from any other influence (wood, reach, meander etc.) as control.

3.2.1. Invertebrate Samples

For the Invertebrate samples, a Surber sampler has been used with a mesh of 500 μm . The front sampling area was 24x24 cm and the sampling was repeated 5 times around each sampling point (total of 0.29 m^2). The Surber has been located on the bottom with the front iron frame against the flow, while with an iron stick all the sediment enclosed in the frontal area has been strongly “plowed”. The objective was to dislodge all the animals living in and on the sediment, while the current carried them into the net.



For the samples from the wood surface, a similar strategy has been followed, with the only difference that a hard brush was used to dislodge the animals from the surface and collected with the same Surber. The surface sampled was the same (0.29 m^2).

The samples collected (biotic and abiotic part) were than stored in plastic bottles, filled with Ethanol 70%.

In each sampling point on the stream bottom, also a sample of the sediment has been taken. A corer sampler has been used, with an opening of 10cm. Sediment was collected for the first 5 cm (Friberg *et al.*, 1998), than saved in plastic bags and frozen to avoid decomposition of the organic matter.

3.2.2. Flow Velocity

Parallel to the macroinvertebrates, flow measurements were carried out at the same sampling points. With a **FlowTracker Handheld-ADV**[®] 3D data on the near – bed flow velocity were collected, with a frequency of 1 measure every 1s (1Hz) for 2 minutes.



The **FlowTracker Handheld-ADV**[®] uses acoustic Doppler technology to measure flow velocity of a small sampling volume at a fixed distance from the probe (10cm). Sound bounces on the particles suspended in the water, the reflected sound is recorded by the probe, averaged by the software and results in the flow velocity of the water for the three axis (SonTek/YSI, 2013).

3.2.3. Habitat Survey

A visual survey of mesohabitats of the full width of the channel for 2 meters upstream and 2 meters downstream from the wood was carried out. Squares of 1m² were drawn around the LWD (Figure 19), creating two transects upstream and two downstream. Then the area was visually divided in mesohabitats, assigning a percentage relative to the coverage of that habitat for the square. The area was divided among Sand, FPOM, Gravel, Log, CPOM. Other information like secondary coverage (e.g. Macrophytes, suspended branches, etc.) and water depth were also recorded.

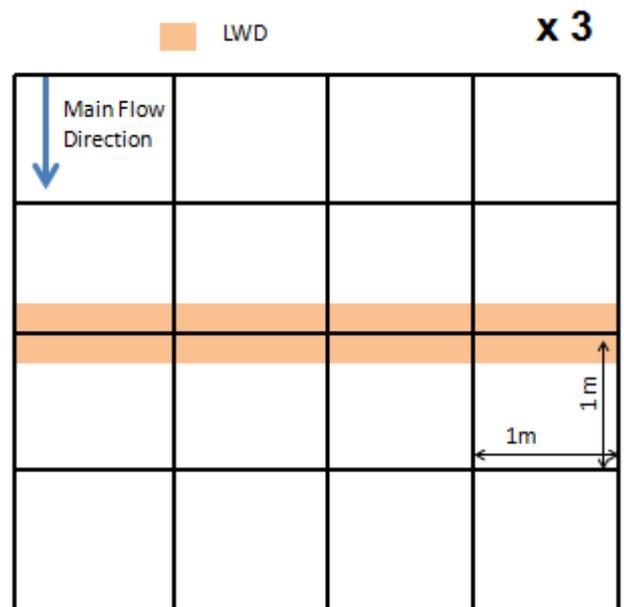


Figure 19 Habitat survey scheme. Every square is 1sqm. Total primary substrate always covered 100%, while secondary substrate could be partial.

3.3. Elaboration of the samples

After the field campaign, samples were taken to the laboratory for further elaboration.

3.3.1. Invertebrate samples

Invertebrate samples stored in alcohol have been washed with water in a 500 μm sieve, to remove alcohol and finer material. Samples with high quantity of sand have been “washed”: in large plastic trays water was added to the sample, the tray was thoroughly shaken and the supernatant was poured in a 500 μm sieve. The procedure was repeated until several consecutive washing (at least 5) didn't produce any material on the sieve. Residual sand was randomly checked to assure that no macroinvertebrates were overlooked.

Using a stereo microscope, every sample was checked for animals, separating the invertebrates from organic matter and sediment. The animals have been counted and identified, when possible at the specie level, or at the lower level possible given the keys available (see Annex 6 for a bibliography list of the books and keys used). The samples, divided by main taxa, have been also preserved in 70% ethanol.

3.3.2. Sediment samples

Sediment samples have been processed in order to get information about grain size and organic content. The protocol followed for the analysis of the sediment samples was the following:

1. The frozen samples were dried for at least 36 hours at 60°C
2. Every single sample has been dry-sieved with a sieving shaker for at least 20 minutes to separate every different granulometry, than collected. Dimension of the sieves it's reported in the side table.
3. After a small period at 60°C again (to dry eventual humidity collected during the sieving) all the fractions have been weighted.
4. To evaluate the content of fine and coarse particulate organic matter contained (FPOM and CPOM), the fractions from >20mm to >1.12mm (CPOM) and from >630 μm to <63 μm (FPOM) have been cumulated in two different crucibles, weighted (dry weight), burned at 550°C for 5 hours and then weighted again(ash free dry weight). An Excel sheet has been prepared containing all the sediment data, calculating also the total organic matter and the D16, D50 and D84 of every sample (see Table 2).

Dimension
>20 mm
>11.2 mm
>6.3 mm
>3.55 mm
>2,3 mm
>1,12 mm
>630 μm
>355 μm
>200 μm
>112 μm
>63 μm
<63 μm

3.4. Data Analysis

After collecting all the raw data (Abundance of every taxa in every sample), several metrics were calculated and then collectively analyzed with the program Asterics (fliessgewaesserbewertung.de, 2013). Other values representing sediment data and flow velocity were then added to the database.

Metric	Description
Shannon-Wiener Index	$H' = - \sum_{i=1}^R p_i \ln p_i$ Diversity index
Evenness	$J' = \frac{H'}{H'_{max}}$ Diversity Index
Feeding Types (%)	Divided among Grazers & Scrapers, Miners, Xilophagous, Active Filter feeders, Passive Filter feeders, Total Filters, Predators, Parasites, Other and No Data. This metric was calculated using the ASTERICS software (fliessgewaesserbewertung.de, 2013) ³ . Every taxa get a score from 0 to 10 subdivided between the different feeding behaviours known.
Number of Taxa	Sum of the number of taxa identified.
Abundance	Number of invertebrates found.
D50 (D16, D84)	Average median grain size (size such that 50% or 16% or 84% of the sediment is finer) (Buffington and Montgomery, 1999).
Organic Matter (%)	Calculated as the difference between dry weight of the sediment and ash free dry weight after 5 hours at 550 °C. Divided in Total, Fine (<1.12mm) and Coarse(>1.12mm).
Mean Velocity (or Mean V)	Square root of the sum of the square of the three velocity components: $\sqrt{x^2+y^2+z^2}$
Standard deviation of the flow (or sdVx)	Standard Deviation of the stream – wise flow velocity (sdVx), as a proxy for turbulence (Hart <i>et al.</i> , 1996; Enders <i>et al.</i> , 2003).
Sorting Index	This index shows the sorting level of the sediment, and it is calculated as S. I. = $\frac{D84-D16}{2}$ (Gordon (2004) in Liébault and Piégay (2001)).

Table 2 List and description of the metrics used in this study.

After the creation of the dataframe, statistical analysis were performed using the software R (R Core Team, 2012).

In the creation and testing of models, samples from the wood surface were not aggregated with the samples of the sediment since some variables (Organic Matter and D50 in particular) were not applicable, and to avoid an increase of the variability that could obscure smaller variations in the

³From the ASTERICS manual: “Autoecological information are predominantly taken from:
 FIRST PRIORITY: Moog, O. (Ed.) 1995. Fauna Aquatica Austriaca – a comprehensive species inventory of Austrian aquatic organisms with ecological data. First edition, Wasserwirtschaftskataster, Bundesministerium für Land- und Forstwirtschaft, Wien.
 SECOND PRIORITY: SCHMEDTJE, U. & M. COLLING 1996. Ökologische Typisierung der aquatischen Makrofauna. Informationsberichte des Bayerischen Landesamtes für Wasserwirtschaft 4/96.
 THIRD PRIORITY: Information sampled by the AQEM consortium.

benthic community surrounding the wood. Anyway wood samples were considered in the general information and generic statistics (“General Information” p. 25; “Feeding guilds distribution” p. 30).

3.4.1. Canonical Correspondence Analysis (CCA)

Ordination techniques were used to identify the environmental variables most closely associated with species and sites distribution. After an initial Detrended Correspondence Analysis showed that the axis length of the data set was 2.4, a CCA was applied. Rare Taxa (taxa with less than 5 specimens) were eliminated. A permutation test (ANOVA, 1000 permutations) was run to check the significance ($p < 0.05$) of the full model and of all the axis.

Forward selection of the environmental variables (1000 permutations) was used to identify the variables that exerted a significant influence on invertebrates’ distribution.

In the biplot, taxa and sites are reported with an identification code and the environmental variables are represented with an arrow. Arrows point roughly toward the maximum variation, while the length of the segments represent the influence of that variable.

For this analysis the R package “vegan” has been used (Oksanen *et al.*, 2013).

3.4.2. Analysis Of Variance (ANOVA)

Using the packages “lawstat” (Gastwirth *et al.*, 2013) and “agricolae” for R (Felipe de Mendiburu, 2012), an ANOVA 2-way test have been run to identify specific relations between the calculated metrics against Habitat and Distance (see section “Sampling Methods”). The metrics chosen for this test were Shannon, Evenness, Total Taxa and Abundance. The test was run on all the parameters that showed homoscedasticity, verified with a classical Levene's test based on the absolute deviations from the mean ($p > 0.05$). Data were eventually transformed, if necessary, with Log+1.

3.4.3. General Linear Model

The influence of mean flow velocity (MeanV) and standard deviation (sdVx) on the metrics values was then tested by introducing them as covariable in General Linear Models together with the factors Habitat type and Distance from the wood.

1. Metric~habitat+distance+MeanV
2. Metric~habitat+distance+sdVx

Small-scale distribution of macroinvertebrates on Large Woody Debris and nearby sediment in a lowland river.

Results are shown only for the models that follow the criteria: Adjusted R² of the model >0.25, a p-value of the model <0.05 and normally distributed residuals (Shapiro test: p>0.05). For this modeling the package “agricolae” was used (Felipe de Mendiburu, 2012).

