The effects of increased fine sediment load on the benthic invertebrate community in five upper Austrian streams.

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ABSTRACT

The reasons for siltation and its impact on the benthic invertebrate fauna in running waters have been studied in various previous researches. Nevertheless benthic invertebrate indicator species that signal distinct stages of siltation and the potential applicability of the newly introduced Proportion of Sediment sensitive Invertebrate index (PSI) by Extence et al. (2011) could provide a tool for cost-efficient monitoring in areas prone to the effects of siltation. The aim of this study is therefore to evaluate the effects of siltation on benthic invertebrates in the study area and to compare them with similar studies, and to identify taxa preferring silted conditions that can be potentially used as indicator species for increased sediment load and deposition in running waters and furthermore to assess whether the PSI is applicable in the study area. Therefore, 92 benthic invertebrate single sampling units from eleven sites of five rivers (Osterbach, Gusen, Rodl, Kleine Mühl and Große Mühl) in the north-eastern part of upper Austria have been taken in the period of 14^{th} to 16^{th} of April 2014. The number of taxa, biomass, % EPT and PSI didn't react until a certain threshold when they decreased alongside a gradient of increasing fine sediment. The analysis of indicator species showed exactly an opposite pattern than expected; only indicator species for near natural, unsilted conditions were found, while with increasing fines the taxa shift from specialists to generalists. The slow reaction of the different metrics underline the importance of small scale structures, like woody debris, within the river bed that provide habitat for species that otherwise would not occur in silted conditions. The PSI shows a weak reaction to increasing fine sediment share. The reason is that the finest fractions in the study area are consisting of granite grit, which is typical for the study area and comprises psammal to akal fractions whereas the PSI ratings consider effects of agryllal to psammal fractions. Furthermore the sensitivity ratings by Extence et al. (2011) could be improved to a lower taxonomic level and extended to Chironomidae to possibly mitigate the weak reaction of the PSI in the study area. This study shows that sedimentation of fine sediments have distinct effects on the lotic environment.

Keywords: *siltation, benthic invertebrates, proportion of sediment sensitive invertebrates (PSI).*

KURZFASSUNG

Die Ursachen von Versandung und ihr Einfluss auf das Makrozoobenthos wurden bereits in früheren Studien gründlich untersucht. Ergänzend dazu können benthische Invertebraten, welche als Zeigerarten für verschiedene Stufen der Versandung im Fließgewässer fungieren, als auch der potentielle Einsatz des Proportion of Sediment Sensitive Invertebrate Index (PSI) von Extence et al. (2011) die Grundlage für ein kosteneffizientes Monitoring Tool in Gegenden mit potentiellen Versandungsproblemen darstellen. Das Ziel dieser Masterarbeit ist es, den Einfluss von Versandung auf die Makrozoobenthos Gemeinschaft in den fünf Flüssen des Studiengebietes festzustellen, die beobachteten Effekte mit denen aus ähnlichen Studien zu vergleichen, herauszufinden, ob es spezifische Zeigerarten für zunehmende Versandung im Fließgewässer beziehungsweise versandete Bedingungen bevorzugende Arten gibt, als auch zu prüfen, ob sich der PSI im Forschungsgebiet anwenden lässt. Hierzu wurden 92 Makrozoobenthos Einzelproben und zwölf MHS Proben an elf Standorten an fünf Flüssen (Osterbach, Gusen, Rodl, Kleine Mühl und Große Mühl) vom 14. bis 16. April 2014 im nord-östlichen Oberösterreich genommen. Die Anzahl an verschiedenen Taxa, die Biomasse, der Anteil an EPT- Taxa und der PSI reagierten - bis zu einem bestimmten Schwellenwert - nicht, obwohl ein genereller Trend der Abnahme dieser Werte bei steigendem Feinsedimentanteil zu beobachten war. Die Berechnung der Zeigerarten ergab exakt das gegenteilige Ergebnis zur Erwartungshaltung, es wurden nur Zeigerarten für naturnahe, unversandete Bedingungen gefunden und ein genereller Wechsel von Spezialisten zu Generalisten bei steigender Versandung beobachtet. Der PSI zeigte nur eine schwache Reaktion auf den steigenden Feinsedimentanteil. Der Grund für diese schwache Reaktion ist der als feinste Sedimentfraktion dominierende Granitgrus, welche typisch für die Studiengegend ist. Granitgrus umfasst Psammal bis Akal Fraktionen und bildet daher die Obergrenze der Sedimentfraktionen bezüglich des Begriffes Feinsediment. Ein weiterer Grund für die schwache Reaktion ist die Tatsache, dass sich der PSI durch eine Verfeinerung der Sensitivitätseinstufungen von Extence et al. (2011) durch die Verwendung eines niedrigeren taxonomischen Levels als auch durch die Berücksichtigung von Chironomiden verbessern lassen würde.

Schlagwörter: *Versandung, Makrozoobenthos, Proportion of sediment sensitive Invertebrates (PSI).*

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1. INTRODUCTION

It has long been noted that fine sediment can have a detrimental effect on aquatic invertebrate communities. Such observations have led to efforts to describe and quantify the impact of fine sediments on invertebrates. Largely, these investigations have been driven by an interest in the effects of fine sediment per se (Jones et al., 2007).

Frequently the terms ‘’fines’’ and ‘’sedimentation’’ are used in their broadest sense by many freshwater scientists. For examples, Pinder et al. (1987) refer to ‘’soft sediments’’ to characterize the entire range of fine particles in riverine deposits. Wright et al. (1983) adopt an even broader definition, which encompasses sand and silt (as determined by physical size) as well as fine and coarse organic material such as leaves (Wood & Armitage, 1997).

Geologically, silt is defined as a grain of a size between 0.004 and 0.063 mm, when using the Udden-Wentworth size classification. However, concern over fine sediment can include both sand fractions and clays, an overview on the size classes is given in table 2. On the other hand, siltation can be defined as the deposition of fine sediment either on the surface of the stream bed or within a gravel substrate. Riverbeds are not static. Sediment movement is a natural process, although it may occur only rarely during extreme floods, depending on the mobility of the channel (Scarlett & Hornby, 2000).

In their 2005 publication Owens et al. summarized the present effects of fine sediments on a global scale: Fine-grained sediment is a natural and essential component of river systems and plays a major role in the hydrological, geo- morphological and ecological functioning of rivers. In many areas of the world, the level of anthropogenic activity is such that fine-grained sediment fluxes have been, or are being, modified at a magnitude and rate that cause profound, and sometimes irreversible, changes in the way that river systems function. Furthermore they state that, although human activity has generally resulted in increased fine-grained sediment delivery to rivers and increased sediment transport in rivers, it must be recognized that this is not always translated to increased sediment yields in downstream reaches. In some instances, downstream sediment yields have remained broadly constant, for example due to changes in sediment storage, or have decreased over time due to the construction of impoundments, dams and reservoirs (Trimble, 1983; Vörösmarty et al., 2003; Walling and Fang, 2003). Vörösmarty et al., (2003) for example, estimate that reservoirs trap about 25–30% of the global sediment flux to the oceans. Owens et al. (2005) synthesized the existing information from Salomons (2004, 2005) in table 1 to give an overview on the present drivers, impacts and states regarding siltation worldwide.

Table 1: Drivers, impacts and states regarding worldwide siltation by Owens et al. (2005) derived from Salomons (2004, 2005).

Region	Ranking	Anthropogenic drivers	Major state changes and coastal impacts	Present pressure status	Trend expected
South America	1	Urbanization	Eutrophication	Major	Increasing
	2	Damming/diversion	Erosion/sedimentation	Major	Increasing
	3	Industrialization	Pollution	Medium	Increasing
	4	Agriculture	Eutrophication/pollution	Medium	Increasing
	5	Deforestation	Erosion/sedimentation	Medium	Increasing
Africa	1	Damming/diversion	Erosion/sedimentation	Major	Increasing
			Salinization	Local	
			Nutrient depletion	Local	
	2	Various drivers	Biodiversity loss	Major	Increasing
	3	Deforestation	Erosion/sedimentation	Medium	Increasing
	4	Agriculture	Eutrophication/pollution	Medium	Increasing
	5	Urbanization	Eutrophication/pollution	Medium	Increasing
East Asia	1	Urbanization	Eutrophication/water abstraction	Major	Increasing
	2	Agriculture	Eutrophication/pollution/ reclamation/disease	Major	Increasing
	3	Damming/diversion	Erosion/sedimentation/nutrient depletion	Major	Increasing
	4	Industrialization	Pollution	Minor	Increasing
	5	Deforestation	Erosion/sedimentation	Major	Increasing
Russian Arctic	1	Industrialization	Pollution	Major	Increasing
	2	Navigation	Erosion/sedimentation	Medium	Increasing
	3	Damming	Erosion	Medium	Decreasing
	4	Agriculture	Nutrient/water/sediment	Minor	Stable
	5	Urbanization	Nutrient/water abstraction	Minor	Stable

On a European scale Vanmaercke et al. (2011) gathered data from 1794 locations through an extensive literature review. Despite potential uncertainties, due to the heterogeneous dataset they used, important regional differences in sediment yield can be noted. Whereas most catchments in northern, western and central Europe are characterised by relatively low sediment yield values, Mediterranean and mountainous regions generally have higher sediment yield values. As these differences are based on large numbers of observation, they cannot only be attributed to uncertainties on the available data but must be the result of regional variation of the controlling factors of fine sediment input and potentially of different dominant erosion processes (Vanmaercke et al., 2011). The importance of local factors on fine sediment input can be expected to be smaller for large river systems (>10,000 km²). Larger catchments generally consist of a patchwork of land uses and geomorphic units. Local conditions are therefore less likely to override general trends, as they often average out at larger scales. Furthermore, mean slope gradient generally decreases with increasing catchment size and the importance of sediment sinks can be expected to further increase (De Vente & Poesen, 2005).

Figure 1 (Wood & Armitage, 1997) gives a holistic overview on the inherent processes and effects of an increased fine sediment amount in running water systems showing the sources and processes that bring fine sediments in the system and the effect increased fine sediment load has on the aquatic fauna.

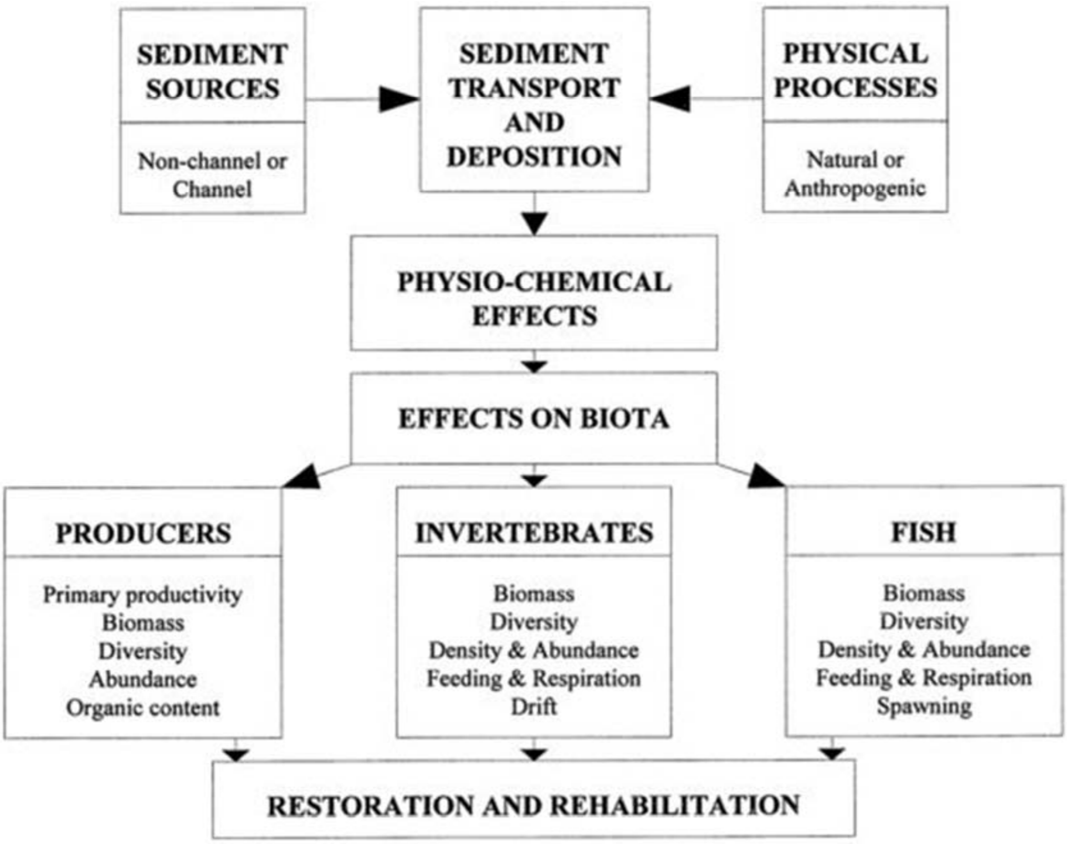

Figure 1: A holistic overview of fine sediment in the lotic ecosystem after Wood & Armitage (1997).

Generally two main sources of sediment input can be identified; channel sources, which are principally derived from the bed and banks of the stream and its tributaries, and non-channel sources within the catchment, such as bare soils that are susceptible to erosion (Grimshaw & Lewin, 1980). The principle sources of fine particles available to a stream from channel sources are river banks subject to erosion due to high shear, long exposure to water, and location (e.g., on a meander bend); mid-channel and point bars subject to erosion; fine bed material stored within the interstices or from surficial deposits; natural backwaters where sediment may accumulate during base flow conditions; fine particles trapped within aquatic macrophyte stands or associated with the seasonal growth and decline of aquatic vegetation; and other biotic particles including phytoplankton and zooplankton. In some instances, there may be some on-site generation of fine particles due to the decay of aquatic macrophytes, biofilms and invertebrate material. Benthic invertebrate faecal material has been shown to constitute a significant source of fine particulate matter. The main non-channel sources of fine sediment supplied to a stream are exposed soils subject to erosion—this material is transported to the channel via gullies, rills, and other features associated with runoff erosion; mass failures within the catchment, such as landslides and soil creep; urban areas, which markedly increase sediment delivery by increasing both the volume and timing of runoff; anthropogenic activities; litter fall, principally leaf material from vegetation adjacent to the channel; and atmospheric deposition, due to aeolian processes and precipitation (Wood & Armitage, 1997).

The erosion and deposition of fine sediment are intrinsic and natural components of the hydro-geomorphic processes of fluvial systems (Jones et al., 2007). Two types of fine sediment transport can be identified; along the surface of the substrate as bedload by rolling, sliding, or saltating and as turbulence increases, the weight of the particle may be upheld as suspended load by a succession of eddy currents (Petts & Foster, 1985).