4. RESULTS

4.1. General Information

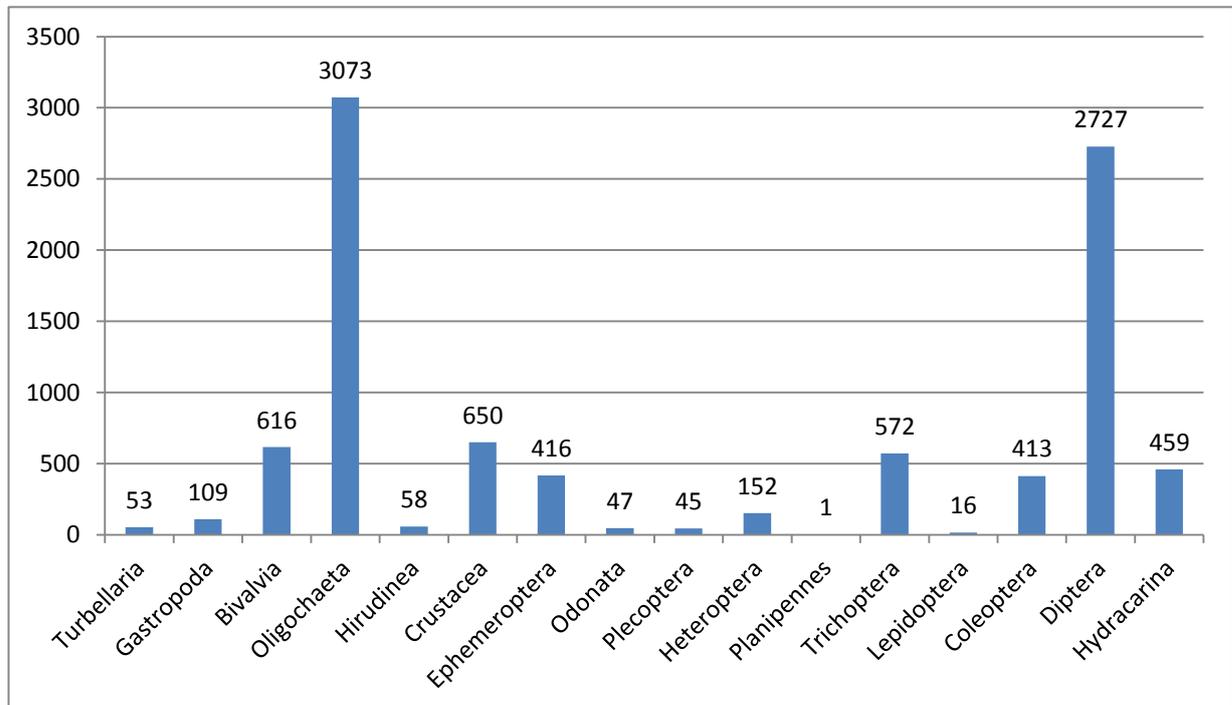
36 samples coming from the sediment around 3 different LWD, 9 samples from the wood surface and three control samples have been analyzed.

Mean water dept was 80cm (max=157cm, min=28cm) and mean flow velocity 0.26 m/s, but with a really high variability: max= 0.60, min=0.06. Other information on the three sampling sites are reported in the following table.

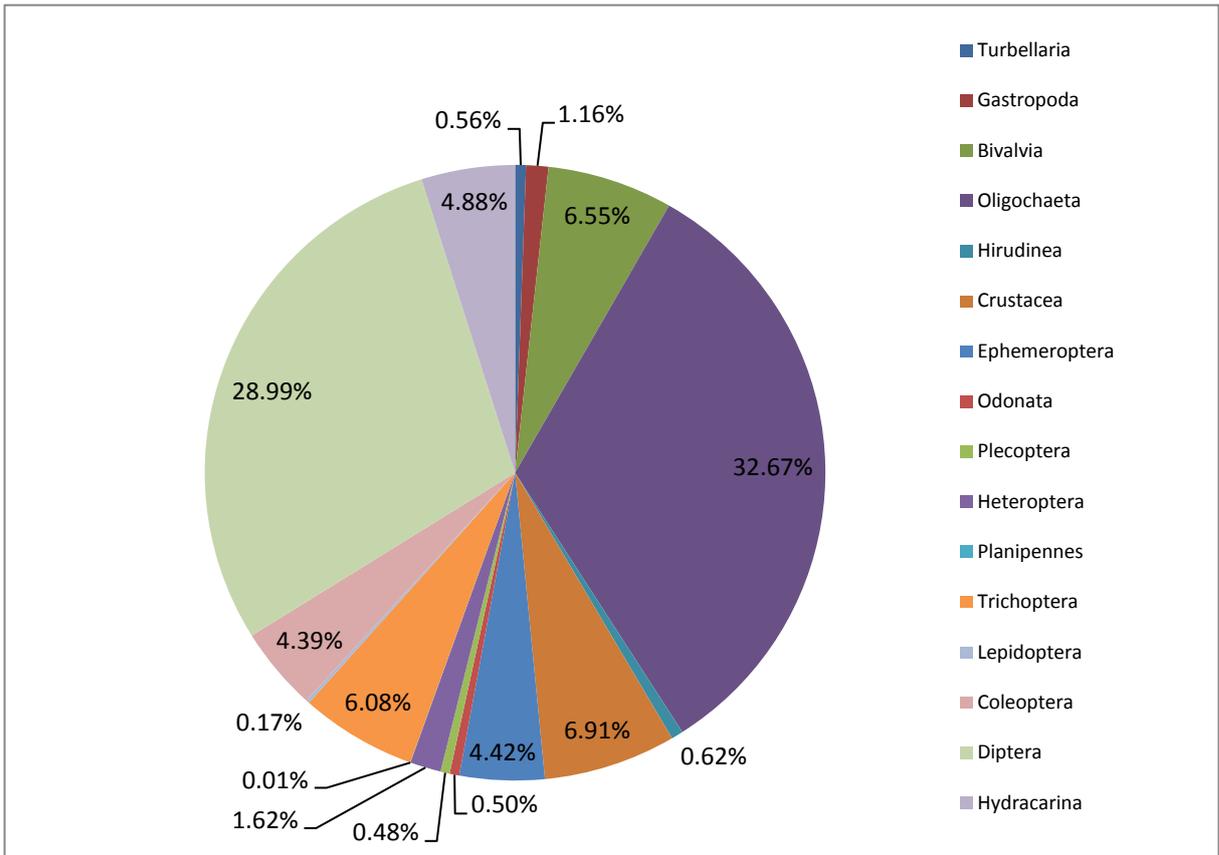
	Log Diameter (cm)	Log Length (m)	Channel width (m)
LWD1	40	13.6	10.5
LWD2	25	9	9
LWD3	23	10	8

Table 3 General information on the sampling sites.

A total of 9407 animals were collected (5655 from the sediment, 3384 from the wood, 368 from the control samples) representing 85 different taxa. The list of all the taxa collected is reported in Annex 1.



Graph 1 Total abundance, divided by taxa. The number is the number of specimens belonging to that taxa.

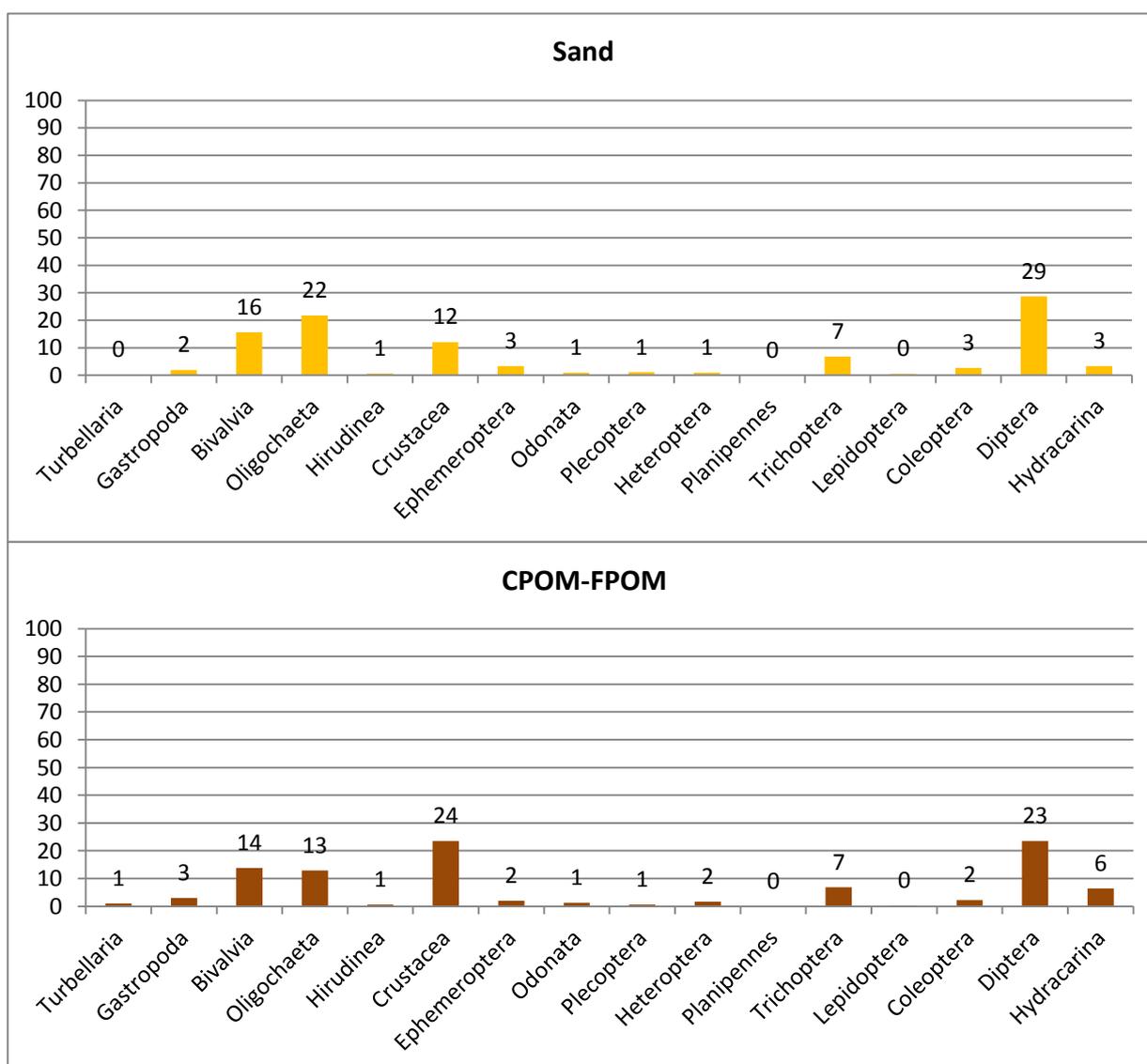


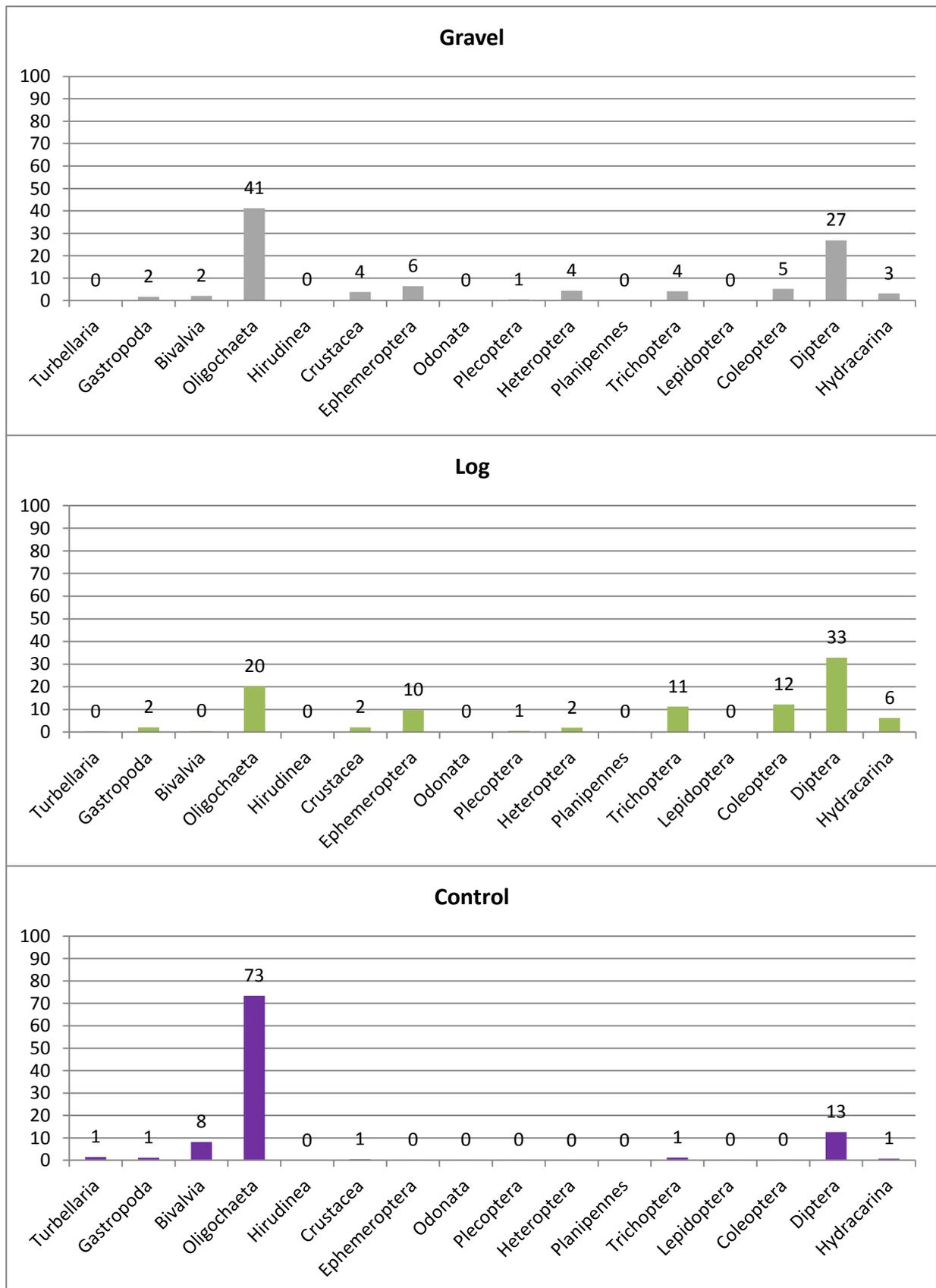
Graph 2 Mean abundance distribution of the macroinvertebrates among the taxa.

On the sediment, the mean number of specimens sampled was 157 (sd 145), on the wood 376 (sd 510) while on the control 122 (sd 80). The most abundant order was Oligochaeta (33%) followed by Diptera (29%). The least common order was Lepidoptera (0.16%), Plecoptera (0.48%) and Odonata (0.50%) (Graph 2). Only one specimen of the group Planipennes was found. In the Table 4 Total Abundance data are reported for every distance and habitat transects.

	Sand	Gravel	CPOM_FP OM	Log	Control	Distance 1	Distance 2	Distance 3	Distance 4
Total Abundance	861	1920	2874	3384	368	1277.0	939.0	1712.0	1727.0
Mean	71.7	160	239.5	376	122.7	141.9	104.3	190.2	191.9
Std. Deviation	82.2	104.1	183.9	509.7	79.8	158.2	71.7	179.9	152.6
Standard Error	23.7	30.1	53.1	169.9	46.0	425.7	313	570.7	575.7

Table 4 Total Abundance data on the specimens sampled, broke down to every habitat transect (Sand, Gravel, CPOM_FPOM, Log) and to every distance transect (1=160cm, 2=60cm, 3=10cm, 4=10cm upstream)





Graph 3 Mean percentage of animals sampled for every habitat transect, divided by taxa.

In Graph 3 is reported the mean percentage of every taxa present in the different habitats. Oligochaeta and Diptera are always dominant with the exception of the CPOM – FPOM transect that present an high presence of Crustacea. Control and, to a less degree, Gravel seems to have a less diverse composition with higher dominance of Oligochaeta and Diptera.

The maximum number of taxa collected in a single site was 36, the minimum 4. In the Gravel sample the mean was 22, in the Sand 16, in the CPOM_FPOM 24, in the Log 23 while in the control 11.

	Sand	Gravel	CPOM – FPOM	Log	Control
Mean Shannon value	2.04	1.98	2.13	2.298	0.976
Mean Evenness value	0.79	0.66	0.70	0.758	0.414

Table 5 Mean values of diversity index considered for every habitat transect, the log and the control.

In both the diversity indexes in Table 5, the habitats close to the wood (first three from the left) have much higher mean values than the control samples.

Sediment granulometry was evaluated through the D50 values and the Sorting Index (S.I.) (see Table 2).

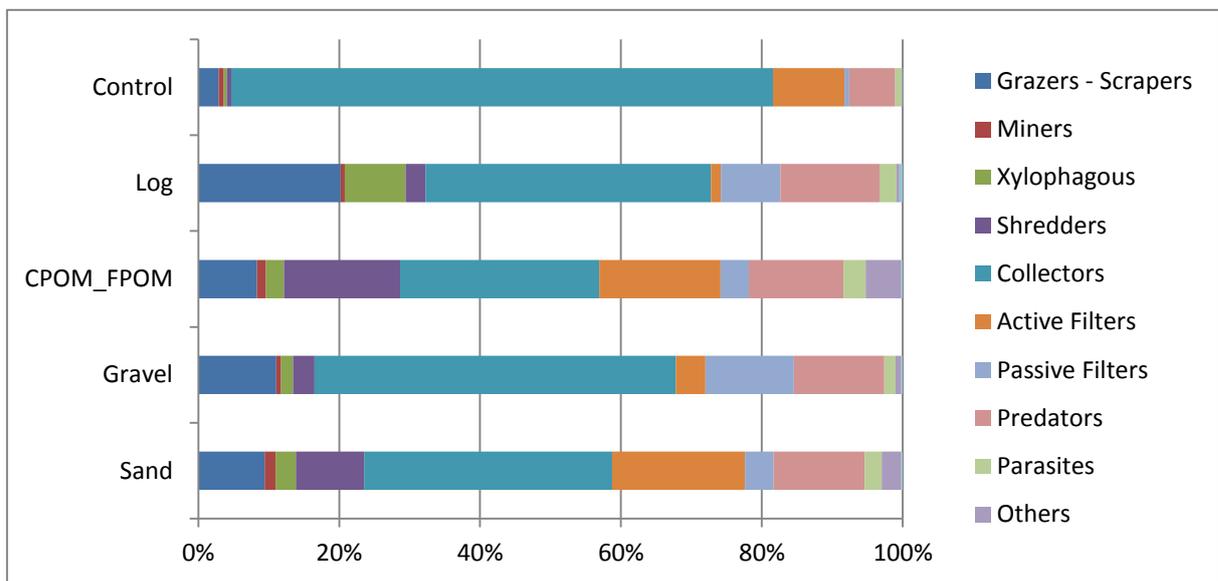
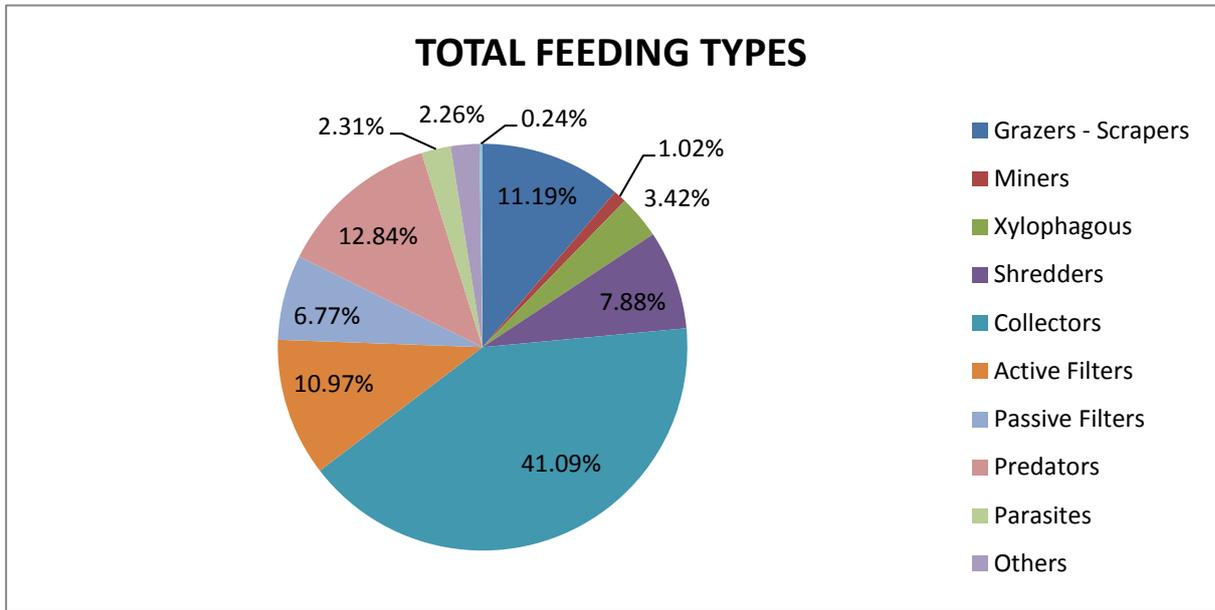
	Sand	Gravel	CPOM - FPOM	Control
Mean D50 (mm)	0.31	2.31	0.24	0.51
Median S.I.	0.17	3.38	0.14	0.35

Table 6 Values for the granulometry of the transects. See Table 1 for the calculation method of D50, D16 and D84.

As can be seen from the table, while CPOM – FPOM show similar values in both the metrics, Gravel on one hand and Control on the other show marked differences.

4.2. Feeding guilds distribution

The percentage of each feeding type is reported in the following graphs both as percentage of the total and divided by habitat.