Wood & Armitage (1997) summarized the effects of fine sediments on benthic invertebrates. Fine sediment suspension and deposition affects benthic invertebrates in four ways: (1) by altering substrate composition and changing the suitability of the substrate for some taxa (Erman & Ligon, 1988; Richards & Bacon, 1994); (2) by increasing drift due to sediment deposition or substrate instability (Culp et al., 1985; Rosenberg & Wiens 1978); (3) by affecting respiration due to the deposition of silt on respiration structures (Lemly 1982) or low oxygen concentrations associated with silt deposits (Eriksen 1966); and (4) by affecting feeding activities by impeding filter feeding due to an increase in suspended sediment concentrations (Aldridge et al., 1987), reducing the food value of periphyton (Cline et al., 1982; Graham 1990) and reducing the density of prey items (Peckarsky, 1984).

Figure 2 by Jones et al. (2007) illustrates the direct and indirect mechanics described above and shows the main physical effects of increased fine sediment load on benthic invertebrates.

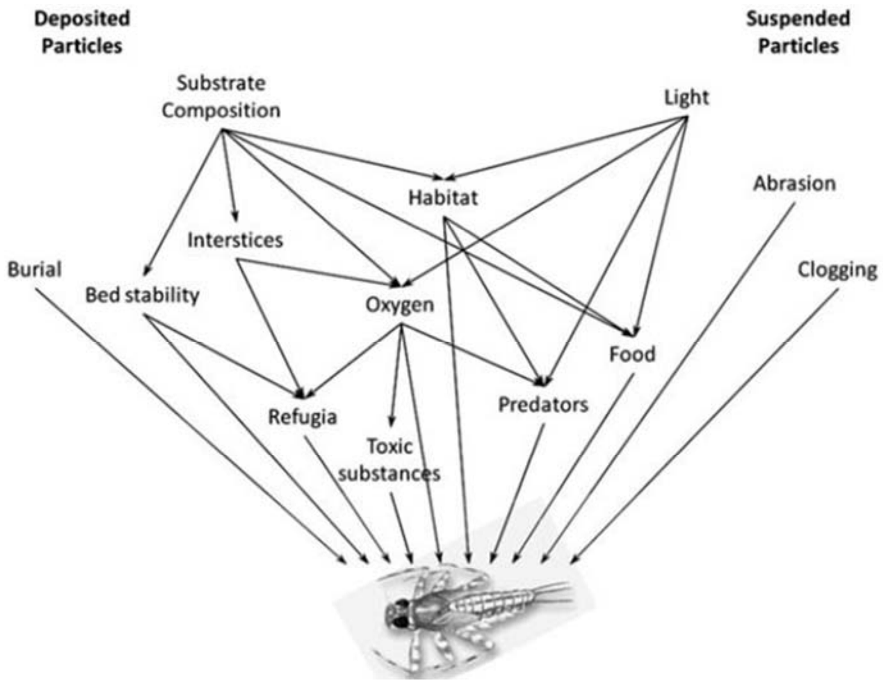

Figure 2: Summary of fine sediment caused mechanisms affecting benthic invertebrates. The strength and direction (+/−) of effects are not given as they are dependent upon the taxa affected; some taxa and communities respond positively to changes, others negatively (Jones et al., 2007).

These physical effects are clogging as well as physical damage by abrasion, which can result in a build-up on the organs of benthic invertebrates, disrupting the normal functioning of gills and filter-feeding apparatus, making respiration and feeding difficult. Burial can present difficulties for sedentary animals and, where rates of deposition are high, even motile animals can be affected. Yet, many species of invertebrates are adapted to live in fluvial depositional zones and benefit from the rate of influx of organic (food) particles (Jones et al., 2007). Issues arise when the rate of accretion exceeds the ability of individuals to excavate themselves, which is highly dependent upon individual taxa and the particle size of deposited materials (Wood et al., 2005). Together with increased accretion, fine sediment deposition results in changes to the composition of the bed of rivers. Where inputs of fine sediment to catchments are increased, the average size of particles becomes smaller, interstices between larger particles become filled and, where a surface drape of deposited sediment occurs, the stability of the bed may be reduced (Kaufmann et al., 2009).

Most invertebrate species have specific requirements of the substrate they live in and tend to avoid patches that fail to meet these requirements (Jones et al., 2007).

The deposition of fine sediments on river beds is usually associated with profound changes to the chemical environment. Reduced percolation of water through the substrate results in the establishment of more pronounced gradients of oxygen and other dissolved substances (Pretty et al., 2006). Jones et al. (2007) add that where deposited material has a high organic content, microbial activity can lead to oxygen depletion and a build-up of potentially toxic substances such as ammonium, ferrous and manganous ions. However, the increased supply of particulate organic matter may benefit some particular species of invertebrates (Welton & Clarke 1980; Lemly, 1982; Arruda et al., 1983; Hart 1992; Jackson et al., 2007).

Furthermore, Jones et al. (2007) identified the indirect effects of increased fine sediment load on benthic invertebrates as decreasing habitat availability, reduced food availability and quality as well as changes in the food web. There is a strong relationship between substrate composition and invertebrate distribution at the patch scale (Culp et al., 1983), and invertebrate community is strongly correlated with mesoscale habitat patches (Pardo & Armitage 1997; Collier et al., 1998; Armitage & Cannan, 2000; Buffagni et al., 2000) defined by water depth, substrate composition and the presence and type of macrophytes (Kemp et al., 2000). Moreover, changes in habitat patches are often the best indicators of change in an invertebrate community (Petts et al., 1993). Dependent upon the key sources of fine sediment, loadings to rivers can contain substantial quantities of organic matter, influencing the concentration of particulate organic matter suspended in the water and therefore, the food available to filter-feeding invertebrates. Where concentrations of particulate organic matter are increased above intrinsic levels, there is a potential for, filter feeders to expand. However, the overall quality of particulate food resources may decline if the concentration of particulate inorganic matter increases disproportionally, influencing ingestion rate (Gaugler & Molloy, 1980). As invertebrates comprise the main food resource of many species of fish, invertebrates are released from predation where fish populations decline as a consequence of increased fine sediment loading. Even where fish populations are not **impacted**, increased turbidity can reduce the visibility of invertebrate prey to fish, thus reducing predation risk (Gardener, 1981; Berg & Northcote 1985; Zamor & Grossman 2007). The implications for the invertebrate community will depend on the extent to which predation controls population growth; in unproductive or frequently disturbed conditions, change in fish density has little impact on invertebrates (Peckarsky, 1991), whereas fish can exert significant control on invertebrate populations in more productive environments

(Jones & Jeppesen, 2007). Rabení et al. (2005) investigated functional response of benthic invertebrates to increasing surface cover by fine sediment in four streams in the United States. Rabení et al. (2005) investigated the relation of surface cover and functional feeding guilds (FFG) as well as functional habitat groups (FHG). He states that with increasing surface cover of fine sediments a general decrease in the density of individuals is to be expected but some functional feeding and habit groups might be favoured, due to physical adaptions and higher resilience when affected by high amounts of fine sediment.

The percentage of Ephemeroptera, Plecoptera and Trichoptera (EPT) is usually negatively correlated with the increase in fine sediment (Descloux et al., 2013) while Baetidae, Oligochaeta, Chironomidae and Elmidae show an opposite pattern in an experimental study of Angradi (1999). Furthermore according to Rabení et al. (2005) and Matoni & Kep (2010), expressed in metrics, % Limnephilidae to Trichoptera, % Leptoceridae to Trichoptera, % Baetidae to Ephemeroptera are expected to increase at the silted sites. Whereas number of total taxa, number of EPT taxa, % of filter feeders, % shredders and % scrapers are expected to decrease with increasing amount of fine sediments (Matoni & Kep, 2010).

Extence et al. (2011) developed a sediment-sensitive macro-invertebrate metric (PSI — Proportion of Sediment-sensitive Invertebrates) which provides a proxy to describe the extent to which the surface of river beds are composed of, or covered by, fine sediments. The PSI technique is a relatively simple approach based on the proportion of sediment-sensitive taxa recorded in a benthic macro-invertebrate sample. Where suitable biomonitoring data exists, the index can be calculated retrospectively to track trends in fine sediment deposition, and its ecological impact, through time. Furthermore Extence et al. (2013) state that the PSI is an effective methodology, with sufficient capability to first identify and then help managing sedimentation impacts. The biological basis of the metric is in keeping with the philosophy of the WFD, and the ability of the macro-invertebrate community to integrate impacts over time is an advantage compared with the alternative of direct monitoring of siltation, which is time-consuming and relatively expensive.

This thesis contributes to the "Interreg Project Bayern-Österreich – "Feststoffmanagement im Mühlviertel und im Bayerischen Wald". The project is done on behalf of the Bavarian state agency for environment, the federal state of Upper Austria and the water management office Deggendorf, funded by the European Union and the Austrian Federal Ministry of

Agriculture, Forestry, Environment and Water Management. Priv.-Doz. Dipl.-Ing. Dr. Christoph Hauer and Univ.-Prof. Dipl.-Ing. Dr. Helmut Habersack coordinate and lead the project. The aim of the project is to address the specific problems that arise due to entry of, typical for the crystalline bohemian mass, granite grit into the running waters of the study area. The increased amount of granite grit leads to flood risk issues due to a decrease of the cross section and increasing colmation and ecological quality issues that arise due to increasing habitat homogeneity and degradation of the river bed. The project consists of eleven working packages dealing with assessment of existing data, definitions, creation of maps, land use- and grain size analyses, mineralogical analyses, installation of sediment traps, assessment of the aquatic habitat quality, principles of sediment management, flood risk management, physical experiments, model creation and impacts on the freshwater pearl mussel *Margaritifera margaritifera.* This thesis is part of the working package "aquatic habitat quality".

1.1. Aims and goals

- To evaluate if the effects of siltation on benthic invertebrates in the study area and to compare them with similar studies, and

- to identify taxa preferring silted conditions that can be potentially used as indicator species for increased sediment load and deposition in running waters.

- To assess whether the Proportion of Sediment-sensitive Invertebrate Index (PSI), by Extence et al. (2013) is applicable in the study area.

- To identify certain thresholds in the grade of siltation and its ecological effect.

1.2. Hypotheses

1. Increasing amounts of fine sediment lead to a decrease of species diversity, abundance and biomass in the benthic invertebrate community.

2. Specific benthic invertebrate taxa prefer silted conditions and can be used as indicator species for siltation processes.

3. Increased sediment load in running waters causes a decrease of the ecological quality class.

4. The PSI is applicable in the study area.

2. Study Area

Eleven sampling sites at five rivers (Osterbach, Gusen, Rodl, Kleine Mühl and Große Mühl) in the north-eastern part of upper Austria have been sampled from 14th to 16th of April 2014. All sites are located in the Ecoregion Central Highlands and in the Bioregion (Österreichisches) Granit- und Gneisgebiet der Böhmischen Masse (Illies, 1978). The sites have been selected by the Interreg Project Bayern-Österreich – "Feststoffmanagement im Mühlviertel und im Bayerischen Wald" and are mapped in Figure 3.

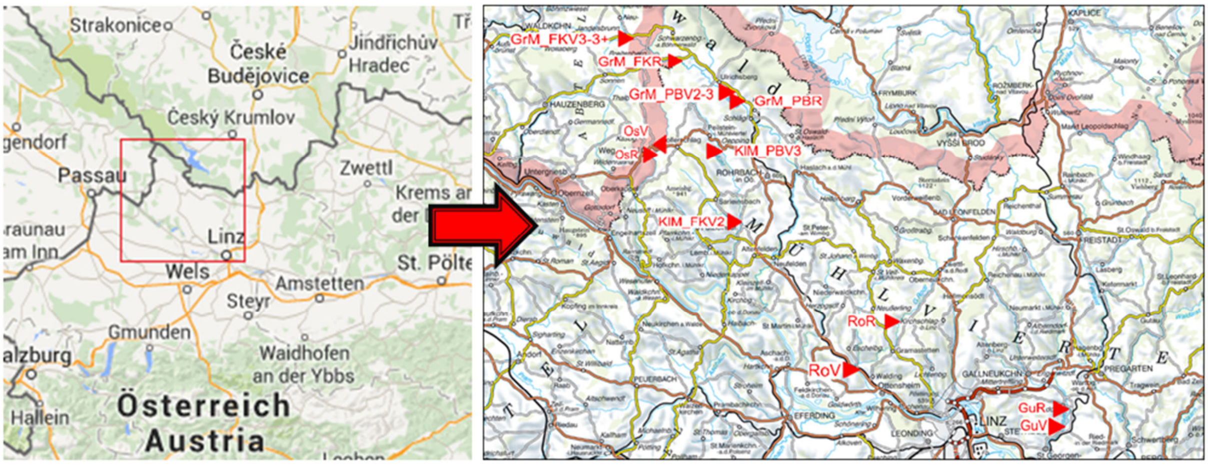

Figure 3: Overview and locations of the 11 sampling spots (left: maps.google.com, right: www.austrianmap.at).

2.1. OSTERBACH

The Osterbach has a stream order four and is located between at the German-Austrian border between the villages Kollerschlag in Austria and Wegscheid in Germany at approximately 600 meters above sea level. Benthic invertebrate samples at two sites of different degrees of siltation have been taken there. The **reference** site with a siltation degree of zero (OSR) is located at 48°36´02´´ N/ 13°48´31´´E and the **impacted** site with a siltation degree of 3 (OSV) is located at 48°36´31´´ N / 13°48´38´´ E. Figure 4 shows that the two sites are embedded in an area of grassland with some trees and shrubs at the shorelines. The dominating substrate at the **reference** site is mesolithal with very little fine sediments whereas the **impacted** site is dominated by akal fractions.

Figure 4: left: Reference site at the Osterbach (OSR), right: impacted site at the Osterbach (OSV).

2.2. RODL

The Rodl has a stream order four and is located in central upper Austria. Benthic invertebrate samples at two different sites of different siltation have been taken near the village of Edt. The **reference** site (ROR) downstream of Untergeng, 507 meters above sea level is located at 48°24´37´´ N/ 14°12´11´´E and the **impacted** site with a siltation degree of two (ROV) near Rottenegg, 260 meters above sea level is located at 48°21´39´´ / 14°08´10´´. Figure 5 shows that the **impacted** site ROV on the right has stronger alterations by human activities than the **reference** site. Rip-rap and other regulation measures are non-existent at the **reference** site. The **reference** site is situated in between forest and grassland and the substrate is dominated by meso- and macrolithal. The **impacted** site is bordered by a road and a small grass stripe next to a settlement and in the middle of the river the substrate consists mostly of mesolithal. However, due to low flow velocities at the riparian zones, fine sediments increase at the shoreline.

Figure 5: left: Reference site ROR, right: Impacted site ROV.

2.3. KLEINE MÜHL

The Kleine Mühl has a stream order five and is located in north-eastern upper Austria. Benthic invertebrate samples at two sites with different degrees of siltation and different bed forms have been taken. The KLM_FKV2 site has a Riffle-Pool bed form and siltation degree of 2. It is located near the town of Hühnergeschrei, 444 meters above sea level at 48°31´23´´ N / 13°56´48´´ E. The KLM_PBV3 has Plain-Bed bed form and a siltation degree of 3. It is located downstream of the town of Peilstein, 493 meters above sea level at 48°36´04´´ N / 13°54´47´´ E. Figure 6 shows the differences between the two sites. It is visible that the high siltation class of KLM_PBV3 could be a consequence of the lacking shoreline vegetation in the middle of grassland, the sediment is very soft and beneath the, pelal to akal dominated, fine sediment there is soil. On the other hand, KLM_FKV2 has better developed shoreline vegetation, therefore, a lot of woody debris and organic matter has been found at this site. The most dominant substrate fractions are akal and mesolithal but all other substrate fractions have been found too.