Graph 4 Percentage distribution of feeding groups. Top: Total mean. Bottom: mean of every habitat transect.

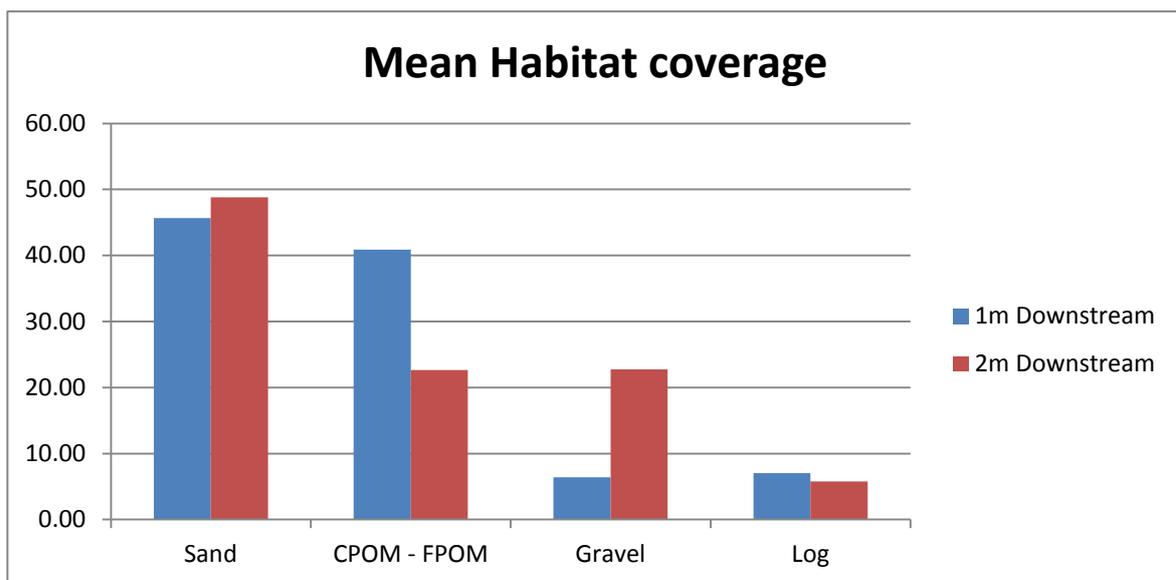
Sample_code	Sand	Gravel	CPOM-FPOM	Log	Control
Grazers - Scrapers	9.45%	11.08%	8.34%	20.26%	2.90%
Miners	1.55%	0.68%	1.25%	0.55%	0.74%
Xylophagous	2.88%	1.68%	2.56%	8.65%	0.38%
Shredders	9.67%	3.06%	16.49%	2.86%	0.68%
Collectors	35.27%	51.25%	28.29%	40.58%	76.90%
Active Filters	18.86%	4.19%	17.26%	1.43%	10.16%
Passive Filters	4.08%	12.54%	3.99%	8.42%	0.67%
Predators	12.92%	12.84%	13.42%	14.12%	6.55%
Parasites	2.44%	1.62%	3.16%	2.41%	0.92%
Others	2.80%	0.82%	5.07%	0.41%	0.10%
No Data	0.19%	0.18%	0.22%	0.48%	0.04%

Table 7 percentage of feeding groups for every transect.

4.3. Habitat Survey

A survey of the habitat coverage was carried out, to get some general information about the relative importance of the habitats evaluated during the sampling (Figure 19 Habitat survey scheme. Every square is 1sqm. Total primary substrate always covered 100%, while secondary substrate could be partial..

To highlight general variations between the closer (0 to 1 m) and the furthest downstream (1 to 2 m) transects, mean total coverage of every habitat were then compared.



Graph 5 Comparison between habitat coverage (%) of the first and the second transect downstream.

Habitat surface follow a similar pattern in both transects, but with some significant differences. Sandy habitats, the most common mesohabitat, slightly increase downstream, as well as the gravel habitats. CPOM - FPOM shows a sharp decrease downstream. The full report with coverage for every square meter and every upstream and downstream transect, as well as values for secondary coverage can be found in Annex 4: Habitat Survey.

4.4. Canonical Composition Analysis (CCA)

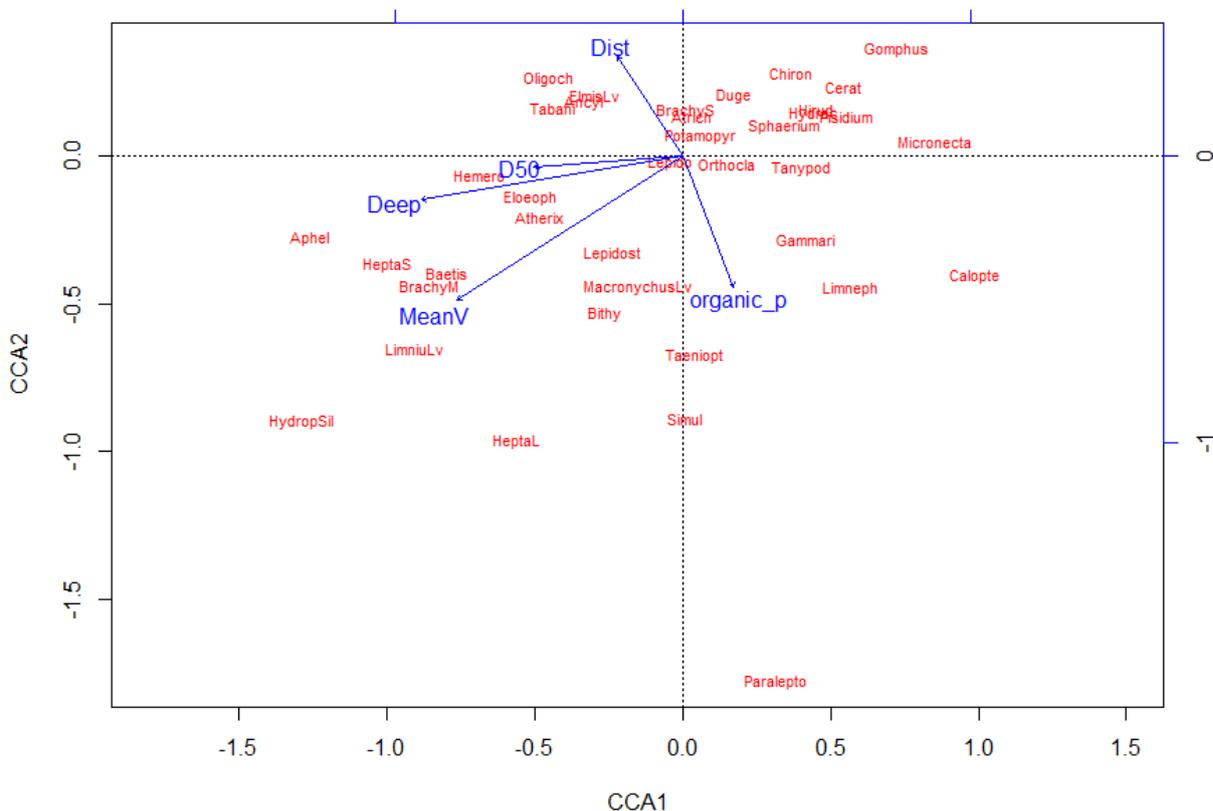
The objective of this analysis was to identify particular distribution patterns of the taxa collected and of the sites distribution in relation to the environmental variables. The scope in the first case is, in particular, to see which taxa are “outsider” and to which environmental variable they are more related. In the second case is to check the presence of different cluster of sites justified by the environmental variables.

Log samples were excluded, as well as rare taxa (n<5). The proportion of variance explained from the constrained CCA is 0.29, and the first 2 axes (CCA1, CCA2) explain 74% of the variance.

	F	P-value
Overall Model	2.7492	0.015
CCA1	6.1644	0.001
CCA2	4.0108	0.009

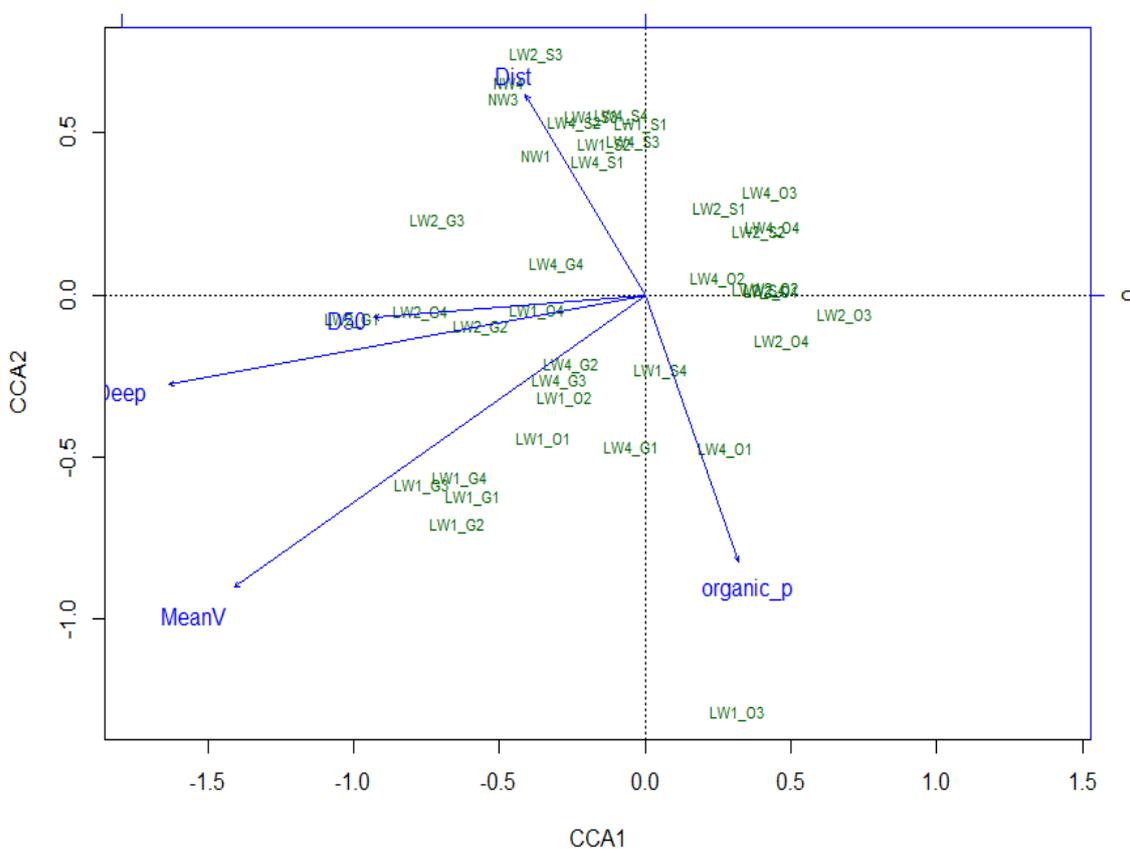
Table 8 Output of ANOVA test on the overall model (1000 permutations) and of an ANOVA by axis (1000 permutations)

The two most important variables along CCA1 are Depth and Mean Velocity, while on the CCA2 are still Mean Velocity and Organic Matter.



Graph 6 CCA plot with the taxa distribution. Taxa are reported with a code that univocally identify that taxon.

The Graph 6 show that D50, depth and Velocity are closely correlated. Several taxa show a relation with these variables: Heptagenia sp. and Baetis sp. (Ephemeroptera), Hydropsiche Siltalai and Brachycentrus Montanus (Trichoptera), Aphelocheirus Aestivalis (Heteroptera) and Limnius sp. Lv (Coleoptera) show the highest correlations with these axis. Along the Organic content axis, the outsider taxa are mainly Paraleptophlebia sp. and Limnephilidae gen. sp. (Trichoptera), Taeniopteryx nebulosa (Plecoptera) and Calopteryx sp. (Odonata). Gomphus sp. (Odonata) and Micronecta sp. (Heteroptera) seems on the other hand have a strong negative relation with both velocity and organic content.



Graph 7 CCA plot with the site distribution. Every site is represented with a code (e.g. LW1_O1) reporting sampling log (e.g. LW1), Habitat transect (e.g. O) and distance transect (e.g. 1). Habitats are reported as Gravel (G), Organic (O), Sand (S), or control (NW). The code “LW4” refers to LWD3.

From the site point of view, the distribution shows three main groups of sites. Gravel sites (G) are mainly distributed along the MeanV, D50 and Deep axis, while organic sites (O) and sand sites (S) show two separated clusters characterized by low flow and depth. They are separated along the CCA2, probably due to a slightly different organic content.

Finally, the forward selection of the environmental variables showed that Depth and Organic Matter are the two variables that explain the most variance and that are the most reliable.

	F	P-value
Deep	3.9693	0.001
organic_p	2.3960	0.041

Table 9 Results of the forward selection for the best model (perm. 1000)

4.5. Analysis Of Variance (ANOVA)

The analysis of variance was performed to identify relations between the metrics and the environmental variables. A 2-way ANOVA approach was used. After checking the assumptions of the test, three metrics were chosen and tested against habitat, distance and the combination of this two (Abundance was discarded for a p-value of the Levene Test <0.05). Samples coming from the wood surface were not considered for this test.

		Habitat	Distance	Habitat+Distance	Residuals
Shannon	Df	3	3	6	26
	Sum Sq	3.3169	0.5267	1.3303	6.698
	Mean Sq	1.10563	0.17556	0.22171	0.25761
	F value	4.2918	0.6815	0.8606	
	Pr(>F)	0.01378	0.57135	0.53634	
Evenness	Df	3	3	6	26
	Sum Sq	0.36016	0.03078	0.10145	0.47246
	Mean Sq	0.120052	0.01026	0.016908	0.018172
	F value	6.6066	0.5646	0.9305	
	Pr(>F)	0.001819	0.643215	0.489853	
Taxa Richness	Df	3	3	6	26
	Sum Sq	727.41	224.56	117.11	1096
	Mean Sq	242.47	74.852	19.519	42.154
	F value	5.752	1.7757	0.463	
	Pr(>F)	0.003715	0.176521	0.829128	

Table 10 F and P values for the different metrics in relation to the different variables. * show p-values <0.05

All the metrics show a significant relation with the habitat transect ($p < 0.05$) while the distance from the wood and the combination between the two present p-values too high to consider the model reliable.

4.6. General Linear Model

For the analysis of the covariance, the same parameters and samples were considered as in the ANOVA test: Shannon, Evenness, Taxa Richness and Abundance. However, abundance was than discarded due to the low p-value of the Shapiro-test, always lower than 0.05. Here we report the results of the tests, first for the Mean Velocity and than for the standard deviation of the streamwise velocity.

4.6.1. Mean Velocity

	F value	Df	P value	Habitat (P value)	Distance (P value)	MeanV (P value)
Shannon	3.046	31	0.0147	0.0069	0.5176	0.0446
Evenness	5.902	31	0.0002	0.0002	0.5201	0.0016
Taxa Richness	3.65	31	0.0055	0.0018	0.1413	0.4143

	Shannon				Evenness				Taxa Richness			
	Estimate	Std. Error	t value	Pr(> t)	Estimate	Std. Error	t value	Pr(> t)	Estimate	Std. Error	t value	Pr(> t)
(Intercept)	0.56814	0.06894	8.241	2.62E-09	1.74823	0.28432	6.149	8.04E-07	26.0628	3.6932	7.057	6.33E-08
Habitat= Gravel	0.18698	0.06361	-2.939	0.00616	0.52125	0.26232	-1.987	0.05582	-0.7742	3.4075	-0.227	0.82175
Habitat= Sand	0.14217	0.0496	2.867	0.00739	0.04061	0.20453	0.199	0.84391	-9.181	2.6568	-3.456	0.00161
Habitat= sandNW	0.40019	0.08426	-4.75	4.40E-05	1.38496	0.34747	-3.986	0.00038	12.2494	4.5135	-2.714	0.01076
Distance 2	0.04566	0.05464	-0.836	0.40976	0.19291	0.22534	-0.856	0.3985	-3.5352	2.9271	-1.208	0.23627
Distance 3	0.01511	0.05574	0.271	0.78819	0.2008	0.22987	0.874	0.38909	2.6937	2.986	0.902	0.37395
Distance 4	0.04707	0.05501	-0.856	0.3988	0.01415	0.22687	-0.062	0.95065	1.7638	2.9469	0.599	0.55383
MeanV	0.73833	0.213	3.466	0.00157	1.8386	0.87842	2.093	0.04462	-9.4422	11.4104	-0.828	0.41427

Table 11 Top: F value, Df and P value of the model. P values of the variables. Bottom: coefficients.

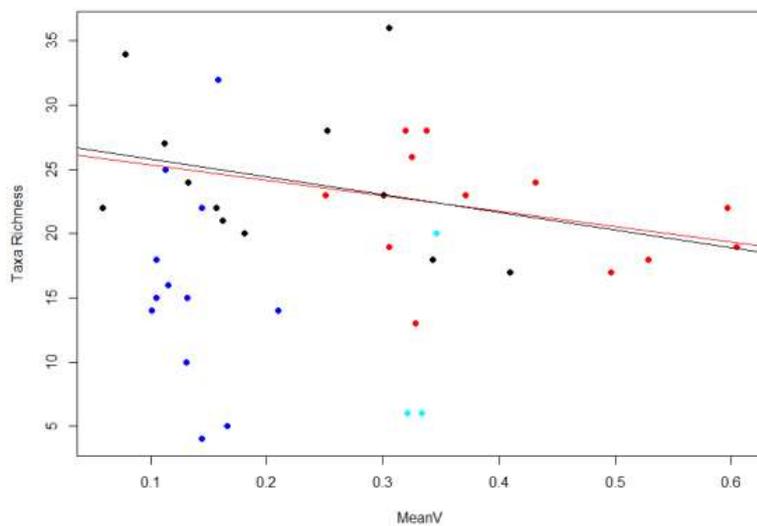
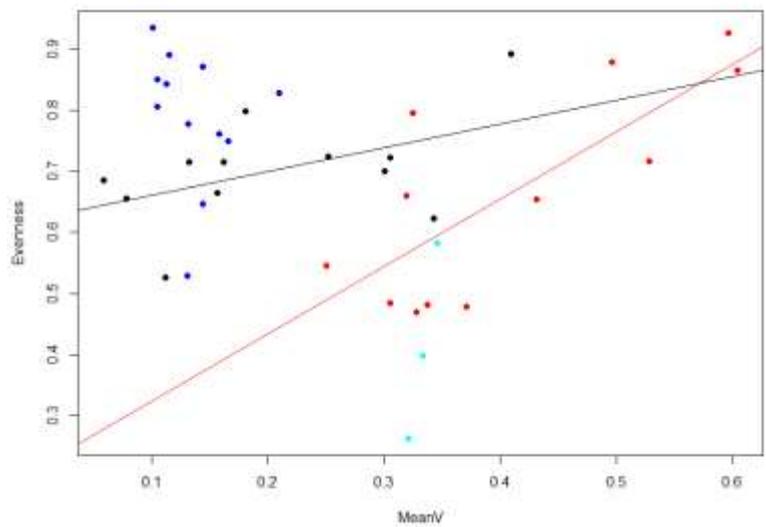
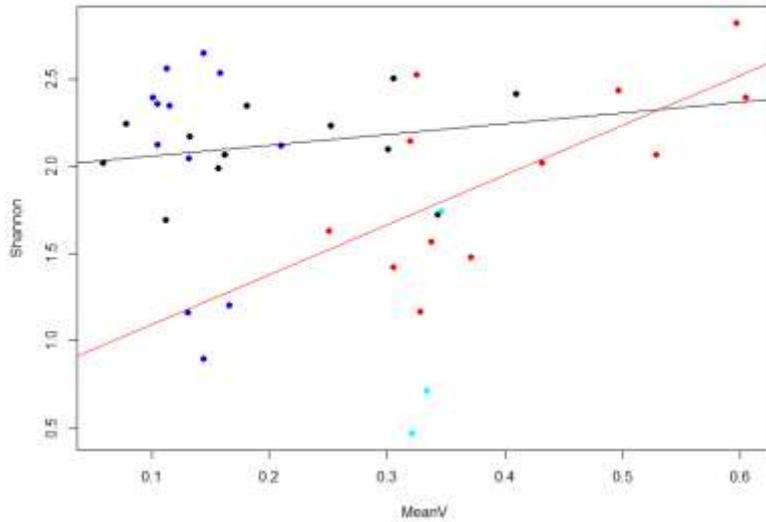
4.6.2. Standard deviation of the flow

	F value	Df	P value	Habitat (P value)	Distance (P value)	sdVx (P value)
Shannon	2.864	31	0.0200	0.0030	0.5834	0.3756
Evenness	3.254	31	0.0104	0.0013	0.6378	0.3275
Taxa Richness	3.509	31	0.0069	0.0020	0.1467	0.7166

	Shannon (Log+1)				Evenness				Taxa Richness			
	Estimate	Std. Error	t value	Pr(> t)	Estimate	Std. Error	t value	Pr(> t)	Estimate	Std. Error	t value	Pr(> t)
(Intercept)	1.229123	0.110288	11.145	2.27E-12	0.586535	0.047725	12.29	1.88E-13	3.24046	0.22419	14.454	2.55E-15
Habitat= Gravel	-0.00148	0.101154	-0.015	0.988425	0.000468	0.043772	0.011	0.991538	-0.03743	0.20562	-0.182	0.85674
Habitat= Sand	-0.03094	0.078609	-0.394	0.696601	0.058122	0.034016	1.709	0.097508	-0.48608	0.15979	-3.042	0.00475
Habitat= SandNW	-0.51485	0.132124	-3.897	0.000486	-0.21247	0.057173	-3.716	0.000798	-0.84562	0.26857	-3.149	0.00362
Distance 2	-0.09038	0.088098	-1.026	0.312871	-0.03695	0.038122	-0.969	0.339975	-0.14994	0.17908	-0.837	0.40883
Distance 3	0.016929	0.089397	0.189	0.85104	-0.02022	0.038684	-0.523	0.604929	0.17566	0.18172	0.967	0.34121
Distance 4	-0.04136	0.090606	-0.456	0.65122	-0.05183	0.039207	-1.322	0.195884	0.14512	0.18418	0.788	0.43672
sdVx	-1.22469	1.362205	-0.899	0.375559	-0.55946	0.58946	-0.949	0.349906	-1.44998	2.76902	-0.524	0.60425

Table 12 Top: Analysis of the variance table. Bottom: coefficients.