Figure 6: left: Riffle-Pool site KLM_FKV2, right: Plain-Bed site KLM_PBV3.

2.4. Große Mühl

The Große Mühl has a stream order five and is located in north-eastern upper Austria. Benthic invertebrate samples have been taken at four sites of different degrees of siltation and different bed forms, two of them being **reference** sites. The GRM_PBR site has a Plane-Bed bed form and a siltation degree of zero. It is located upstream of Aigen im Mühlkreis, 550 meters above sea level at 48°39´18´´ N/ 13°56´36´´ E. The GRM_PBV2-3 site has a Plane-Bed bed form and a siltation degree of 2.5. The site is located downstream of Ulrichsberg, just a few kilometres upstream of GRM_PBR. It has an elevation of 560 meters above sea level and is located at 48°42´01´´ N/ 13°55´30´´ E. The GRM_FKR site has a Riffle-Pool bed form and siltation degree of zero. It is located near the town of Vorderanger, 590 meters above sea level at 48°42´01´´ N/ 13°49´41´´ E. GRM_FKV3-3+ has a Riffle-Pool bed form and a siltation degree of 3,5. GRM_FKV3-3+ is located near the German town of Breitenberg next to the German-Austrian border at 695 meters above sea level, at 48°43´32´´ N/ 13°45´38´´ E. Figure 7 gives a visual comparison between the Plain-Bed **reference** and **impacted** site, both of which are embedded in grassland with little woody vegetation at shorelines. GRM_PBR is dominated by macro- and mesolithal, whereas at GRM_PBV2-3 a lot of macro- and mesolithal also occur. However, all of these stones are embedded in very fine fractions. Figure 8 shows the two Riffle-Pool sites at the Große Mühl. Both are surrounded by grassland with some woody vegetation at the shorelines. GRM_FKR has a high amount of mesolithal alongside macro- and microlithal with very little fine sediments whereas the substrate at GRM-FKV3-3+ is dominated by akal to psammopelal fractions.

Figure 7: left: Plain-Bed reference site GRM_PBR, right: Plane Bed impacted site GRM_PBV2-3.

Figure 8: left: Riffle-Pool reference site GRM_FKR, right: Riffle-Pool impacted site GRM_FKV3-3+.

2.5. GUSEN

The Gusen has a stream order five and is located in central upper Austria. One site with a siltation degree of 4 has been sampled for benthic invertebrates. The GUV site is located near Schörgendorf, 272 meters above sea level at 48°17´34´´ N / 14°28´23´´ E. Figure 9 shows the very slow flow velocity at the GUV site and that the shoreline vegetation is wide and dense as it is at the other sites. GUV is the site with the highest degree of siltation and very fine sediment fractions of akal and finer.

Figure 9: Two views of the heavily silted GUV site.

3. Methods

3.1. Sampling Design

The eleven study-sites each were classified into different siltation classes based on the definitions of the Interreg Project Bayern-Österreich – "Feststoffmanagement im Mühlviertel und im Bayerischen Wald" (Hauer et al., 2015).

- The **reference** class represents river stretches that have a natural substrate composition, without any unnatural fine substrate/granite grit depositions.

- **Siltation class 1** represents river stretches that are nearly natural, with at the maximum, little occurrences of granite grit with zero impacts on the heterogeneity and morphology of the river.

- In **siltation class 2** a considerable amount of fine sediments is present. Important habitats like riffles are negatively affected due to filling of the deeper parts.

- In **siltation class 3** the entire river bottom is covered with granite grit and no other substrate types are visible.

- In **siltation class 3+** the entire river bottom is covered with granite grit and no other substrate types are visible and the material is highly manoeuvrable. Even at low flow conditions sediment is transported.

At each of the rivers (except the Gusen, where only samples from the **impacted** site were taken) there was at least one **reference** site (siltation class 0) and at least one **impacted** site sampled. In addition to the siltation classes two different river types were sampled, namely plain-bed dominated stretches (Große Mühl) and riffle-pool characterised reaches (Kleine Mühl). At the **reference** sites mostly macro-, meso- and microlithal were predominant while microlithal, akal and psammal dominated the **impacted** sites, table 2 gives an overview on the different choriotope types in running waters. At each site four compartments consisting of five pooled sampling units each were taken per substrate type. Additionally, samples from macrophytes and xylal were collected. Table 3 gives an overview of the benthic invertebrate samples. In total 92 benthic invertebrate single sampling units, plus one multiple habitat sampling (MHS) sample per site, from eleven sites of five rivers (Osterbach, Gusen, Rodl, Kleine Mühl and Große Mühl) in the north-eastern part of upper Austria were taken from 14th to 16th of April 2014.

Table 2: Choriotope types in running waters after Moog et al. (1999).

Mineral substrates	Definition
Hygropetric sites	water layer on mineral substrata
Megalithal (>40 cm)	upper sides of large cobbles, boulders and blocks, bedrock
Macrolithal (>20 cm to 40 cm)	coarse blocks, head-sized cobbles, with a variable percentages of cobble, gravel and sand
Mesolithal (>6 cm to 20 cm)	fist to hand-sized cobbles with a variable percentage of gravel and sand
Microlithal (>2 cm to 6 cm)	coarse gravel, (size of a pigeon egg to child's fist) with variable percentages of medium to fine gravel
Akal (>0.2 cm to 2 cm)	fine to medium-sized gravel
Psammal/psammopelal (>6 µm to 2 mm)	sand and mud
Argyllal (<6 µm)	silt, loam, clay (inorganic)
Biotic microhabitats	**Definition**
Phytal	floating stands or mats, lawns of bacteria or fungi, and tufts, often with aggregations of detritus, moss or algal mats (interphytal: habitat within a vegetation stand, plant mats or clumps)
Algae	filamentous algae, algal tufts
Submerged macrophytes	macrophytes, including moss and Characeae
Emergent macrophytes	e.g. *Typha, Carex, Phragmites*
Living parts of terrestrial plants	fine roots, floating riparian vegetation
Xylal (wood)	tree trunks, dead wood, branches, roots
CPOM	deposits of coarse particulate organic matter, e.g. fallen leaves
FPOM	deposits of fine particulate organic matter
Sewage bacteria and –fungi and saprobel	sewage bacteria and –fungi, (*Sphaerotilus, Leptomitus*), sulfur bacteria (e.g. *Beggiatoa, Thiothrix*), sludge
Organic mud	mud and sludge (organic) = pelal
Debris	organic and inorganic matter deposited within the splash zone area by wave motion and changing water levels, e.g. mussel shells, snail shells

Table 3: Sampling design, classification of siltation degree and number of sampling compartments. Samples marked with a * indicate samples where 4 sampling units were stored in one box.

River	Site	Siltation Class	MHS	Macro-/Mesolithal	Mesolithal	Microlithal	Akal	Akal/Psammal	Psammal	Roots	Xylal	Shoreline grass	Moss
Rodl	Reference	0	1		4					1*			
	Impacted	2	1		4		4				1*		
Gr. Mühl	Reference (Riffle/Pool)	0	1			4	4				1*		
	Impacted (Riffle/Pool)	3 to 3+	1			4	4	4			1*		
	Reference (Plain)	0	1	4								1*	
	Impacted (Plain)	2 to 3	1	4								1*	
Kl. Mühl	Impacted (Riffle/Pool)	2	1	4	4					1*			
	Impacted (Plain)	3	1				4		4 (+MP)				
Osterbach	Reference	0	1		4	4				1*	1*		
	Impacted	3	1				4		4		1*		1*
Gusen	Reference	0	1										
	Impacted	3+	1			4	4				1*		

At each site abiotic parameters including water temperature, O_2 saturation and content, pH and conductivity were measured with a Hach Lange Hq30d portable meter. Mean and maximum flow velocity as well as mean and maximum water depth were measured with a Hach-Lange flow mate.

The samples were taken with a AQEM Consortium conform 25 cm edge length benthic invertebrate Multi-Habitat-Sampling net with 500 um mesh size (AQEM Consortium, 2002). Directly after sampling each sample has been stored in separate plastic boxes and fixed with Formaldehyde to a final concentration of 4%. After 3 weeks of exposure to the Formaldehyde solution the samples were ready for determination.

3.2. LABORATORY WORK

Of the 92 taken single sampling units 81 were used for the analyses, the others were stored for the case that samples got lost or became unusable in another way. The 81 single sampling units were sieved in a scaled sieve tower with a mesh size from 1 cm down to 500 µm, to get rid of inorganic and plant material. In cases where it was obvious that there are far more then hundred individuals of a certain taxon, mostly Chironomidae and *Brachycentrus maculatus,* subsampling was applied for these taxa. After the determination to at least genus level, the samples were weighted in family or other taxa groups to calculate the biomass. Abundances and biomass for the determined taxa were entered in a taxa list in Microsoft Excel 2010.

Afterwards the samples were stored in 70% ethanol.

The individuals were determined using a Leica stereo microscope with a magnification up to 35 times. For determination, keys by Waringer & Graf (2011) for Trichoptera, Bauernfeind & Humpesch (2001) for Ephemeroptera and the screening level key by Moog et al. (2010) for the other taxagroups were used.

3.3. DATA PROCESSING

The results of the determination process as well as the biomass were entered into a taxa list, in Microsoft Excel. This taxa list was the base for further analysis via Ecoprof 4.0 software (Moog et al., 2013), PCORD 5.33 (McCune & Mefford, 2006) and IBM SPSS Statistics 20.

Via Ecoprof the saprobic status and metrics like number of total taxa, number of EPT taxa and functional feeding guilds (FFG) were calculated according to benthic invertebrate data.

For the assessment of the saprobic status of a sampling site, the multiple habitat sampling samples (MHS), which were processed by Dr. Patrick Leitner were used. For all other analyses the 92 single sampling units were used.

3.3.1. METRICS

The calculation of the number of taxa, abundance, biomass, dominance of EPT taxa and the functional feeding guilds was based on the single sampling units and was done via Ecoprof 4.0 software (Moog et al., 2013).

3.3.2. PROPORTION OF SEDIMENT-SENSITIVE INVERTEBRATE INDEX (PSI)

The Proportion of Sediment-sensitive Invertebrate Index (PSI) by Extence et al. (2011) was calculated on the base of the single sampling units. To calculate the PSI every taxon has a fine sediment sensitivity rating (FSSR) from A - highly sensitive, to D - highly insensitive.

$$\mathrm{PSI}(\Psi) = \frac{\sum \text{Scores for Sediment Sensitivity Groups A \& B}}{\sum \text{Scores for all Sediment Sensitivity Groups A; B; C \& D}} \times 100$$

According to Extence et al. (2011) any appropriate sampling method can be used to collect raw data for PSI calculation, although Extence et al. (2011) recommend that for application in Britain, where the PSI was invented, the UK TAG methodology for macro-invertebrate sampling and analysis is used (Murray-Bligh et al., 1997; Chadd, 2010). This stipulates timed pond-net surveys, with different habitats being sampled in proportion to their occurrence. This approach is advocated because it accommodates ecological change associated with shifting sediment cover.

In this study the calculation of the PSI was done based on the data of the single sampling units, in order to work out the distinct differences between the different microhabitats.

3.3.3. Nonmetric Multidimensional Scaling (NMS) and Indicator species Analysis (ISA)

PCORD was utilized to perform Nonmetric Multidimensional Scaling (NMS) and Indicator species Analysis (ISA) to gather information on similarities and dissimilarities between each single sampling unit based on the benthic invertebrate species composition. The parameters that were included in the analyses are: river and site name, siltation class, whether organic material was present, water depth, flow velocities at the bottom and at 40 percent depth, biggest and smallest sediment fractions, the proportion of sediment-sensitive invertebrate index (PSI) and the total number of taxa.

Non-metric Multidimensional Scaling (NMS) is an ordination method that is well suited to data that are non-normal or are on arbitrary, discontinuous, or otherwise questionable scales. For this reason, NMS should probably be used in ecology more often than it is. This method can be used both as an ordination technique and as a method for assessing the dimensionality of a data set (McCune & Mefford, 2006).

A very common goal in community analysis is to detect and describe the value of different species for indicating environmental conditions. If environmental differences are conceptualized as groups of sample units, then Dufrene and Legendre's (1997) method of calculating species indicator values provides a simple, intuitive solution. The method combines information of the concentration of species’ abundance in a particular group and the faithfulness of occurrence of a species in a particular group. It produces indicator values for each species in each group. These are tested for statistical significance using a randomization technique (McCune & Mefford, 2006). In this study 0.005 has been chosen as the level of significance, taxa which fulfil the criteria are displayed in bold in the results, and the different taxa are sorted in descending order corresponding to their p-value. The indicator values range from zero, no indication, to 100, perfect indication. Perfect indication means that presence of a species points to a particular group without error (McCune & Mefford, 2006).

3.3.4. Ecological Quality Class and Saprobity

The ecological quality class and the saprobity were based on the Multi Habitat samples (MHS), which were taken by Dr. Patrick Leitner according to the AQEM method (AQEM Consortium, 2002).It aims to apply a multi- habitat approach designed for sampling major habitats proportionally according to their share within a sampling reach. One MHS sample consists of 20 individual samples, each representing 5% of the total sample. The aim of the multi habitat sampling approach is that the 20 partial samples represent the present distribution of microhabitats at a given sampling site. For example, if 20% akal, 50% microlithal and 30% mesolithal are observed, than 4 akal, 10 microlithal and 6 mesolithal partial samples have to be taken to represent the site properly. For the sampling itself a 25 cm edge length benthic invertebrate Multi-Habitat-Sampling net with 500 um mesh size is used thus covering 1,25 m^2 of the river bottom with each MHS sample. The calculation of the ecological quality class was done according to the water framework directive via the detailed benthic invertebrate method. According to Ofenböck et al. (2010) three assessment modules contribute to the calculation of the ecological quality class. The saprobitiy module, where the saprobic index after Zelinka and Marvan (1961) is calculated. The module of general degradation, where one or two, depending on the river typology, multi metric indices are calculated which aim to address, potamalisation effects, rhithralisation effects and toxic stress. The third module is the module of acidification. For the final classification all three modules are combined following a worst case approach. The overall ecological quality class can’t be better than the worst result of a single module, for example when the acidification and the general degradation module result in quality class 1 but the saprobic index results in quality class 3 then the overall ecological quality class is 3.

4. RESULTS

4.1. SINGLE SAMPLING UNITS

4.1.1. NUMBER OF TAXA

The analysis of the number of taxa per siltation class, shown in figure 10, indicates that the number of taxa in the **reference** class, class 2 and class 2-3 is similar. At siltation class 3 the mean number of taxa decreases by 50%, this marks the lowest mean number of taxa of all siltation classes. At the higher classes 3-3+ and 3+ the number of taxa increases slightly but are still noticeably lower than in the first three siltation classes.

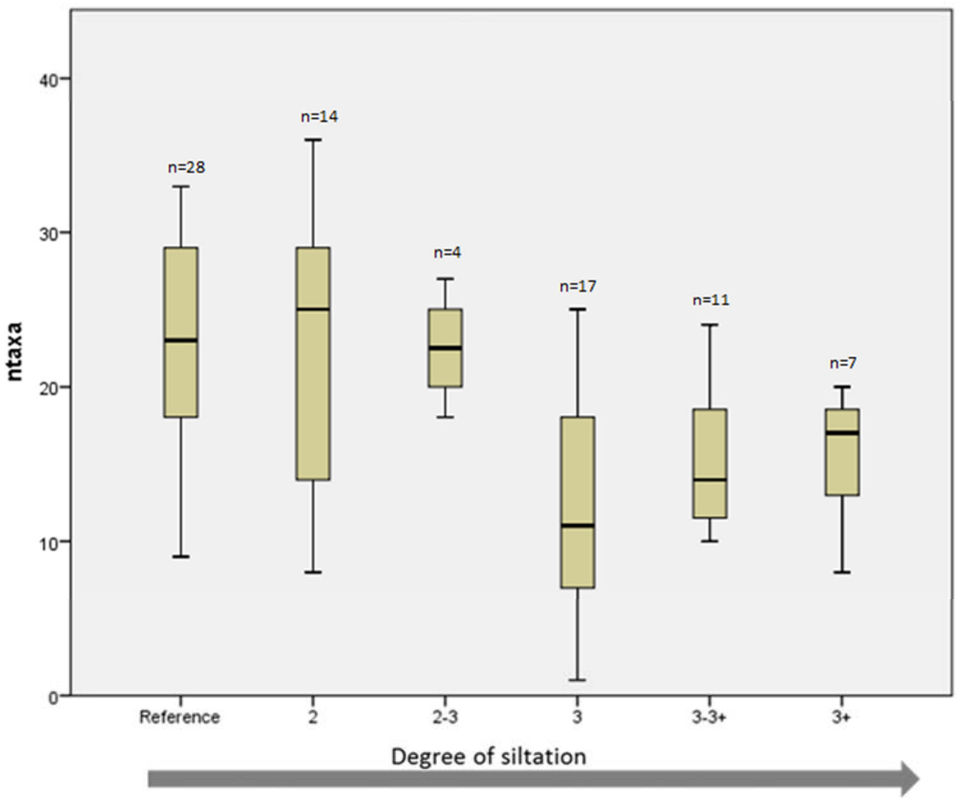

Figure 10: Number of taxa per siltation class.

The analysis of number of taxa per microhabitat shows a similar result like the number of taxa per siltation class. As it is visible in figure 11, the number of taxa stays at a comparable level from the coarse macrolithal to the microlithal samples from the **reference** site. In the macrolithal samples from **impacted** sites, the number of taxa is lower than in the coarser fractions and decreases alongside the grain size gradient. The samples from the akal **impacted** sites have a slightly higher number of taxa than the akal samples from the akal **reference** site. The lowest number of taxa occurs at the finest fraction, the psammal from **impacted** sites. The samples containing organic matter range between the coarse fractions and the fine fractions show high variability.

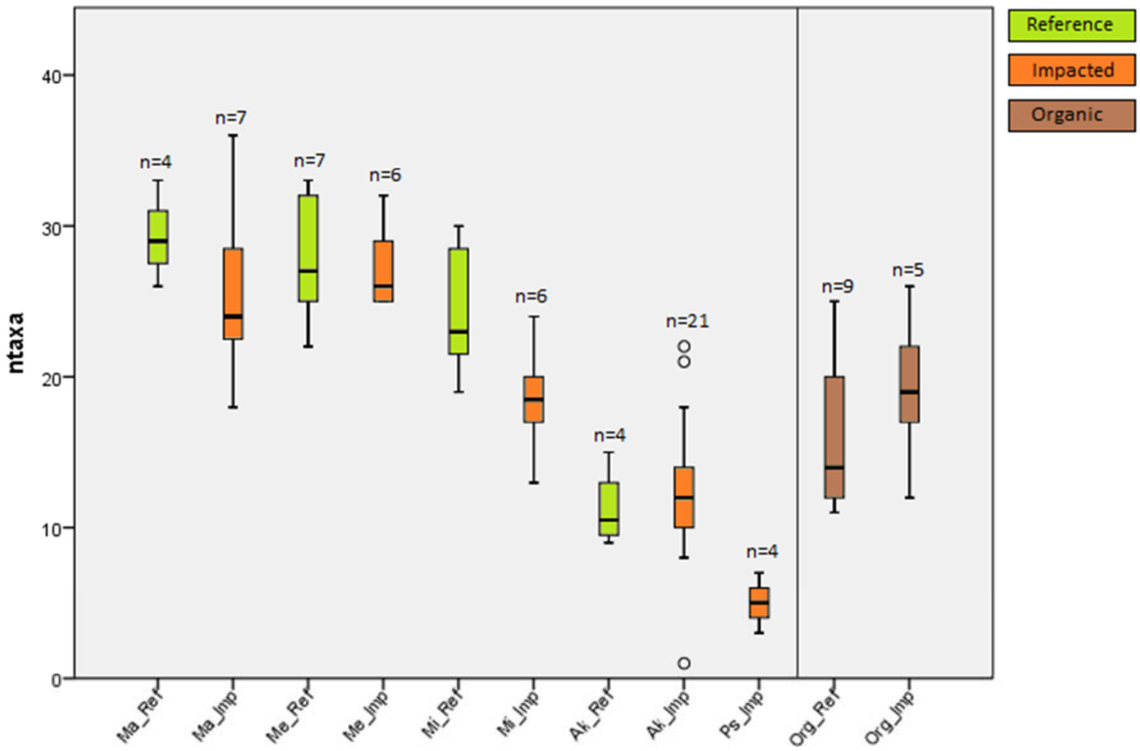

Figure 11: Number of taxa per microhabitat and condition.

4.1.2. ABUNDANCE

The analysis of abundance of individuals per siltation class, illustrated in figure 12, shows that the average abundances in the first three siltation classes are at a comparable level. At siltation class 3 and 3-3+ the abundances are noticeably lower, whereas siltation class 3+ sites are comparable to the first three classes. What is clearly visible is that sites from siltation class 2 have by far the highest abundance with up to six times more individuals than in any other class.

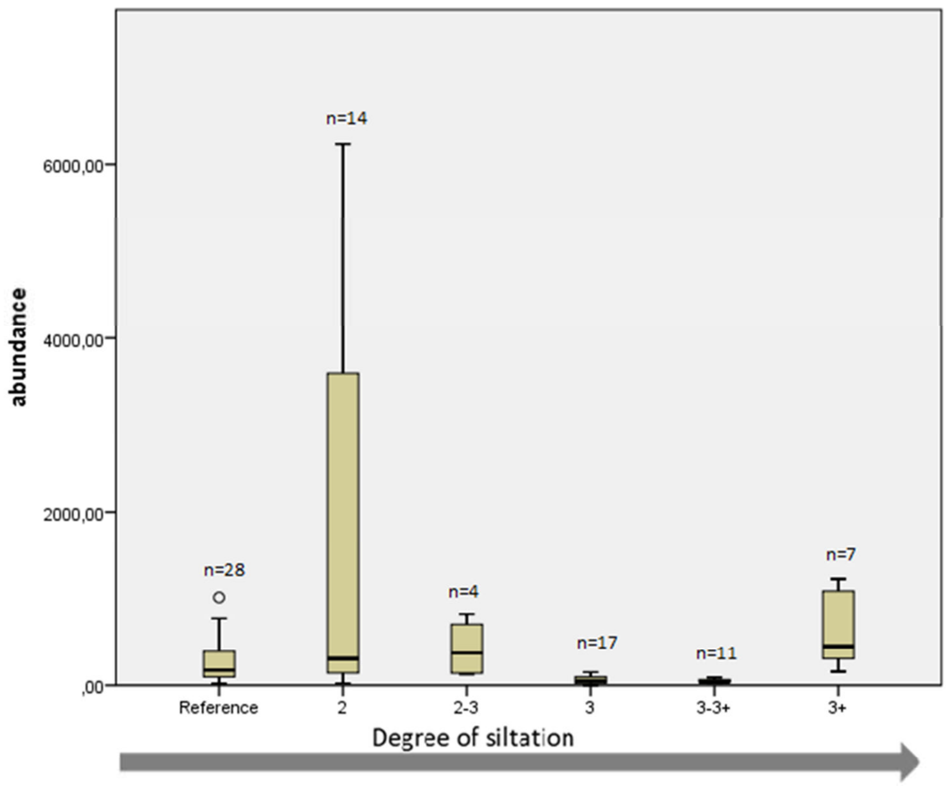

Figure 12: Abundance of individuals per siltation class.

The analysis of abundance of individuals per microhabitat, depicted in figure 13, shows a slight gradient from coarser to finer fractions, with the exception of the **impacted** macro- and mesolithal fractions that have significantly higher abundances. A real drop in the abundance is visible at akal **reference** sites and the numbers stay low up to the finest fractions of **impacted** psammal sites. The samples with organic matter have very low abundances and are comparable to the finer fractions.

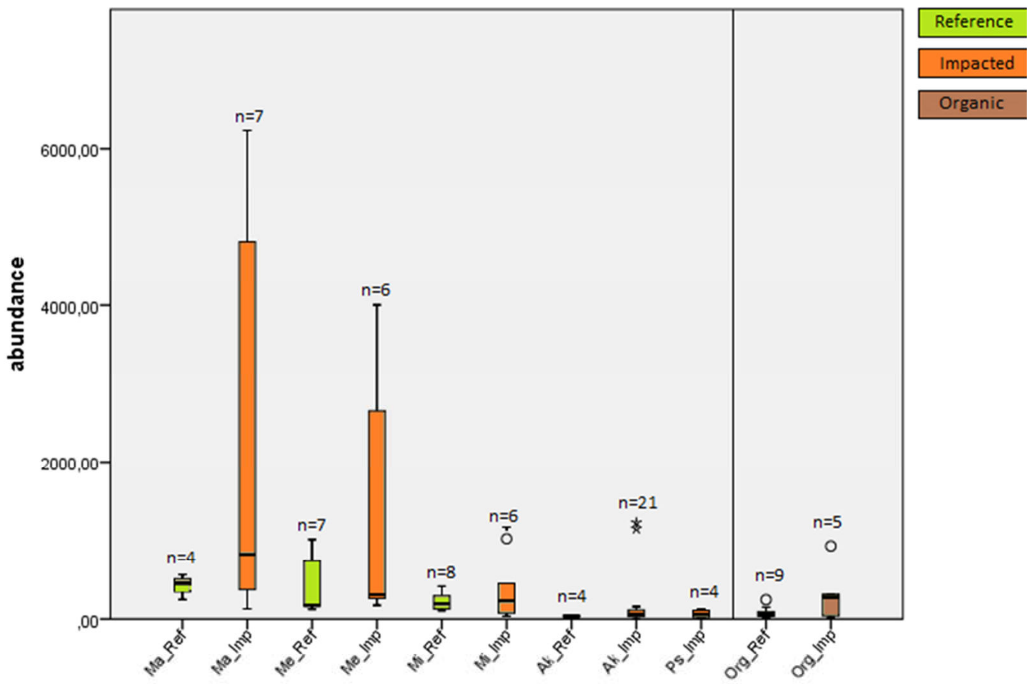

Figure 13: Abundance of individuals per microhabitat and condition.

4.1.3. Biomass

The analysis of biomass per siltation class, represented in figure 14, shows that the **reference** class, class 2-3 and class 3+ have a similar biomass averaging at around 1 gram per sample. The highest biomass is present in samples from siltation class 2-3 with an average biomass of 2 gram per sample. In siltation classes 3 and 3-3+ the biomass is about 50% lower than in the **reference** class. After the low levels of the classes 3 and 3-3+ the 3+ class shows an increased biomass in comparison to the neighbouring class.

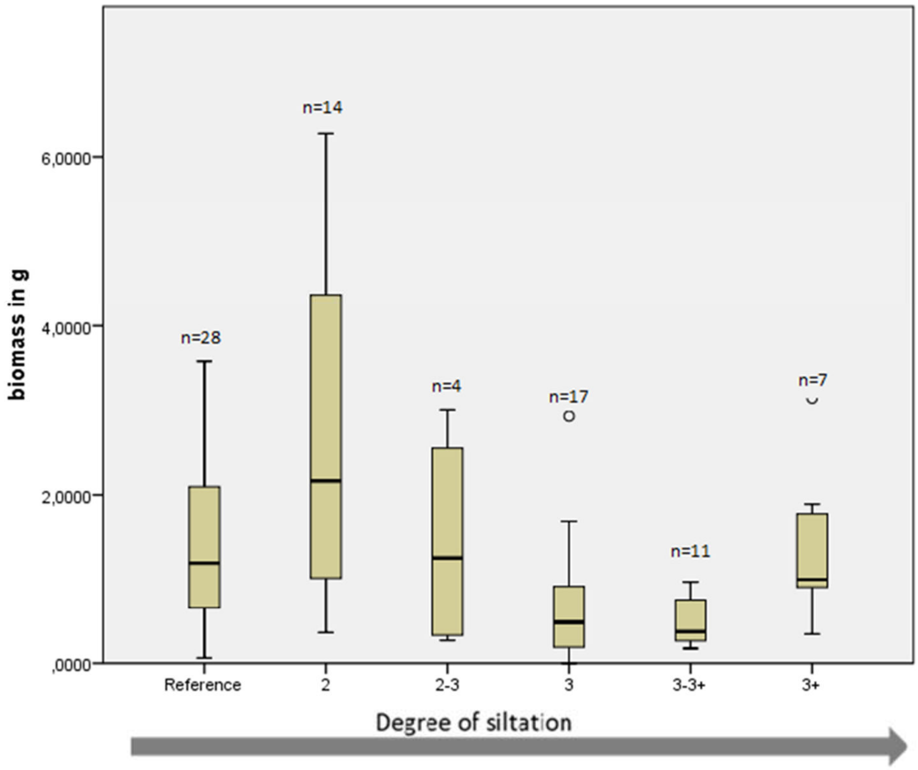

Figure 14: Biomass per siltation class.

The analysis of biomass per microhabitat, shown in figure 15, displays a general trend of biomass decrease alongside the grain size gradient. The samples from the **impacted** macrolithal represent the samples with the highest biomass, whereas the samples from **reference** akal and **impacted** psammal are the ones with the lowest biomass. The samples containing organic matter are similar to the coarser grain sizes. The analysis of the abundance of individuals per microhabitat and condition given in figure 17 shows that only few individuals are present in the organic habitats, the low abundance but fairly high biomasses, displayed in figure 19, indicate the occurrence of bigger individuals in the organic habitats.