From the results, we can see that Habitats and mean velocity reveal relations with some of the metrics. Several scatterplot have then been created, to visually show the correlations.



Graph 8 Scatterplots of sites with different metrics plotted against MeanV. Points of different colors belongs to different Habitats Transects: Black= Organic, Red= Gravel, Blue= Sand, Light Blue= Control. Gravel shows some kind of correlation: higher velocity correspond to higher Shannon and Evenness values. Also Organic seems to follow a similar trend, although less clearly.

5. DISCUSSION

5.1. Invertebrates distribution and feeding guilds patterns.

As expected, the macroinvertebrates mean abundance was much higher in wood samples than in sediment samples (Table 4). This result is coherent to several studies that highlighted the importance of wood in sand bed streams as a biodiversity hotspot (Benke *et al.*, 1984; Weigelhofer and Waringer (1999) in Benke and Wallace (2003); Piégay and Gurnell, 1997). In turn, the sediment samples around the wood were on average richer in taxa than control samples, and this result seems to confirm the assumption that wood has some direct or indirect influence on the surrounding environment. The cause of this higher richness it is probably the greater structural complexity triggered by the presence of the wood (Schneider and Winemiller, 2008).

Strangely, the mesohabitat at a first glance closer to the control samples (the “sand” transect) had a significantly lower invertebrate abundance than control samples (71.7 against 122.7, Table 4). This can be due either to a different stability of the substrate, but it can maybe be caused by an higher predation in the sand transect than in the control. In fact, the taxa richness in the sand was on average 16, while in the control only 11, so the higher richness was given almost exclusively from the higher number of Oligochaeta. Predators In the control were only the 6.5%, while in all the other samples they were always between 12.9% and 14.1%. Another factor that can maybe explain this difference is the Sorting Index. As shown in Table 6, the index has a significantly lower value (around 50% lower) in the Sand transect than in the Control samples, presenting a more uniform granulometry in the first than in the latter.

From a mere numerical point of view, Oligochaeta and Diptera were always the dominant taxa, particularly in gravel, log and control transects. Sand and CPOM-FPOM were the only transects to show other taxa numerically comparable: Bivalvia and Crustacea.

More significant can be the distribution of feeding types. Globally, this reach of the Pliszka River is highly dominated by collectors (41.1%) and filters (Active 11% + Passive 6.8% = 17.8%). The reach, due to the small size and the high shading, can be than considered a typical heterotrophic river. If we consider the River Continuum Concept (Vannote *et al.*, 1980), this river can be however sited together with large, lowland rivers, since the dominating feeding guilds are all based on processed organic matter (FPOM). These values are in agreement with Benke *et al.*, (1984) who found that sandy and muddy sediments were always dominated by collectors, with little contribution by filters.

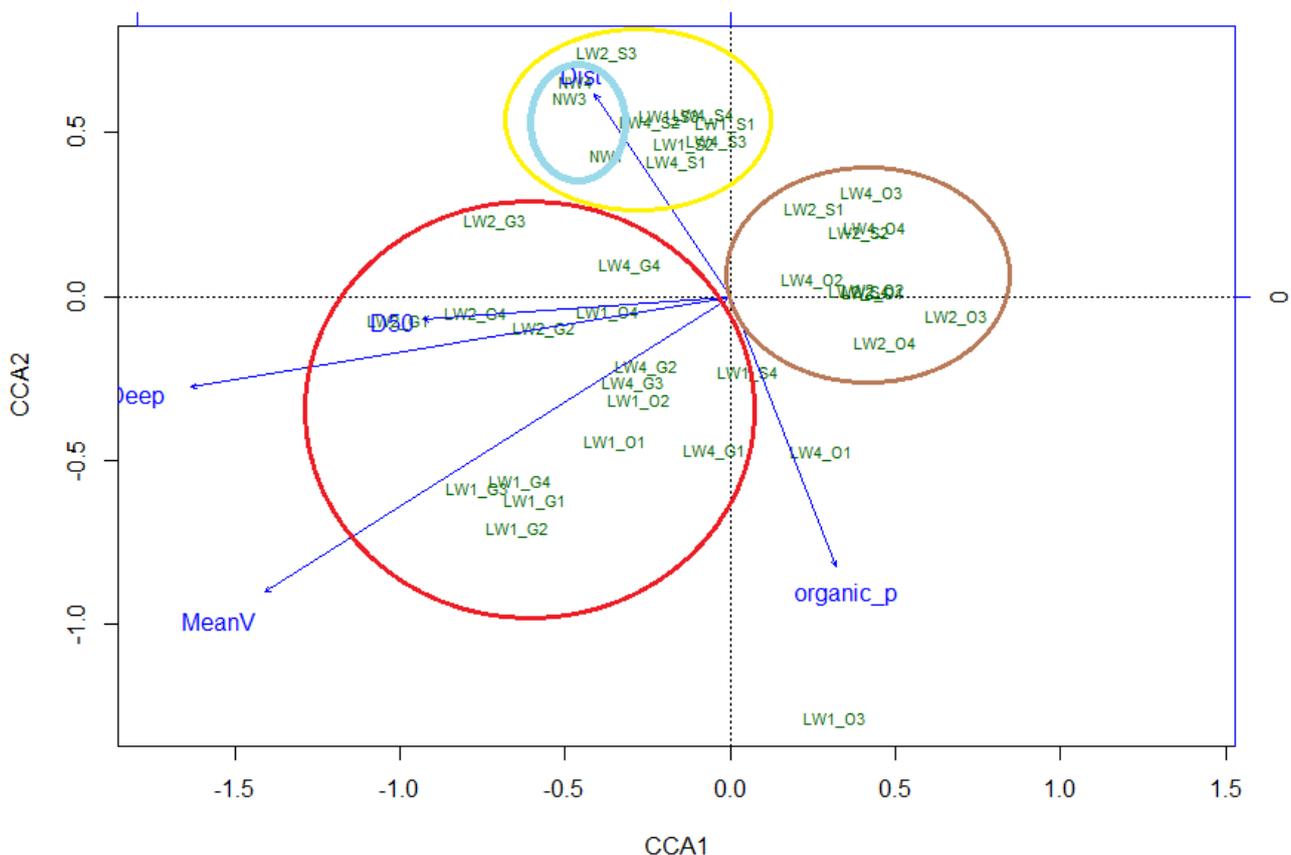
Here, also gravel transects followed a similar pattern, and it can be due to the fact that the sediment size was not so different among the transects (see Table 6).

Wood form a stable substrate that enhance the production of high quality biofilm (Hax and Golladay, 1993). As expected, wood surface samples show an higher presence of grazers – scrapers and xylophagous invertebrates, even if in other studies were found much higher percentages of xylophagous and of shredders (Warmke and Hering, 2000). This lower value of wood-eating taxa it is probably due to a different evaluation of the xylophagous trichoptera *Lype sp.* In fact, this is one of the few recognised taxa that actually eats wood, but while Moog (1995), principal source of the ASTERICS software, consider the specie only partially Xylophagous (2 points out of 10), Warmke and Hering (2000) study supports a probable exclusive wood diet. These data, however, are in complete accord with experimental studies conducted on wood colonization, which found logs dominated by collectors with contribution by scrapers and xylophagous (Bond *et al.*, 2006).

Shredders have an higher incidence in CPOM-FPOM samples and passive filters peak in the gravel transect. Predators seems evenly distributed among all the samples, except the control sample. This distribution seems related to both the river power and the type of substrate. As described in Harper and Everard (1998), rivers like the Pliszka that have low power and high shaded area are dominated by collectors. Anyway relatively higher percentage of different feeding groups were found in microhabitats with higher power (Gravel transects with filters) that had faster flowing water (Mean Velocity: Sand 0.14 m/s, CPOM-FPOM 0.21 m/s, Gravel **0.41** m/s, Control 0.33 m/s) or with particular substrates (CPOM-FPOM with shredders and Log samples with scrapers).

5.2. Mesohabitats

The arbitral division of the sediment among Gravel, Sand and FPOM-CPOM mesohabitats seems strongly supported by the CCA analysis. From the graphical output, three main groups are easily identified, each one encompassing most if not all the sites from every habitat transect.



Graph 9 CCA plot with the site distribution. Clusters are highlighted with circles.

All the Gravel sites (red circle) are distributed along the mean velocity, Depth and D50 axis, while Sand (yellow) and CPOM-FPOM (brown) are characterized by low depth and velocity. These two groups are divided along the organic content axis. Control sites (Light blue) are grouped together with the sand sites, also supporting the assumption that the sand habitat is the closest to the background habitat of this reach.

From the forward selection, Depth and Organic Content are the best variables to explain the variance. D50 and Mean Velocity were not selected probably because strongly connected with the

water depth. In fact, the Gravel transects, characterized by higher D50 values, were positioned in scour areas of the river (higher water depth), where flow was forced to pass under the LWD, accelerating and causing erosion of the riverbed.

5.3. Diversity metrics and environmental variables

The subdivision among habitat is significant for the invertebrate Diversity (Shannon), Evenness and Taxa Richness, as showed by the ANOVA analysis (Table 8). On the other hand, the distance from the wood in all the statistical analysis applied (ANOVA, General Linear Model and CCA) it is not significant, demonstrating that in this study a longitudinal division of the habitats downstream a LWD can be justified, while up to a range of 160cm downstream the wood and 10cm upstream there is no particular variation of the invertebrate metrics. Anyway, the habitat survey (Graph 5) showed an actual simplification of the habitat distribution moving further downstream from the LWD. Probably a more complete comparison of macroinvertebrates assemblages between these two transects (0 to 1m and 1 to 2m) would show a different community composition.

Contrary to Shannon, Evenness and Taxa Richness, Abundance doesn't relate to any physical variable. This results agree with other studies (Álvarez-Cabria *et al.*, 2011), that suggested no relation between abundance and abiotic conditions.

The General Linear Model revealed that Standard Deviation of the stream-wise flow (sdVx) doesn't relate to any of the invertebrate variables chosen. This could mean that invertebrate assemblages are more influenced by habitat and flow velocity than by turbulence, however some studies (e.g. Brooks *et al.*, 2005) demonstrate a relation between taxa and turbulence, particularly a negative one (the community seemed to prefer areas with lower turbulence). So, more likely, the results show a weakness in the recording method. Since the FlowTracker Handheld-ADV® used records with a frequency of 1 Hz, this frequency it is probably too low to give reliable data on the turbulence at such high scale. In alternative, the sdVx reliability as a proxy for turbulence can be not good. (Reid and Thoms, 2008) suggest the use of standard deviation of the vertical flow (sdVy) instead of the sdVx. Analysis using this other variable were tried, but with no better results.

Mean flow velocity showed a close relation with Shannon and Evenness values, and as can be easily read from the Graph 8, this relation is a positive one: higher flow velocity means higher values of Shannon and Evenness. This is particularly true for the Gravel transect, the most influenced by the flow velocity. The gravel transect can be considered a riffle habitat, and flow velocity has been

already proved to be an important variable in defining invertebrate communities (Brooks *et al.*, 2005; Effenberger *et al.*, 2006). Taxa richness however doesn't respond in our study to MeanV.

5.3.1. Particular Taxa preferences⁴

The CCA analysis revealed some taxa positively or negatively correlated with our environmental variables (Graph 6).

- *Positive relations with flow velocity (MeanV)*

Hydropsyche siltalai DÖHLER, 1963 (Trichoptera) positively respond to flow, appearing highly related. This pattern has already been documented, and it is probably due to its feeding behaviour (passive filter) (Statzner and Bretschko, 1998).

Brachycentrus montanus KLAPÁLEK, 1892 (Trichoptera), it is already known to be bound to areas with strong current, and to prefer gravel areas. This is also probably related to his feeding behavior, principally a passive filter feeder.

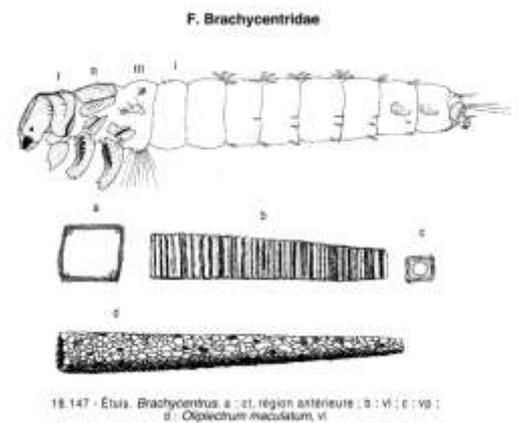


Figure 20 *Brachycentrus sp.* larvae and appearance of the case (a,b,c) (Tachet *et al.*, impr. 2010)



Figure 21 *Aphelocheirus aestivalis* (FABRICIUS, 1794). Adult and head details (Tachet *et al.*, impr. 2010)

Heptagenia longicauda (STEPHENS, 1835) and *Heptagenia sulphurea* (MÜLLER, 1776) (Ephemeroptera) are usually found on boulders or coarse sediment (are grazers) but, unlike what was found in this study, are normally thought to prefer slow flowing areas. The flow stress here it is probably overcome by an higher availability of grazing surfaces, in an otherwise fine sediment river.

Aphelocheirus aestivalis (FABRICIUS, 1794) (Heteroptera). The preferred habitat of this predator it's deep and fast flowing water, and it is verified by these samples.

Limnius sp. (Coleoptera) are also known to prefer moderate to fast flowing water, and since their feeding guild is grazers, it can be

⁴ If not otherwise stated, autoecological information are taken from Schmidt-Kloiber and Hering (2013). A detailed list of the bibliography sources for the information can be found in Annex 4: Autoecological Bibliography

related to the fact that in this environment availability of grazing surfaces (gravel, boulders, etc.) it's normally higher.

- *Positive relation with Organic Matter Content Percentage (organic_p)*

Paraleptophlebiasp. (Ephemeroptera) is the most important outsider in our CCA analysis. In our study, this specie seems slightly more related to the organic matter presence than to the flow velocity, and it can be because of his feeding behaviour as a gatherer/collector feeding on CPOM and FPOM. Actually, *Paraleptophlebia* species have been found to prefer fast flowing waters with leaf litters (Holomuzki and Messier, 1993), preferences that are completely in agreement with these results.

Limnephilidae Gen. Sp. (Trichoptera) is a too heterogeneous taxa to see some patterns, since its members colonize a wide range of habitats and have very different feeding adaptations. However, in this study they seems to be related to increasing particulate organic content.

Taeniopteryx nebulosa (LINNAEUS, 1758) (Plecoptera). This stonefly it is considered to live in slow flowing waters, preferring fine sediment with organic matter. Some studies showed an association also with slow flowing sandy reaches (0.3 m/s) and a strong relation with macrophytes and moss habitats (Langford and Bray, 1969). Here, this specie seems also related to flow velocity and the 0.3 m/s are compatible with the Pliszka mean flow (0.26 m/s), but also with organic matter. However macrophytes and moss were not present in the sampling sites.

Calopteryx sp. (Odonata) is a predator, normally thought to prefer moderate to fast flowing waters. Here, the relation with the velocity seems outweighed by the importance of organic matter, but since this invertebrate is a predator, this cannot be the main factor. Low

level of BOD (that some *Calopteryx* species can survive (Hofmann and Mason, 2005)) is more probable with high organic level and weak flow, so this can be the reason of this results.

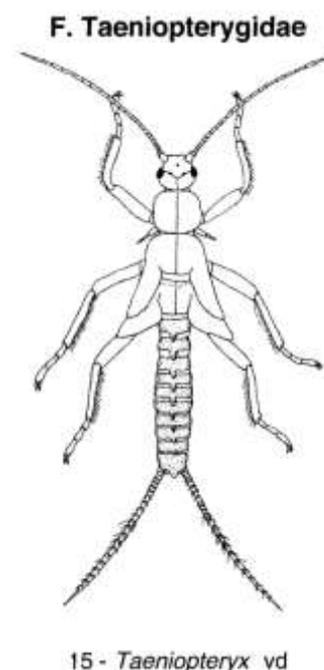
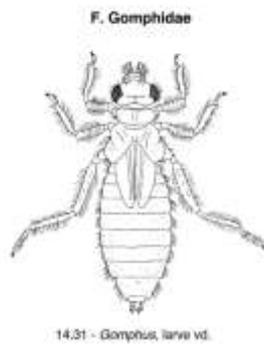


Figure 22 *Taeniopteryx* sp. larvae (Tachet *et al.*, impr. 2010)

- Negative relation with organic content and flow velocity



Gomphus sp. (Odonata) and *Micronecta sp.* (Heteroptera) are both genus that can be also found in still waters, and accordingly in this study were found negatively correlated with flow velocity.

Figure 23 *Gomphus sp.* larvae
(Tachet *et al.*, impr. 2010)

CONCLUSIONS

The results of this work show clearly the modifications of the benthic environment and population resulting by the presence of LWD on a sand river. These logs alter significantly the habitat diversity and invertebrate population of the sand – bottom rivers, creating longitudinal mesohabitats with higher diversity, evenness and taxa abundance, sometimes with values twice as high as the non – wood areas for every single habitat. The subdivision in habitat of the sediment surrounding a wood log perpendicular to the flow seems justified, since differences in both abiotic features and macroinvertebrates distribution can be found. The characteristics that define every single mesohabitat are on one hand a combination of Mean Flow Velocity, Depth and Sediment Granulometry, on the other hand the amount of Organic Matter available to the invertebrates. Distance from the wood, at least inside 160 cm of the wood, didn't results as significant variable, so it was not possible to identify the expected gradient in macroinvertebrate assemblages. Knowing distribution patterns and influences can be of great help in future sampling efforts. This study also highlighted a big difference between sand bars downstream logs and “normal” sand bottom of the river: these two habitats, the same at a first glance, are actually very different.

This study needs further development on the feeding guilds and particular species distribution, as well as different measurements of the environmental variables. Particularly, the application of better turbulence and shear stress measurements are desirable, to identify to a better scale differences in microhabitats created by the wood disturbance. It is also desirable a comprehensive analysis of the macroinvertebrates assemblages, quantitatively and qualitatively, to better understand where the wood influence on the substrate ends.

The hope is that this study of a pristine environment will help, in the future, to develop better evaluation systems for the efficiency of restoration techniques. Better knowledge of patterns could help to decide how to evaluate these effort, where to sample to understand the efficiency and to adjust restoration techniques.

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Annex 1 : List of taxa and species found

	Total
Bivalvia	617
SPHAERIIDAE	611
Pisidium sp.	593.63
Sphaerium sp.	17.37
UNIONIDAE	6
Anodonta anatina	3
Unio sp.	3
Coleoptera	413
[Ord:Coleoptera]	1
Coleoptera Gen. sp. Lv.	1
ELMIDAE	408
Elmis sp. Lv.	121
Limnius sp. Ad.	6
Limnius sp. Lv.	57
Macronychus quadrituberculatus Ad.	1
Macronychus quadrituberculatus Lv.	198.78
Oulimnius sp. Lv.	1
Riolus sp. Lv.	23.22
GYRINIDAE	4
Orectochilus sp. Lv.	4
Crustacea	650
ASELLIDAE	2
Asellidae Gen. sp.	2
GAMMARIDAE	648
Gammaridae Gen. sp.	648
Diptera	2728.99
ATHERICIDAE	232
Atherix ibis	121
Atrichops crassipes	94
Ibisia marginata	17
CERATOPOGONIDAE	75
Ceratopogonidae Gen. sp.	75
CHIRONOMIDAE	1457.99
Chironominae Gen. sp.	915.56
Orthocladiinae Gen. sp.	476.21
Tanypodinae Gen. sp.	66.22
EMPIDIDAE	276
Chelifera sp.	4
Hemerodromia sp.	272
LIMONIIDAE	28
Eloeophila sp.	26
Limoniidae Gen. sp.	2
MUSCIDAE	1
Muscidae Gen. sp.	1
PEDICIIDAE	2
Dicranota sp.	2
PSYCHODIDAE	4
Psychodidae Gen. sp.	4
PTYCHOPTERIDAE	2
Ptychoptera sp.	2
SIMULIIDAE	642
Simuliidae Gen. sp.	642
TABANIDAE	6
Tabanidae Gen. sp.	6
TIPULIDAE	3
Dolichopeza sp.	1
Tipula sp.	1
Tipulidae Gen. sp.	1
Ephemeroptera	416.01
[Ord:Ephemeroptera]	5
Ephemeroptera Gen. sp.	5
BAETIDAE	309.89
Baetidae Gen. sp.	2