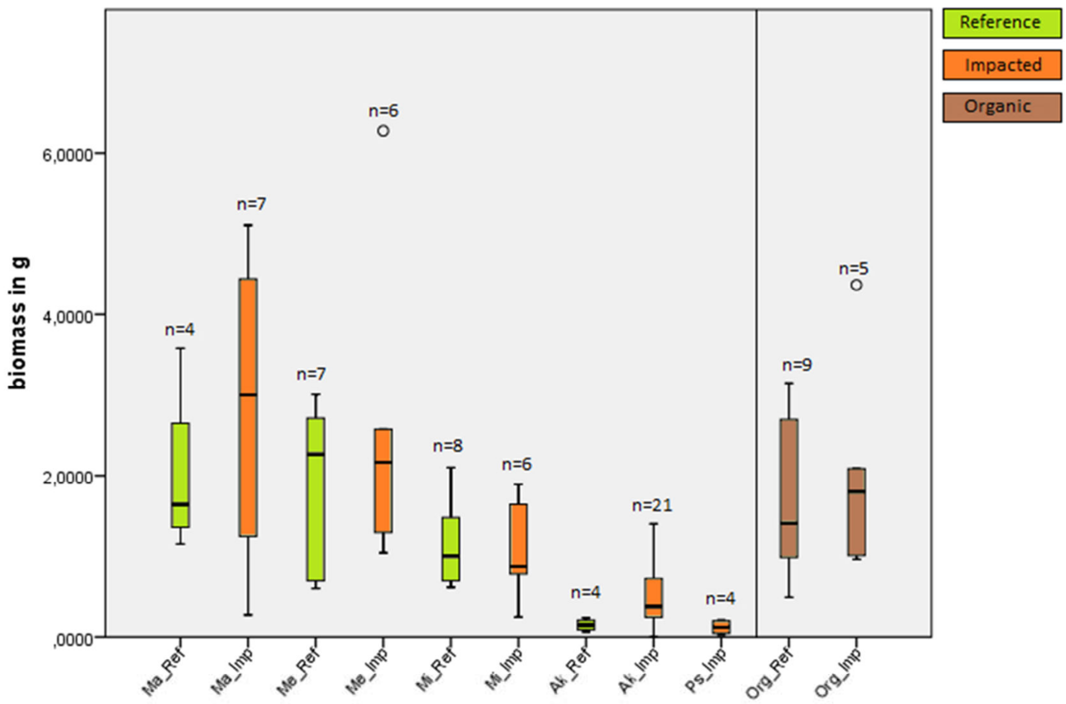

Figure 15: Biomass per microhabitat and condition.

4.1.4. ORDINATION OF THE SINGLE SAMPLES

Figures 16 and 17 show the non-metric multi-dimensional scaling ordination of the 81 benthic invertebrate samples. The red vectors indicate increasing biomass, abundance, number of taxa and PSI.

Figure 16 shows the joint plot of all sites with the overlay of the different siltation classes. A gradient from higher siltation classes, 3 and 3-3+ in the top right corner to the **reference** sites, siltation class 0, in the middle can be observed. The highest siltation class 3+ is grouped near the **reference** sites. In the bottom left corner six sampling sites with siltation class 2 cluster as a group of outliers. Even though a gradient is visible, the different groups of siltation classes are overlapped and not sharply separated.

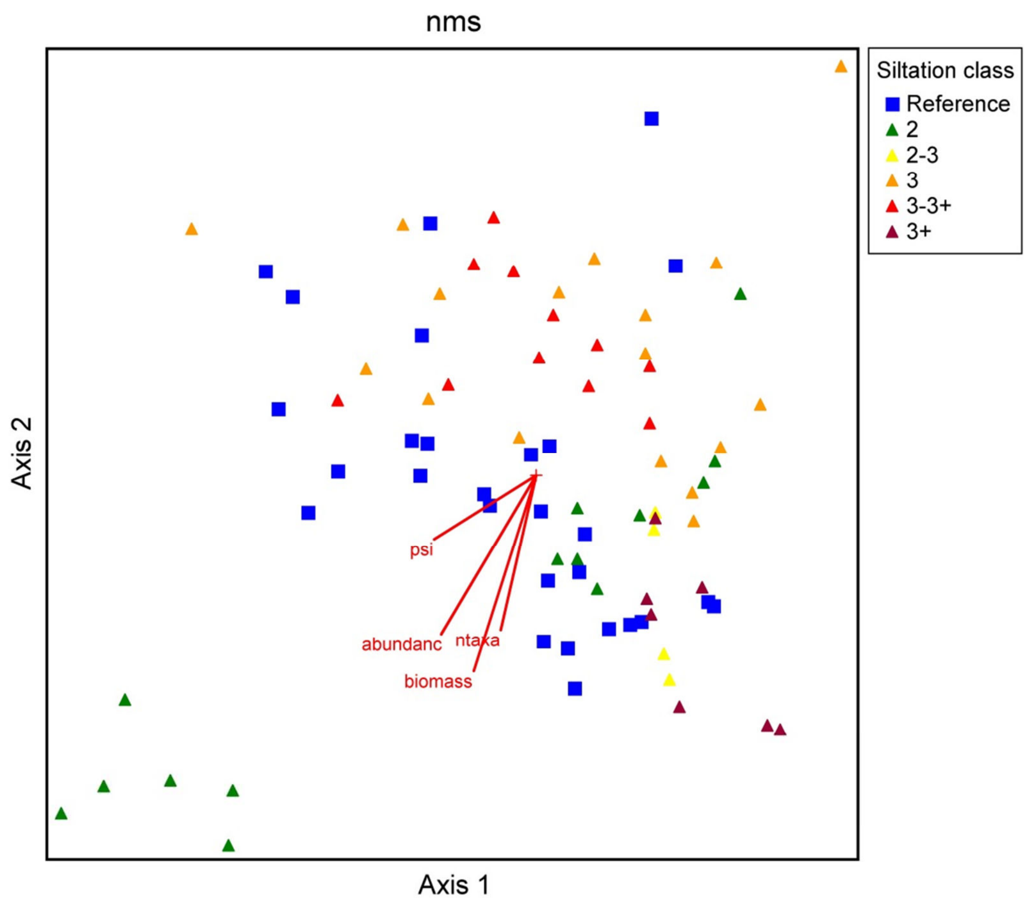

Figure 16: Joint plot of the NMS analysis for all macroinvertebrate samples (n = 81); colours represent the different siltation classes, vectors (joint plot cut-off value r2= 0.5) represent psi, abundance (abundanc), total number of taxa (ntaxa) and biomass. Final stress 16.31 for 2 – dimensional solution.

Figure 17 shows the overlay of the five rivers indicating differences between **reference** and **impacted** sites. A gradient from **impacted** towards **reference** sites is visible but no sharply separated groups occur, with the exception of six samples from the Kleine Mühl.

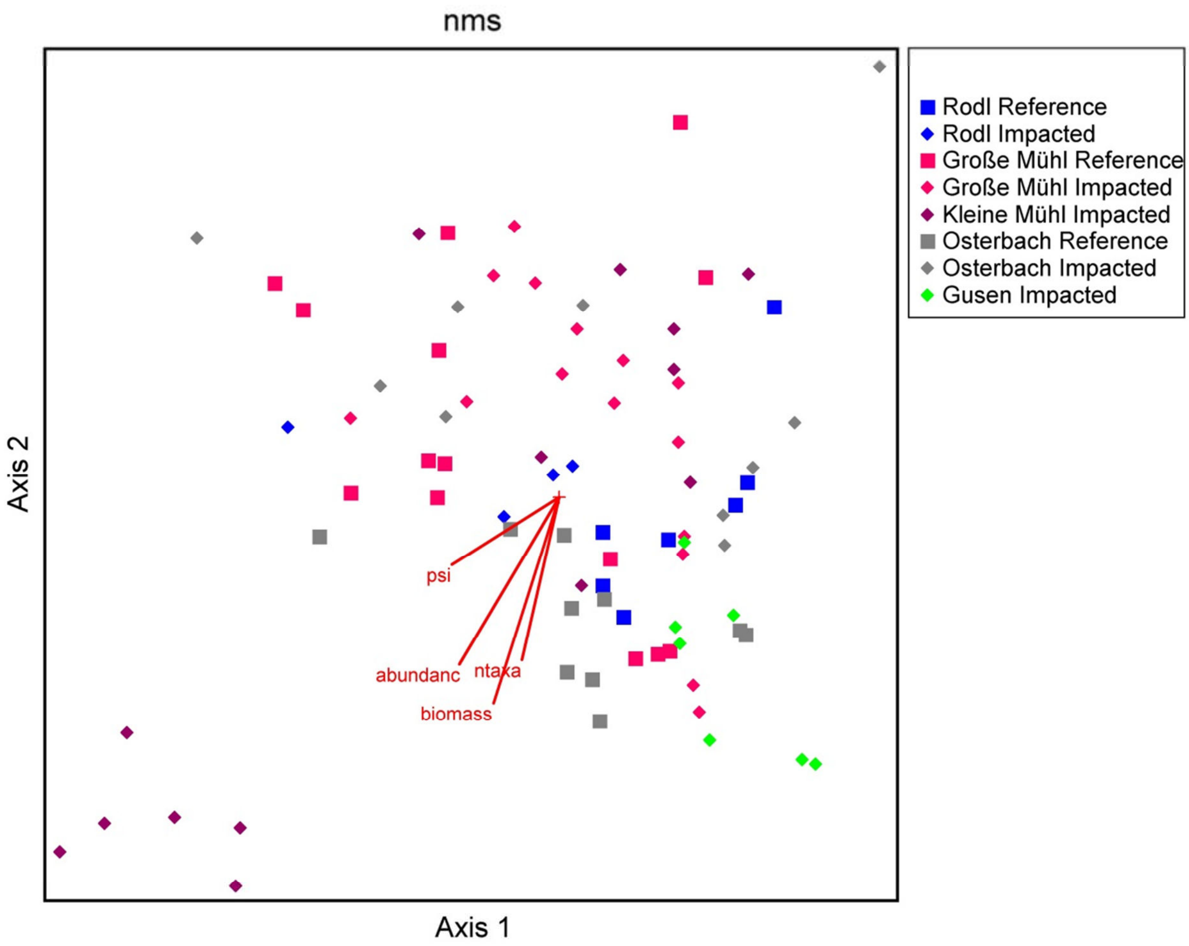

Figure 17: Joint plot of the NMS analysis for all macroinvertebrate samples (n = 81); colours represent the different sampling sites, vectors (joint plot cut-off value r2= 0.5) represent psi, abundance (abundanc), total number of taxa (ntaxa) and biomass. Final stress 16.31 for 2 – dimensional solution.

Figure 18 shows the differences between the different grain sizes and whether they are from **reference** or **impacted** sites. All **reference** and **impacted** samples from the coarser fractions (macro-, meso- and microlithal) taken from the same site form distinct groups and are distinguishable from each other. On the contrary, no distinct groups of samples can be observed at the akal fraction. The samples from **reference** and **impacted** sites overlap.

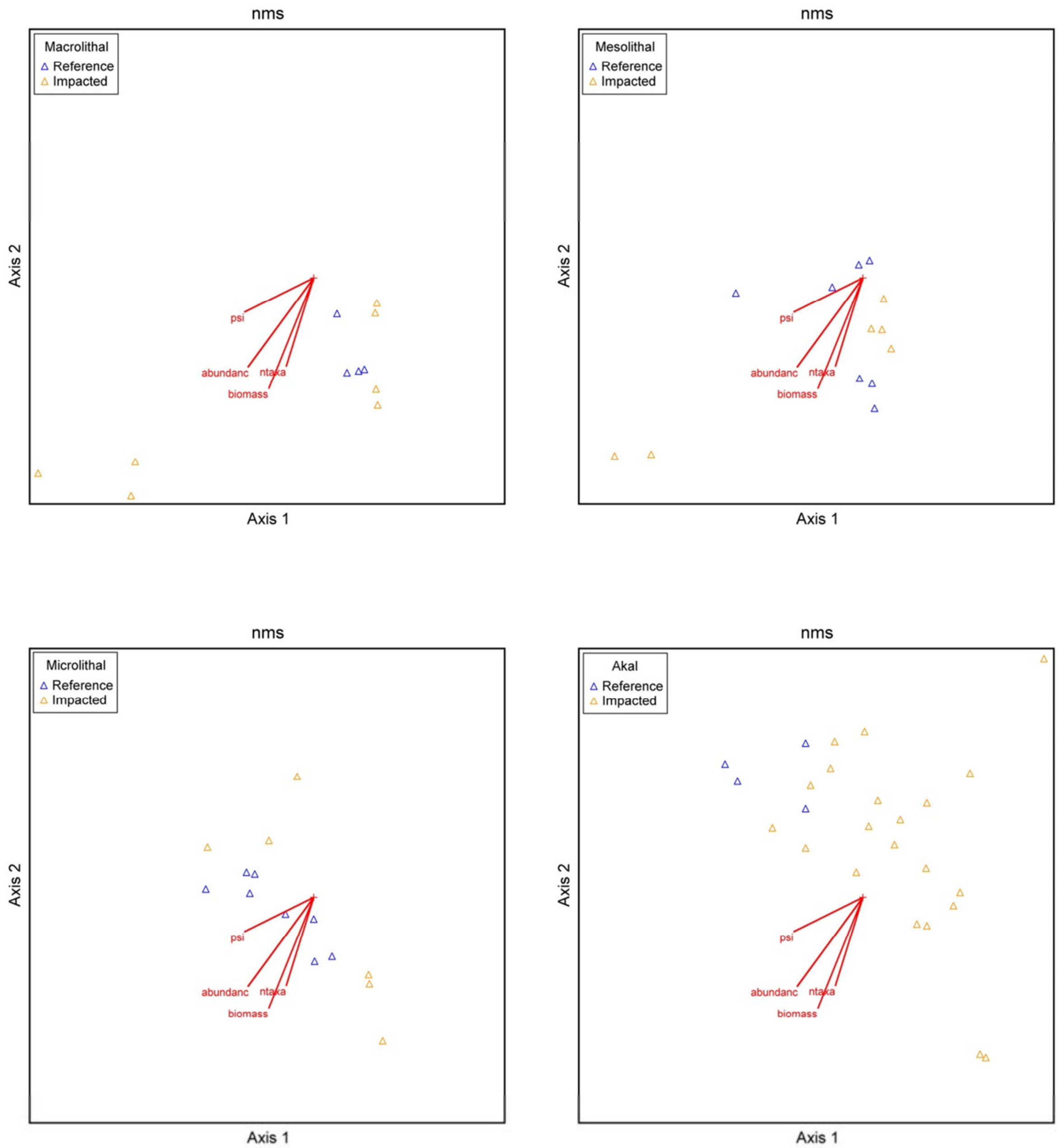

Figure 18: Four plots of the NMS analysis for four different grain sizes; blue represents reference samples whereas brown represents impacted samples, vectors (joint plot cut-off value r2= 0.5) represent psi, abundance (abundanc), total number of taxa (ntaxa) and biomass. Final stress 16.31 for 2 – dimensional solution.

4.1.5. Dominance of EPT-taxa

The analysis of the proportion of EPT taxa, the percentage of individuals belonging to either Ephemeroptera, Plecoptera or Trichoptera, per siltation class, given in figure 19, shows that there is only a slight variance in the percentage of EPT taxa alongside a gradient of increasing siltation.