	Total
Baetis sp.	307.89
CAENIDAE	1
Brachycercus harrisella	1
Caenidae Gen. sp.	
EPHEMERELLIDAE	1
Ephemerellidae Gen. sp.	1
EPHEMERIDAE	3
Ephemera sp.	3
HEPTAGENIIDAE	79.01
Heptagenia flava	2.29
Heptagenia longicauda	43.57
Heptagenia sulphurea	28.15
Heptageniidae Gen. sp.	5
LEPTOPHLEBIIDAE	17.11
Paraleptophlebia sp.	15.11
Paraleptophlebia submarginata	2
Gastropoda	109
BITHYNIIDAE	50
Bithynia tentaculata	50
ELLOBIIDAE	1
Carychium sp.	1
HYDROBIIDAE	30
Potamopyrgus antipodarum	30
PLANORBIDAE	28
Ancylus fluviatilis	26
Planorbidae Gen. sp.	2
Heteroptera	152
APHELOCHEIRIDAE	110
Aphelocheirus aestivalis	110
CORIXIDAE	42
Micronecta sp.	42
Hirudinea	58
[Kl:Hirudinea]	58
Hirudinea Gen. sp.	58
Hydrachnidia	459
[Ph:Hydrachnidia]	459
Hydracarina Gen. sp.	459
Lepidoptera	16
[Ord:Lepidoptera]	16
Lepidoptera Gen. sp.	16
Odonata	47
CALOPTERYGIDAE	7
Calopteryx sp.	7
GOMPHIDAE	40
Gomphidae Gen. sp.	2
Gomphus sp.	35
Ophiogomphus sp.	3
Oligochaeta	3121
[Kl:Oligochaeta]	3121
Oligochaeta Gen. sp.	3121
Planipennia	1
SISYRIDAE	1
Sisyra fuscata	1
Plecoptera	45
NEMOURIDAE	1
Nemoura avicularis	1
PERLODIDAE	5
Perloides dispar	5
TAENIOPTERYGIDAE	39
Taeniopteryx nebulosa	39
Trichoptera	572
[Ord:Trichoptera]	11
Trichoptera Gen. sp.	11
BRACHYCENTRIDAE	50
Brachycentrus montanus	25

Small-scale distribution of macroinvertebrates on Large Woody Debris and nearby sediment in a lowland river.

	Total
Brachycentrus subnubilus	25
HYDROPSYCHIDAE	80
Hydropsyche pellucidula	5
Hydropsyche siltalai	74
Hydropsychidae Gen. sp.	1
HYDROPTILIDAE	15
Hydroptilidae Gen. sp.	4
Ithytrichia lamellaris	11
LEPIDOSTOMATIDAE	334
Lepidostoma basale	334
LEPTOCERIDAE	1
Leptoceridae Gen. sp.	1
LIMNEPHILIDAE	52

	Total
Limnephilidae Gen. sp.	52
POLYCENTROPODIDAE	2
Plectrocnemia sp.	1
Polycentropus sp.	1
PSYCHOMYIIDAE	27
Lype sp.	19
Psychomyia pusilla	4
Psychomyiidae Gen. sp.	4
Turbellaria	53
DENDROCOELIDAE	1
Dendrocoelum lacteum	1
DUGESIIDAE	52
Dugesia sp.	52

The following table report all the feeding type percentages in every sampling point.

Annex 2: Feeding groups percentages

		graz/scrap	min	xylo	shredd	collect	active_filt	pass_filt	filters	pred	paras	other	No data
Sand	Max	19.121	6.842	7.143	20.37	73.877	59.231	11.607	63.077	20.536	6.842	5.926	0.552
	Min	2.394	0	0	0	11.154	3.736	0	4.444	1.108	0.517	0	0.027
CPOM - FPOM	Max	17.959	3.307	8.056	35.526	60.768	33.024	19.264	39.554	24.285	8.372	9.9	0.571
	Min	2.5	0	0	2.338	16.535	2.519	0	3.908	7.699	0.132	0.469	0.054
Gravel	Max	19.091	2.222	5.227	23.75	75.537	33.024	66.764	70.816	30	4.889	7.829	0.385
	Min	4.098	0	0	0.496	13.178	1.463	1.442	3.17	2.066	0.554	0.098	0.052
Log	Max	29.569	1.496	40.987	12.182	74.274	2.992	23.288	23.521	23.966	3.966	1.324	0.898
	Min	7.655	0.11	0	0	18.966	0.22	0.647	3.075	7.912	0.962	0	0.098
Control	Max	4.46	1.398	1.127	1.408	91.129	22.254	1.075	23.193	15.352	1.398	0.215	0.059
	Min	0.806	0.161	0	0	53.005	3.871	0	4.355	1.398	0.645	0	0.008

Glossary of column headings:

graz/scra	min	xylo	shredd	collect	active_filt	pass_filt	filters	pred	paras	other	no data
Grazers and Scrapers	Miners	Xylophagous	Shredders	Gatherer Collectors	Active filter feeders	Passive filter feeders	Total filter feeders	Predators	Parasites	Other	No Data

Annex 3 : Habitat Survey

Complete report for the visual habitat survey. Surveyed squares are numbered sequentially from the left bank to the right. Sum for Substrate 1 is always 100. Substrate 2 consisting in overhanging branches, macrophytes, CWD, etc. is often less than 100 (only part of the area is covered. Depth (in cm) is taken in the center of every grid square (see Figure 19).

To make the analysis clearer, In the Graph 5 in the Substrate 1 data, CWD has been united with Log and CPOM with FPOM.

Large Woody Debris 1

LWD1 -> 2m Downstream

POINTS	DEPTH	SUBSTRATE 1 (100%)						SUBSTRATE 2			
		Sand	FPOM	Gravel	Log	CPOM	CWD	CPOM	CWD	LOG	Macroph.
1	118				100						
2	115			100						50	
3	110			100							
4	112			100							
5	112	60		40				15			
6	100	80		20							
7	85	100						40			
8	63	100						50			
9	58		100					60			
10	52		100					60			

LWD1 -> 1m Downstream

POINTS	DEPTH	SUBSTRATE 1 (100%)						SUBSTRATE 2			
		Sand	FPOM	Gravel	Log	CPOM	CWD	CPOM	CWD	LOG	Macroph.
1	105				100						
2	110			100						50	
3	108			100						10	
4	116			80		20				10	
5	120			50		50				10	
6	91	70		15	15						
7	63	50			50						
8	60	100				20					
9	62					100					
10	57		100					70			

LWD1 -> 1m Upstream

POINTS	DEPTH	SUBSTRATE 1 (100%)						SUBTRATE 2			
		Sand	FPOM	Gravel	Log	CPOM	CWD	CPOM	CWD	LOG	Macroph.
1	108		50	50						25	
2	108			100						50	
3	110			100						10	
4	120	10		90						10	
5	118	90		10				15			
6	105	100							10	10	
7	90	100						30			
8	80		30			70		30			
9	65					100				10	
10	60					100				10	

LWD1 -> 2m Upstream

POINTS	DEPTH	SUBSTRATE 1 (100%)						SUBTRATE 2			
		Sand	FPOM	Gravel	Log	CPOM	CWD	CPOM	CWD	LOG	Macroph.
1	110			100							
2	110			100					10	10	
3	120	50		50					10		
4	112	80		10	10				20		
5	104	100							15		
6	95	100						15			
7	64	100						10			
8	80					100					
9	85					100				20	
10	39		100					50		50	

Large Woody Debris 2

LWD2 -> 2m Downstream

POINTS	DEPTH	SUBSTRATE 1 (100%)						SUBTRATE 2			
		Sand	FPOM	Gravel	Log	CPOM	CWD	CPOM	CWD	LOG	Macroph.
1	140			100							
2	125			100					25		
3	100	100						20	20		
4	50	50			50				50		
5	50	100						10			
6	30	50	50						30		
7	24		100								10
8	20		100								50

LWD2 -> 1m Downstream

POINTS	DEPTH	SUBSTRATE 1 (100%)						SUBTRATE 2			
		Sand	FPOM	Gravel	Log	CPOM	CWD	CPOM	CWD	LOG	Macroph.
1	140		50	50						10	
2	130		70	30						50	
3	130	50				50				15	
4	100	10			20	70				15	
5	50	25			25	50				10	
6	25	50	40		10				15		
7	25		100							10	15
8	20		100								25

Small-scale distribution of macroinvertebrates on Large Woody Debris and nearby sediment in a lowland river.

LWD2 -> 1m Upstream

POINTS	DEPTH	SUBSTRATE 1 (100%)						SUBTRATE 2			
		Sand	FPOM	Gravel	Log	CPOM	CWD	CPOM	CWD	LOG	Macroph.
1	140			100						15	
2	125	10		90						15	
3	118	100						50		10	
4	95	90			10			50			
5	62	50			20	30		25			
6	50	80			10	10		50			5
7	27	60	30		10			25	10		
8	20		25			75		10			20

LWD2 -> 2m Upstream

POINTS	DEPTH	SUBSTRATE 1 (100%)						SUBTRATE 2			
		Sand	FPOM	Gravel	Log	CPOM	CWD	CPOM	CWD	LOG	Macroph.
1	128			100					25		
2	128			100							
3	112	70		20	10						
4	95	100						50			
5	50	100						10			5
6	30	100						20			5
7	25	100						20			5
8	15		50			50					40

Large Woody Debris 3

LWD3 -> 2m Downstream

POINTS	DEPTH	SUBSTRATE 1 (100%)						SUBTRATE 2			
		Sand	FPOM	Gravel	Log	CPOM	CWD	CPOM	CWD	LOG	Macroph.
1	66		100					25			
2	108	75		15	10						
3	100	100									
4	85	100									
5	88	100									15
6	86	50		50					10		
7	55	100							15		
8	42	100							10		15
9	30	50	25			25			15		25

LWD3 -> 1m Downstream

POINTS	DEPTH	SUBSTRATE 1 (100%)						SUBTRATE 2			
		Sand	FPOM	Gravel	Log	CPOM	CWD	CPOM	CWD	LOG	Macroph.
1	95	20	50			20	10		25		
2	94	90					10		10		
3	92	90			10			10			
4	92	100						10		10	
5	102	100								10	
6	100	75		25						10	
7	72	100							10	10	
8	46	75			25			25		10	
9	25	20	20		10	50					25

LWD3 -> 1m Upstream

POINTS	DEPTH	SUBSTRATE 1 (100%)						SUBTRATE 2			
		Sand	FPOM	Gravel	Log	CPOM	CWD	CPOM	CWD	LOG	Macroph.
1	50		50		50			25			
2	90	70	10		10	10		25			
3	80	90			10			10			
4	82	90			10						
5	85	100								10	
6	102	100						10		10	
7	91	100								10	
8	62	75	25							25	
9	20	25	50		10	15					10

LWD3 -> 2m Upstream

POINTS	DEPTH	SUBSTRATE 1 (100%)						SUBTRATE 2			
		Sand	FPOM	Gravel	Log	CPOM	CWD	CPOM	CWD	LOG	Macroph.
1	30		50		50						
2	75	75			25			25			10
3	85	100						10			10
4	78	100						10			10
5	70	100									
6	90	100									
7	110	75		25				25			
8	63	100							10		
9	30	50	50						10		10

Annex 4: Autoecological Bibliography

Sources of the autoecological information found on (Schmidt-Kloiber and Hering, 2013).

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Annex 5 : Identification Bibliography

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