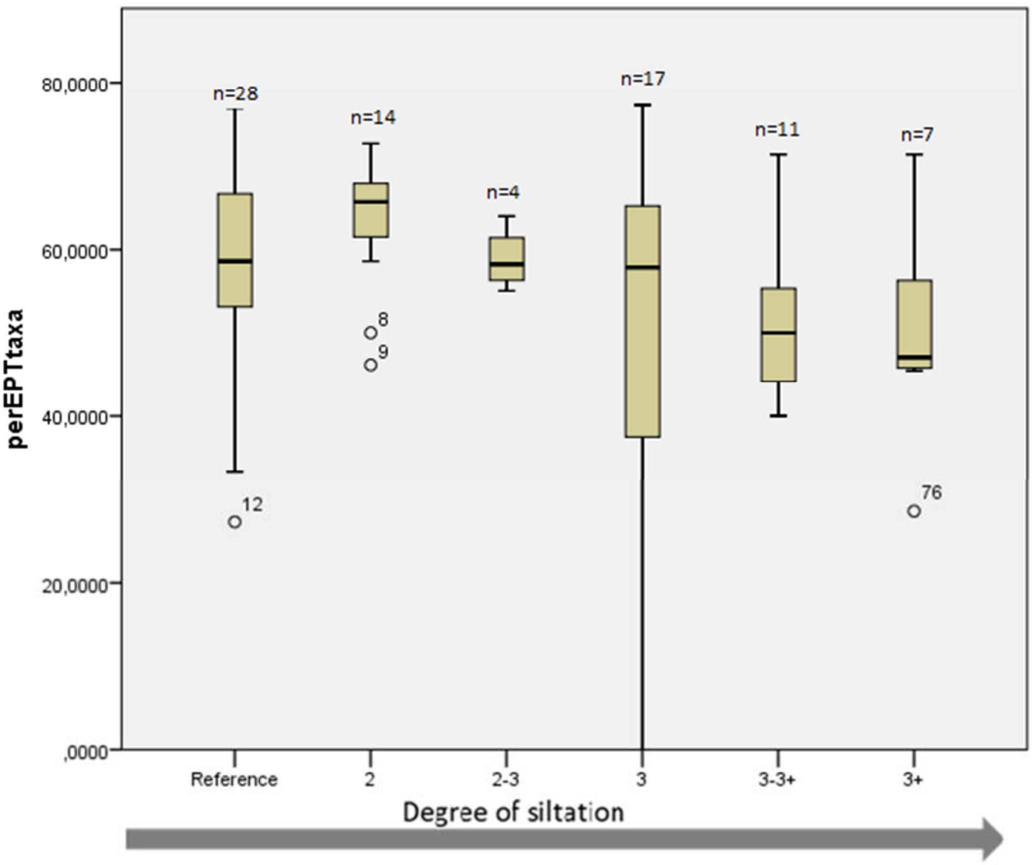

Figure 19: Percentage of EPT taxa per siltation class.

The analysis of the dominance of EPT taxa per microhabitat and preclassification, portrayed in figure 20, shows a trend of decreasing percentage of EPT taxa with decreasing grain size. A clear trend between the **impacted** and the **reference** samples is not observable. The samples that include organic matter have the highest percentage of EPT taxa.

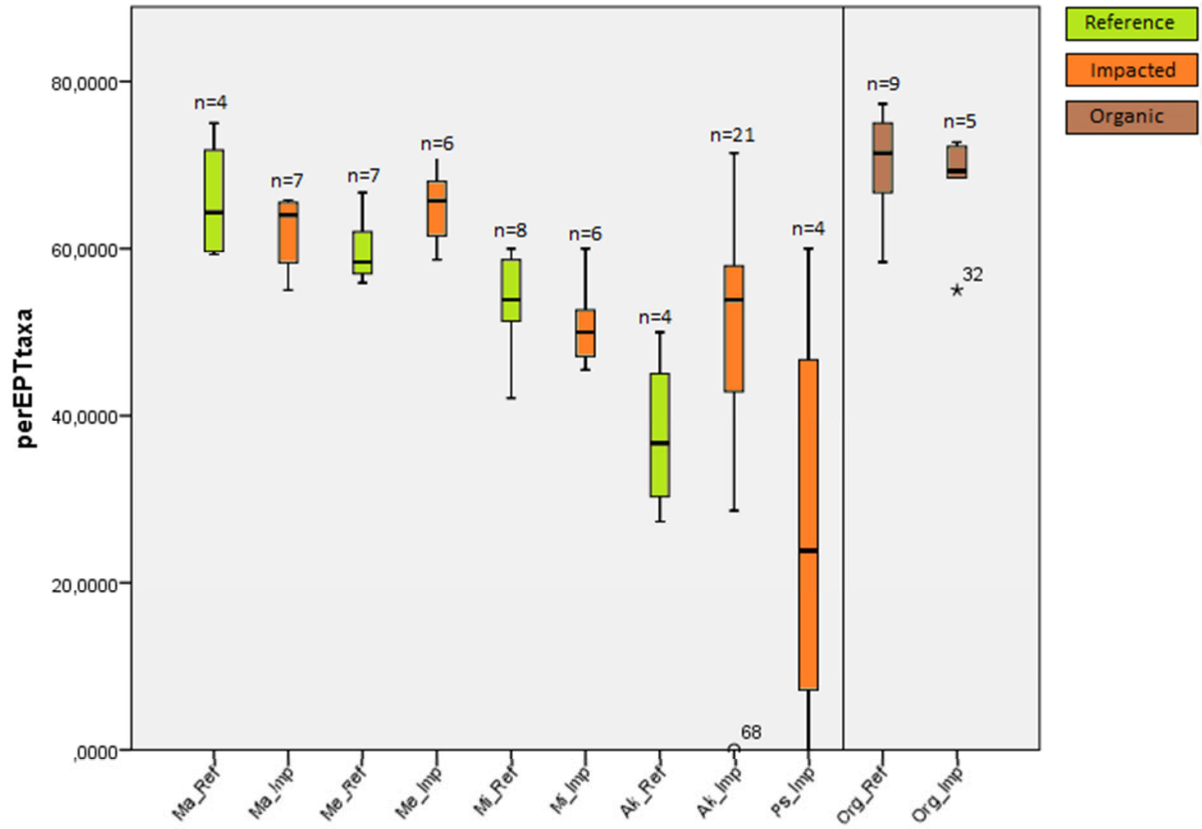

Figure 20: Percentage of EPT- taxa per microhabitat and condition.

4.1.6. Functional Feeding Guilds

The result of the analysis of the functional feeding guild distribution per site is shown in figure 21. Two trends are visible. The percentage of grazers is at a comparable level at the four **reference** sites and the siltation class II site, ranging between 40 and 55%. With increasing siltation class, starting at the KLM_FKV2 site, the percentage of grazers decreases to values between 12% and 30%. Regarding the predators an opposite trend is visible. At the four **reference** sites and three **impacted** sites the percentage of predators is below 25 %. OSV3 has the highest proportion of predators with 52 %. With 27% predators the KLM_PBV3 site has the lowest amount of predators at the highly **impacted** sites. The two sites with the highest siltation class have a share of around 40% predators. Filter feeders have the highest share at the siltation class two and three, whereas the percentage is lower at the **reference** sites and the **impacted** sites with a siltation class higher than three. For the share of detritivorous taxa alongside the gradient of increasing siltation there is no clear trend visible.

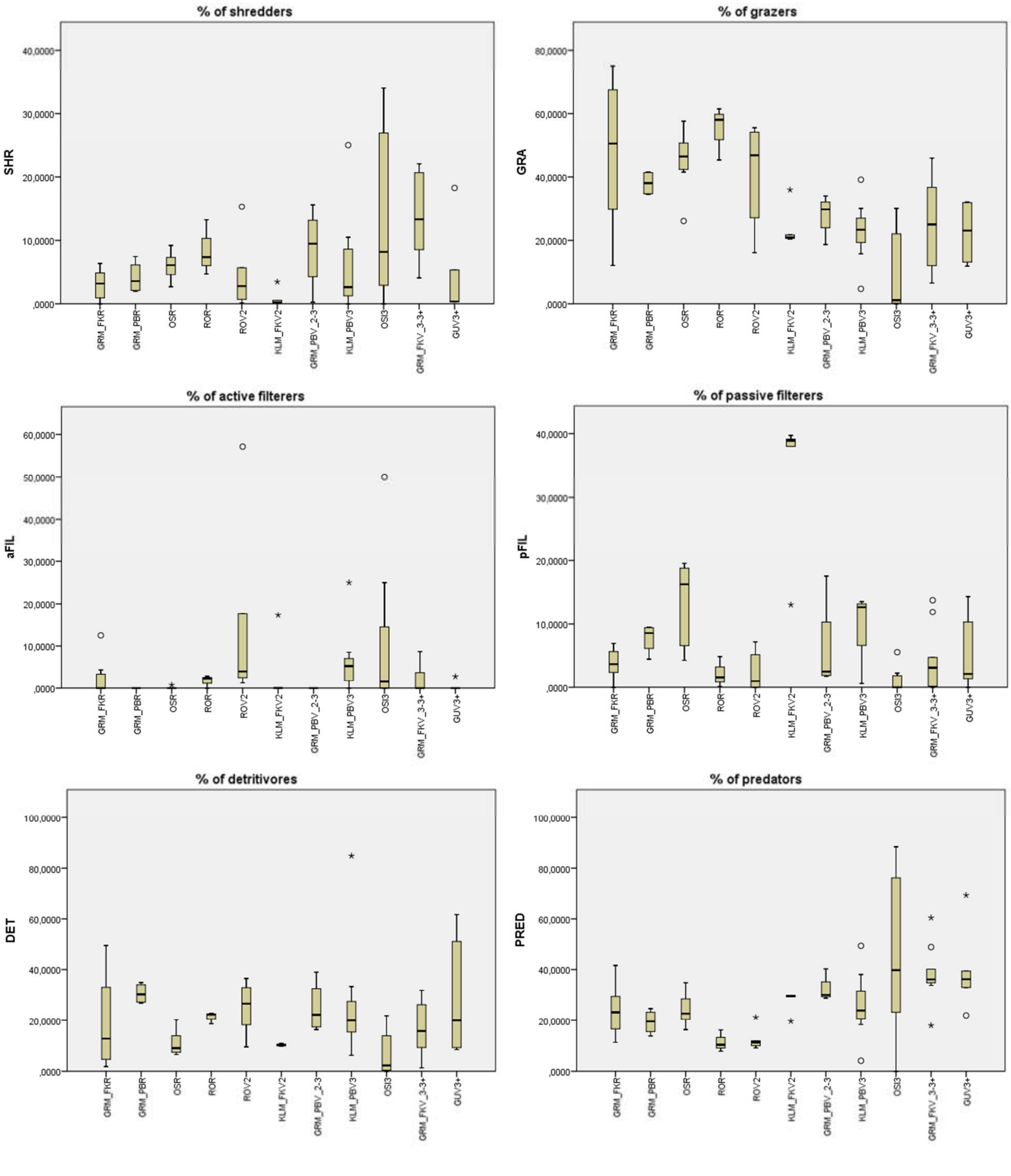

Figure 21: Functional feeding guilds per site.

4.1.7. PROPORTION OF SEDIMENT SENSITIVE INVERTEBRATE INDEX (PSI)

Figure 22 shows the results of PSI calculations per siltation class. It is visible that siltation classes **reference**, 2 and 2-3 have an average calculated PSI between 80 and 100, indicating minimally sedimented or unsedimented conditions. The classes 3, 3-3+ and 3+ have an average PSI between 60 and 80 indicating slightly sedimented conditions. The samples with the highest and the lowest PSI originate both from siltation class 3. The highest calculated PSI is 100 and the lowest 25, with the exception of one sample where no individuals were found and therefore no calculation was possible.

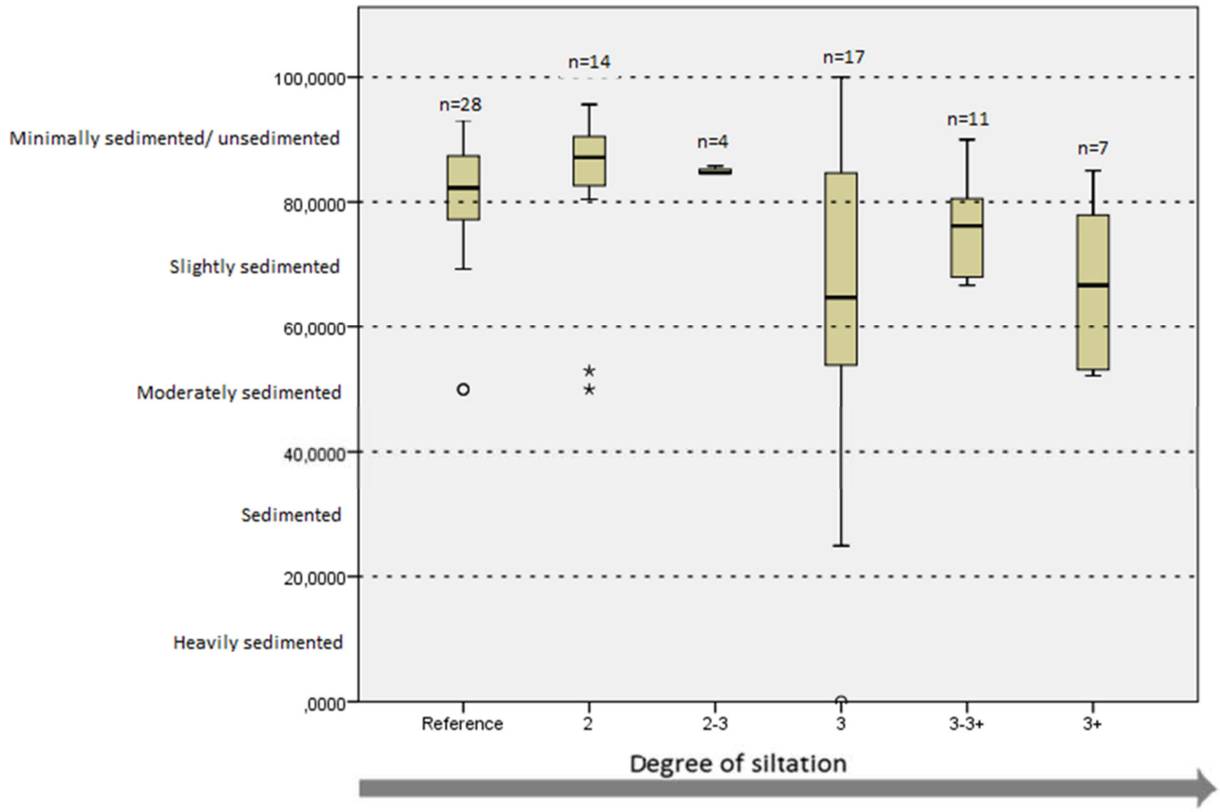

Figure 22: PSI-values per siltation class.

Figure 23 shows the results of the PSI values for the different microhabitats. The macro-, meso- and the microlithal samples from the **reference** site have an average PSI in the highest class between 80 and 100. From the microlithal **reference** samples to the **impacted** psammal, interdependency between decreasing grain size and decreasing PSI is apparent. The samples containing organic matter have an average PSI between 60 and 80 in the second best PSI class.

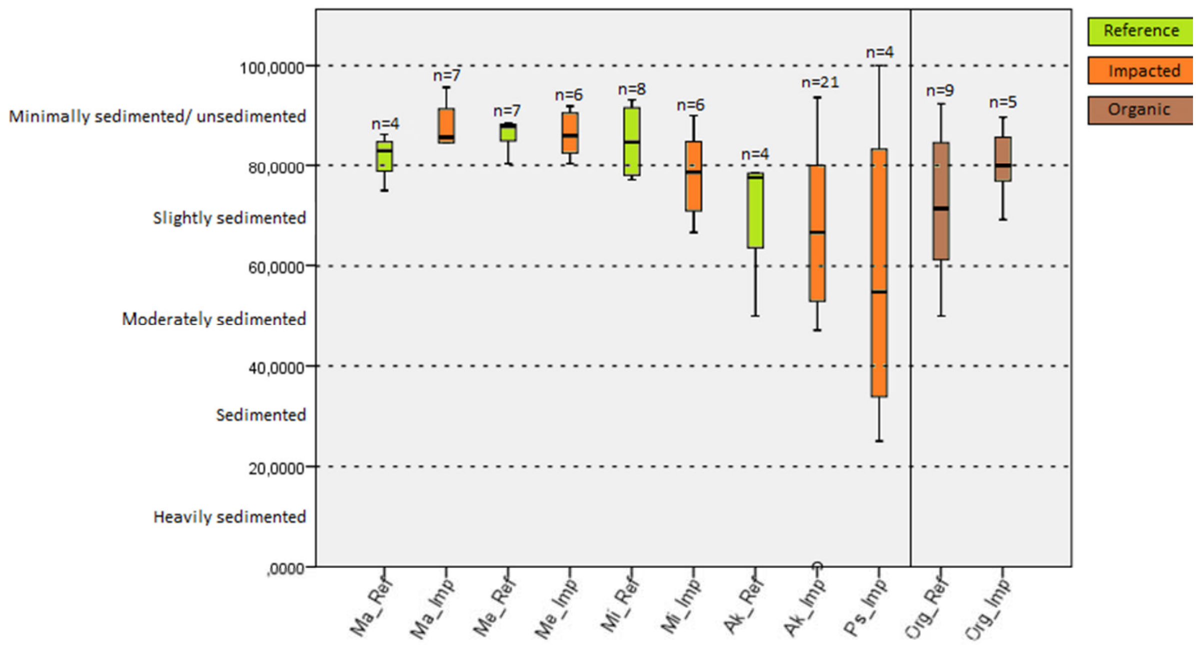

Figure 23: PSI-values per microhabitat and preclassification.

4.1.8. Indicator species Analysis

Table 5 shows the results of the indicator species analysis for the different siltation classes. For each siltation class the three best taxa are displayed in the table. The most significant results are in siltation class 2-3, where three significant taxa were calculated.

Table 4: Indicator species analysis per siltation class. Blue shading indicates the reference class, green shading indicates siltation class 2, very light orange siltation class 2-3, light orange indicates siltation class 3, orange indicates siltation class 3-3+ and red indicates the highest siltation class 3+.In the SR column the fine sediment sensitivity ratings (A –highly sensitive, B – moderately sensitive, C –moderately insensitive and D – highly insensitive) by Extence et al. (2011) are given. Taxa shown in bold are statistically significant (p<0,005).

Group	Taxon	Siltation class	Value	p	SR
Coleoptera	*Hydraena* sp	Reference	46	0,012	B
Ephemeroptera	*Rhithrogena* sp	Reference	39,8	0,014	A
Ephemeroptera	*Baetis* sp.	Reference	33,3	0,0704	A
Gastropoda	***Ancylus fluviatilis***	**2**	**52,5**	**0,0016**	**A**
Trichoptera	***Brachycentrus maculatus***	**2**	**57**	**0,0042**	**A**
Coleoptera	*Elmis* sp *ad*	2	58,7	0,0058	B
Ephemeroptera	***Habroleptoides confusa***	**2-3**	**59,3**	**0,0006**	**B**
Diptera	**Psychodidae (dunkel)**	**2-3**	**65,5**	**0,0006**	**D**
Trichoptera	***Rhyacophila* s. str.**	**2-3**	**51**	**0,002**	**A**
Hirudinea	*Dina punctata*	3	26,4	0,042	D
Crustacea	*Gammarus fossarum*	3	34,2	0,0852	B
Bivalvia	*Pisidium* sp	3	9,4	0,5353	D
Coleoptera	*Oreodytes sanmarkii*	3-3+	30	0,0214	B
Gastropoda	*Radix* sp	3-3+	20,6	0,069	D
Trichoptera	*Allogamus auricollis*	3-3+	26,8	0,0916	B
Ephemeroptera	*Ephemerella notata*	3+	44,4	0,0062	B
Anisoptera	*Ophiogomphus cecilia*	3+	26	0,0162	D
Nematoda	Nematoda	3+	35,7	0,0194	/

No significant taxa were calculated for the **reference** class, for siltation class 2, two significant taxa were calculated namely *Ancylus fluviatilis* and *Brachycentrus maculatus* which both are rated A, highly sensitive by Extence et al. (2011). For siltation class 2-3, three significant taxa were calculated, namely *Habroleptoides confusa*, Psychodidae (dunkel) and *Rhyacophila* sensu strictu type. For the siltation classes 3, 3-3+ and 3+ no significant taxa could be calculated. A trend in the sensitivity ratings with increasing siltation class is visible. The **reference** class and class 2 each have two taxa rated highly sensitive and one taxon rated moderately sensitive. In the higher silation classes, namely 2-3, 3, 3-3+ and 3+ at least one taxon occurs with a moderately sensitive sensitivity rating. Whereas the other taxa in these classes are rated highly insensitive.

Table 6 gives the results of the indicator species analysis for the different microhabitats. The table is sorted alongside a grainsize gradient from top to bottom with the exception of the organic microhabitats at the very end of the table. Green shaded rows indicate samples from **reference** sites whereas orange shaded rows indicate **impacted** samples. The organic samples are shaded in brown. The table shows all the significant taxa and the first taxa above the significance level per category.

Table 5: Indicator species analysis per microhabitat, green shaded rows indicate samples from reference sites, orange shaded rows indicate impacted samples, organic samples are shaded in brown, the increasing intensity of the shading at the reference and impact rows reflects the decreasing grain size. In the SR column the fine sediment sensitivity ratings (A –highly sensitive, B – moderately sensitive, C –moderately insensitive and D – highly insensitive) by Extence et al. (2011) are given Taxa shown in bold are statistically significant (p<0,005).

Group	Taxon	microhabitat	Value	p	SR
Ephemeroptera	***Baetis* sp.**	**Ma_Ref**	**41,9**	**0,0002**	**A**
Ephemeroptera	***Ephemerella mucronata***	**Ma_Ref**	**61,5**	**0,0002**	**A**
Trichoptera	***Rhyacophila* s. str.**	**Ma_Ref**	**55,5**	**0,0002**	**A**
Plecoptera	***Amphinemura* sp.**	**Ma_Ref**	**74,3**	**0,0004**	**B**
Diptera	**Psychodidae (dunkel)**	**Ma_Ref**	**76**	**0,0004**	**D**
Diptera	**Simuliidae Gen sp.**	**Ma_Ref**	**57,2**	**0,001**	**A**
Trichoptera	***Sericostoma* sp.**	**Ma_Ref**	**38,1**	**0,0034**	**B**
Ephemeroptera	*Habroleptoides confusa*	Ma_Ref	37,4	0,007	B
Coleoptera	***Orectochylus villosus***	**Ma_Imp**	**51,6**	**0,0008**	**A**
Plecoptera	*Siphonoperla* sp.	Ma_Imp	45,1	0,009	A
Ephemeroptera	***Epeorus assimilis***	**Me_Ref**	**58,5**	**0,0008**	**A**
Diptera	*Ibisia marginata*	Me_Ref	40,1	0,0054	A
Ephemeroptera	***Ephemera danica***	**Me_Imp**	**64,9**	**0,0002**	**C**
Ephemeroptera	***Ephemerella major***	**Me_Imp**	**48**	**0,002**	**A**
Trichoptera	***Silo nigricornis***	**Me_Imp**	**48,4**	**0,002**	**A**
Gastropoda	***Ancylus fluviatilis***	**Me_Imp**	**44,4**	**0,0038**	**A**
Coleoptera	**Esolus sp. Ad**	**Me_Imp**	**36,9**	**0,004**	**B**
Diptera	*Antocha* sp.	Me_Imp	42,2	0,0082	B
Trichoptera	***Anomalopterygella chauviniana***	**Mi_Ref**	**35,5**	**0,001**	**B**
Coleoptera	**Hydraena sp.**	**Mi_Ref**	**42,6**	**0,0046**	**B**
Ephemeroptera	*Rhithrogena* sp.	Mi_Ref	33,9	0,0076	A
Gastropoda	*Radix* sp.	Mi_Imp	29,9	0,0304	D
Bivalvia	*Pisidium* sp.	Ak_Ref	28,6	0,02	D
Trichoptera	*Anabolia* sp.	Ak_Imp	9,5	0,7341	C
Hirudinea	*Dina punctata*	Org_Ref	32,1	0,0314	D
Trichoptera	***Polycentropus flavomaculatus***	**Org_Imp**	**50**	**0,0012**	**B**
Trichoptera	*Halesus* sp.	Org_Imp	33	0,0188	C

For the macro-, meso- and the microlithal samples at the **reference** site, there are significant indicator species. For the finer fractions, microlithal **impacted** and akal, there are no significant species available. This indicates that there are indicator species for good and unsilted conditions but no indicators for silted conditions. Regarding the fine sediment sensitivity ratings a shift from sensitive to insensitive species is visible alongside the decreasing grain size gradient, with the exception of Psychodidae (dunkel) which are calculated as a significant indicator species for macrolithal but are rated highly insensitive.

4.2. MHS SAMPLES

4.2.1. ECOLOGICAL QUALITY CLASS AND SAPROBITY.

Table 4 comprises the siltation classes, the bed type, saprobic index and the ecological quality class of the five rivers based on MHS samples. From **reference** class up to siltation class 2-3, the sites have a good ecological status. Two sites from the Kleine Mühl and Große Mühl with a siltation class of 3 and 3-3+ indicate an ecological quality class of 3 - moderate. The site with the highest siltation class of 3+ at the Gusen has an ecological quality class of 2, good.

Regarding the saprobic index, all sites, except the two sites at the Kleine Mühl, are in good status. According to the saprobic index, siltation class 2 site at the Kleine Mühl, has a high saprobic status, whereas the siltation class 3 site at the Kleine Mühl has a moderate saprobic status.

There is only a slightly visible interdependency between increasing siltation class and decreasing ecological quality class.

Table 6: Overview of the different rivers, bed types, saprobic indices and the ecological quality classes calculated from the MHS samples. Blue represents high status, green represents good status and yellow represents moderate status. The share of fine sediment has been derived from the share of akal and finer fractions in the MHS samples.

Type	River	Siltation Class	Type 2	SI	MMI 1	MMI 2	Ecological Status	Share of akal and finer fractions in the MHS
Referenz	Große Mühl	0	PB	1,88	0,87	0,95	gut (good)	0
Referenz	Große Mühl	0	FK	1,69	0,87	0,86	gut (good)	5
Referenz	Gusen	0	PB	1,93	0,86	0,94	gut (good)	10
Referenz	Osterbach	0	PB	1,89	0,85	0,81	gut (good)	5
Referenz	Rodl	0	FK	1,83	0,96	0,99	gut (good)	5
Versandet	Rodl	2	PB	1,94	0,81	0,86	gut (good)	35
Versandet	Kleine Mühl	2	FK	1,74	0,78	0,76	gut (good)	40
Versandet	Große Mühl	2-3	PB	1,9	0,74	0,72	gut (good)	0
Versandet	Osterbach	3	PB	1,64	0,65	0,61	gut (good)	[illegible]
Versandet	Kleine Mühl	3	PB	2,35	0,52	0,48	mäßig (moderate)	[illegible]
Versandet	Große Mühl	3-3+	FK	1,59	0,48	0,42	mäßig (moderate)	[illegible]
Versandet	Gusen	3+	PB	2,06	0,6	0,58	gut (good)	[illegible]

Leitner (2015)

5. Discussion

5.2. Community structure and siltation class

Wood & Armitage (1997) pointed out that with increasing sediment load, abundance and diversity of benthic invertebrates decrease. The Ordination, via NMS analysis, of the fauna (figure 16 and 17) shows that this applies for the study area too. There is a group of outliers in the NMS analysis. All of the samples that group together in the bottom left corner of the NMS analysis contain very high number of individuals and different taxa. With 6233 individuals the KLM_FKV2_MaeMo1 sample shows the highest number of individuals in the whole study. KLM_FKV2_MaeMo4 being the sample with 36 taxa, the highest number of different taxa in the whole study. A closer look at the taxa list of the KLM_FKV2 samples reveals that up to 90% of the total individuals are *Brachycentrus maculatus. B. maculatus* occurs in meta- and hyporhithral rivers and prefers stable substrates like micro- and macrolithal, as well as woody structures in a moderate current (Graf et al., 2008). The reason for this mass occurrence of *B. maculatus* could be that, even though the whole site is considered siltation class two, there are patches that meet exactly the requirements of *B. maculatus*, including coarser macrolithal fractions and woody debris. Figure 24 shows a woody branch with *B. maculatus* attached to it from KLM_FKV2.

Figure 24: *Brachycentrus maculatus* at KLM_FKV2.

As observed and according to Jones et al. (2007) this mass occurrence of *Brachycentrus maculatus* does not occur at classes of higher siltation.

This shows the importance of habitat diversity and patchiness and that the invertebrate community is strongly correlated with mesoscale habitat patches like mentioned in Pardo & Armitage (1997).

The analysis of number of taxa for the different siltation classes (figure 10) shows that from **reference** up to siltation class 2-3 the level of number of taxa stay at a similar level, with slightly higher levels at siltation class 2 due to the occurrence of *B. maculatus.*

The Analysis of the biomass for the different siltation classes (figure 14) shows that the highest biomass occurs in siltation class 2. This, again, is the consequence of the above mentioned mass occurrence of *B. maculatus.* The **reference** class and class 2-3 have similar biomass. Siltation class 3 and 3-3+ have less than half of the biomass of the **reference** class, 3-3+ being the class with the lowest biomass. The 3+ class has a slightly lower biomass than the **reference** class but clearly higher than 3 and 3-3+ classes. This unexpected increase after the low biomass level in the less **impacted** classes than 3+ could be explained by the better riparian structures at the 3+ site, this better surrounding seems to mitigate some of the effects of the increased fine sediment load.

Even though a trend in the NMS analyses was visible it is possible that the siltation classes defined by the "Interreg Project Bayern-Österreich – Feststoffmanagement im Mühlviertel und im Bayerischen Wald" do not comply well with the analyses performed in this study and therefore may be the reason for the weak reaction of biomass and number of taxa at the lower, namely 2, 2-3 and 3, siltation classes.

The analysis of the percentage of EPT taxa (figure 19) shows only a very weak trend of decrease alongside the gradient of increasing siltation class.

The observation that there is only a very weak interdependency between the percentage of EPT taxa in the rivers and the siltation classes indicates that the methods used may not be of a sufficient resolution or specialised enough to comply with the defined siltation classes.

The functional feeding guild composition, given in figure 21, changes from the **reference** sites to the sites of highest **impact**. The clearest trend is the increase of predators, which according to Matoni and Kep (2010) is a sign that the community is severely disrupted. They suggest that the disrupted community may be easier to prey on and this is the reason why the predators thrive during the disturbance. Furthermore, the share of grazers decreases with increasing siltation. The suspended particles limit their food resources by eroding the substrate at fast flow on the one hand, and by covering the substrate surface at slow flow on the other, thus changing the stream metabolism (Matoni & Kep, 2010). Filter feeders are known to be prone to fine sediment disturbances (Rabení et al., 2005) but the share of filter feeders at the study sites only decreases at the two most **impacted** sites.

The general weak reaction of all investigations in this study to the increasing siltation classes could be due to the siltation classes that have been defined in Hauer et al. (2015), which focus strongly on the amount of granite grit that is present in the system. Therefore the siltation classes used in this study are not directly comparable to general increase of fine sediments because fine sediment comprises grain sizes from clay to sand fractions (0.004 mm – 2 mm) and the dominantly occurring granite grit in the study area comprises mostly grain sizes between 2 mm and 4 mm (Hauer et al., 2015). So only the effects of the coarsest fine sediment fractions are tackled in this study due to the absence of finer fractions.

5.3. MICROHABITAT-SPECIFIC COMMUNITY STRUCTURES

The NMS Analysis for the different microhabitats, given in figure 18, demonstrates that each site forms individual groups in the coarser fractions, whereas the akal fractions form one big cluster of samples. This indicates the importance of instream structures. Each site has a unique combination of patches and the coarser fractions offer the chance of small scale diversity, whereas the akal fractions act as a uniform substrate. Jones et al. (2007) state that fine sediments may increase the embeddedness of stones, thus reducing erosion during high-flow events and creating areas of low shear stress. However, as interstices fill with fine sediments, the ability of invertebrates to penetrate the river bed by crawling between larger particles becomes reduced, affecting invertebrate distribution (McClelland and Brusven, 1980) and the availability of refugia (Lancaster and Hildrew, 1993): these interstices (and other areas of low shear stress) are used by invertebrates as refugia to avoid ‘wash-out’ during high-flow events (Lancaster and Hildrew, 1993). The availability of flow refugia on the river bed has a significant influence on community composition, with motile species lacking where refugia are scarce (Gjerløv et al., 2003). Under such conditions, the invertebrate community becomes more vulnerable to physical disturbance of the bed during flood events, or any event that depletes the fauna such as pollution or low flow (Dunbar et al., 2010). Recolonization of denuded patches takes longer when motile species are lacking from the community (Gjerløv et al., 2003).

The analysis of number of taxa for the different microhabitats (figure 11) shows similar values for macro- to microlithal, independent of **impacted** or not, samples. The decrease of the number of taxa abruptly starts at the samples taken from akal and finer fractions. Number of taxa is a measure of organic pollution stress and general environmental degradation (Glendell et al., 2014) and indicates that with increasing siltation class and decreasing grain size stress for the benthic invertebrates increases.

The analysis of biomass for the different microhabitats (figure 15) shows that three levels of biomass occur. Macro- and mesolithal fractions have a similar biomass, being the classes with the highest biomass and the **impacted** macrolithal being the fraction with the overall highest biomass. The microlithal fractions only have half the biomass of macro- and mesolithal. And as expected the akal and psammal fractions are the ones with the lowest biomass.

The investigation of the share of EPT taxa for the different microhabitats showed interdependency. The observed trend, given in figure 20, corresponds with the existing knowledge synthesized by Wood & Armitage (1997) and Jones et al. (2007) that with increasing fine sediment load the amount of EPT taxa in an impacted reach will decrease.

It seems that the severe changes starting at the akal fractions correspond with the occurrence of the typical granite grit, which belongs to the akal fraction, in the study area.

5.4. Ecological status, saprobity and siltation

The calculation of the water framework directive compliant ecological quality class of the different sites, after Ofenböck et al. (2010) resulted in an overall good status of the five **reference** sites. From the seven **impacted** sites only two are in a moderate ecological status whereas the other five are in a good status. The two moderate sites are Kleine Mühl Plane Bed, siltation class 3 and Große Mühl Pool Riffle, siltation class 3-3+. With a range of siltation classes from **reference** to 3+ the first reaction of the water framework directive compliant method is at siltation class 3, beforehand class 3 there is no reaction to the increasing amount of fine sediments visible. To calculate the ecological quality class three elements are combined, the saprobic index, the module of acidification and the module of general degradation. The module of general degradation aims to reflect the effects of different stressors and is derived by the calculation of one or two multi metric indices (Ofenböck et al., 2010). In case of the study area in the Granit- und Gneisgebiet der Böhmischen Masse (Illies, 1978), catchment size class one and two, the values used for the multimetric indices in the general degradation module are: degradation index, RETI, number of taxa, number of EPT-taxa, %EPT-taxa, share of litoral zones, %Oligochaeta & Diptera and Margalefs diversity index (Ofenböck et al., 2010). None of these metrics are focus on fine sediment stress indicators, they can indicate degradation through fine sediment but cannot provide certainty that an observed impact is really the consequence of increasing fine sediments or other stressors. Another possible reason for the late reaction of the ecological quality class calculations could be the above discussed patchiness, where the overall impression of a site is silted but due to patches of structure the actual ecological quality class is higher than the siltation class suggests. On the other hand, the discrepancy between siltation and ecological quality class could be a result of the general nature of the ecological quality class approach, maybe the distinct impacts of siltation are too weak for the calculated ecological quality class to react to less severe siltation classes. “As a prerequisite for correct and successful biological sampling careful recording of the microhabitat composition is essential. The sampling itself must be done with the “multi-habitat method”, by distributing 20 sampling replicates according to the distribution of microhabitats” (AQEM Consortium, 2002). The sampling according to the distribution of microhabitats might be the reason for the late reaction of the ecological quality class. For example, when a stretch is silted to a proportion of 50 % (10 sampling units), the remaining 50 % (10 sampling units) of other microhabitats can compensate the poor diversities and abundances of the silted sampling units.

Regarding saprobity, all **reference** sites are in a good status with saprobic indices ranging between 1,63 and 1,93. Five of the **impacted** sites also have a good saprobic index. The Kleine Mühl- Pool Riffle, siltation class two, has a high saprobic index. The Kleine Mühl- Plain Bed, siltation class three, is the only site with a moderate saprobic index of 2,35. This higher saprobic index is probably a result of the missing riparian vegetation in an agricultural spatial context, shown in figure 6, which leads to increased fine sediment and nutrient input.

5.5. PROPORTION OF SEDIMENT SENSITIVE INVERTEBRATE INDEX

The calculation of the PSI for the different siltation classes shows similar results as the analyses of the number of taxa and biomass. The **reference** class, class 2 and class 2-3 have an average PSI that puts them in the best class of minimally sedimented/unsedimented. Classes 3, 3-3+ and 3+ belong to PSI class slightly sedimented. The PSI reacts moderately to the increasing siltation classes. The calculation of the PSI for the different microhabitats shows a better linkage between decreasing grain size and PSI, but still the reaction of the PSI is weaker than expected. The reason could be that the smallest grain size occurring in this study is the psammal fraction. Psammal has a grain size of 0,063 – 2 mm and marks the coarser end of the fine sediment range, but is commonly included in studies regarding the impacts of fine sediments on the lotic environment (Wood & Armitage, 1997). The granite grit that forms this psammal to akal fractions in the study area is typical for north-eastern Upper Austria and a consequence of the geological conditions (Fuchs & Thiele, 1987). The sensitivity rating to fine sediment for each taxa has been derived in a two-stage process involving firstly an extensive literature review, and secondly an assessment of anatomical, physiological and behavioural traits exhibited by individual taxa (Extence et al., 2011). Glendell et al. (2014) and Extence et al. (2013) proofed that the PSI works in the United Kingdom as well as in Guinea. The fact that that the finest fractions sampled in this study are psammal fractions may falsify the results of the PSI calculations since the sensitivity ratings where derived from the reactions of the individuals to not only psammal, but way finer fractions to. The weak reaction can be tracked back to grain size discrepancy between the silt fractions the PSI was designed for and the coarser granite grit that is typical for the study area. A general increase of the sensitivity ratings to a lower taxonomic level where increased information yield is foreseeable could favour the PSI. Furthermore it has to be noted that the exclusion of Chironomidae from the calculation of the PSI and generally not classifying them is a missed chance of adding sensitivity to the PSI. With over 5000 species worldwide and 700 in Europe, comprising species with very distinct habitat preferences like *Rheotanytarsus* sp. which solely occurs on hard substrate, or *Rheosmittia spinicornis, Odontomesa fulva* and *Prodiamesa olivacea* which according to Graf et al. (2016) have a clear preference for fine sediments thus having the potential to be a very good indicator for stress through siltation. The inclusion of Chironomidae could contribute to the improvement of the PSI.

5.6. Indicator Species Analysis and Sensitivity Ratings

As the previous analyses, the calculation of Indicator species has been performed once for the different siltation classes and once for the different microhabitats. Table 4 shows the results for the different siltation classes and the taxa with 3 highest levels of significance per siltation class are displayed. For the **reference** class no significant indicator species have been found, which is a result of the varying choriotopes occurring in the **reference** class. In class 2 there are two characteristic species, *Brachycentrus maculatus* and *Ancylus fluviatilis*. *Ancylus fluviatilis* indicates hard substrate and therefore does not occur in the more silted classes and *Brachycentrus maculatus* is typical for class 2 due to its mass occurrence at the KLM_FKV2 site. All three taxa in siltation class 2-3 are statistically significant (p value below 0.005) indicator taxa for siltation class 2-3. The taxa are *Habroleptoides confusa*, Psychodidae and the *Rhyacophila* sensu strictu group. *Habroleptoides confusa* is a gathering rhithral species that prefers microlithal with macrophytes. For the other siltation classes 3, 3-3+ and 3+ no significant taxa have been found.

The calculation of indicator species for the different microhabitats, given in table 5, shows a clearer picture. All significant taxa and the first insignificant taxa per microhabitat are displayed. Alongside the decreasing grain size gradient, for each microhabitat, from macrolithal to the **reference** samples of microlithal, significant species have been found. However, it is visible that with decreasing grain size the number of significant species decreases. Therefore, instead of finding indicator species for silted conditions, indicator species for good ecological conditions have been found. With decreasing grain size, a shift from a lithal fauna to a more generally tolerant faunal composition is visible.

Regarding the sensitivity ratings of the calculated indicator species the calculated indicator species for the **reference** class and siltation class 2 have sensitivity ratings, with each 2 taxa with an A –highly sensitive rating and one taxon with a B-moderately sensitive rating, as it was expected beforehand. In the higher siltation classes some peculiarities are observable. Psychodidae (dunkel) were calculated as a significant indicator species for siltation class 2-3 and have a sensitivity rating D-highly insensitive, all Psychodidae in the sensitivity rating database of Extence et al. (2011) have a D rating even though not all Psychodidae have the same habitat preferences.. In siltation class 3-3+, the second highest class, two taxa with a sensitivity rating B- moderately sensitive were calculated, namely *Oreodytes sanmarkii* and *Allogamus auricollis*. In the highest siltation class, 3+, *Ephemerella notata* was calculated as an indicator species even though it has a sensitivity rating B-moderately sensitive. Neither of these taxa have strong preference for psammopelal or argyllal habtitats (Schmidt-Kloiber & Hering, 2015), which would be considered as silted. The interdependency between increasing sensitivity rating of indicator species and decreasing grain size is stronger at the indicator species analysis for the different microhabitats. Only one oddity is observable. Similar to the indicator species analysis for the different siltation classes the Psychodidae (dunkel) are calculated as an indicator species for the macrolithal **reference** microhabitats, being the only taxon with a D- highly insensitive rating at the coarse microhabitats. This again could be a consequence of the general bad sensitivity rating for Psychodidae even though not all of them indicate bad conditions. The absence of taxa which really prefer or withstand heavily silted conditions seems to be a consequence of the lack of very fine sediments in the study area. The ratings are fitted to argyllal to psammal fractions, whereas in the study area the coarser granite grit, belonging to the akal fraction, is dominant as the finest fraction.

6. Conclusion

The erosion and deposition of fine sediment are intrinsic and natural components of the hydro-geomorphic processes of fluvial systems, but when the deposition of fine sediment increases, either due to natural or anthropogenic processes, changes in the aquatic system are inevitable. This study shows that increasing amounts of fine sediment lead to a decrease of species diversity, abundance and biomass in the benthic invertebrate community, therefore the first hypothesis, "Increasing amounts of fine sediment lead to a decrease of species diversity, abundance and biomass in the benthic invertebrate community", is confirmed. Contrary to the third hypothesis, "Increased sediment load in running waters causes a decrease of the ecological quality class", only two of the seven **impacted** sites are in a moderate condition. This indicates that the defined siltation classes which arose from the very specific circumstances, created by the presence of granite grit in the study area, do not comply well with the water framework directive conform methodology regarding the sensitivity towards siltation, therefore , the hypothesis has partly to be declined. Regarding the proportion of sediment sensitive invertebrate index, the index has proven to be applicable, therefore the fourth hypothesis, "The PSI is applicable in the study area", is confirmed, but with some limitations. Its weak reaction, similar to the assessment of the ecological quality class, indicates room for improvement and research regarding the level of detail in the fine sediment sensitivity ratings, especially the exclusion of Chironomidae and the weak differentiation between different types of Psychodidae. It has to be noted that the weak reaction could be tracked back to the discrepancy between the grain sizes the PSI was created for and the coarser grain size of the study areas typical granite grit. Silt, sand and granite grit do not only have distinct grain sizes but also distinct effects on the lotic environment. An inclusion of the PSI as a part of the calculation of the multi-metric indices into the module of general degradation in the detailed benthic invertebrate method (Ofenböck et al., 2010) could provide the opportunity to take account of fine sediments as a distinct stressor in the standard evaluation method. Furthermore, regarding the second hypothesis, "Specific benthic invertebrate taxa prefer silted conditions and can be used as indicator species for siltation processes", it has been found out that there are no indicator species that prefer silted conditions, the results of the indicator species analyses indicate in particular that there are indicator species for undisturbed conditions rather than silt indicators, therefore the second hypothesis has to be declined. The absence of indicators for unsilted conditions can be used as an indicator for stress. The problem is that only stress in general is indicated and the absence of indicator species for undisturbed conditions does not automatically imply that the therefore present stress is due to the impact of increasing fine sediment load.

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