



Impacts of riparian zone alternation by hydropower on biodiversity

A comparative analysis of faunistic diversity and environmental factors across various modification levels in riparian zones

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Impacts of riparian zone alternation on biodiversity

A comparative analysis of faunistic diversity and environmental factors across various modification levels in riparian zones

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Keywords: Riparian zones, biodiversity, river regulation, hydropower, restoration

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Abstract

The loss of biodiversity and natural ecosystems and the freshwater crisis are among the most critical global challenges of our time. Hydropower dams cause fragmentation of river systems and have major impacts on aquatic and terrestrial vegetation and fauna. During dam construction, rivers were dredged and sand and sediment were often deposited along the banks, creating new or modified riparian zones. These modified sites exist in large numbers to date and there is a lack of research to understand the dynamics and possible restoration potential of these habitats. The aim of this study is to assess the faunistic biodiversity of riparian zones affected by dredging. For this, several biodiversity measurements were carried out using camera trapping, bioacoustics, insect trapping and eDNA soil sampling. In addition, environmental factors measured using drone surveys and satellite imagery were used to identify variation between sites and their impact on biodiversity, and to provide information on the ecological restoration potential of modified sites. The studied highly modified riparian zone close to the Gallejaur hydropower plant showed the lowest overall diversity, while the moderately modified riparian zone close to the Rusfors power plant showed a diverse species community, indicating the ecological potential of the modified site and highlights the importance of surrounding areas. The natural site along the free-flowing Vindel River (Vormfors) showed the highest overall faunistic diversity. The environmental factors suggest that increased vertical heterogeneity, greater variation in annual flow, higher NDVI values, larger (mean) vegetation and less steep slopes (within the first 10 metres from the river) can be used to distinguish natural and modified riparian areas and are likely to be associated with differences in biodiversity.

Keywords: Riparian zones, biodiversity, river regulation, hydropower, restoration

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Abbreviations

ARUs	Autonomous recording units
ASV	Amplicon sequence variant
CCA	Canonical Correspondence Analysis
CH ₄	Methane
CO ₂	Carbon dioxide
D	Simpson index (in the study, I used: 1/D)
DEM	Digital elevation model
DSM	Digital surface model
eDNA	Environmental DNA
EH	Environmental heterogeneity
GJ	Gallejaur (site)
H'	Shannon-Index
Km / km ²	Kilometre / Square kilometre
L	Liter
M / m ²	Meter / Square meter
NDVI	Normalized Difference Vegetation Index
NIR	Near infra-red band
NMDS	Non-metric Multidimensional Scaling (ordination plot)
PAM	Passive acoustic monitoring
R	Red band
RF	Rusfors (site)
SD	Standard deviation
SMHI	Sveriges meteorologiska och hydrologiska institute
VF	Vormfors (site)

1. Introduction

The biodiversity and water crises are two of the most important global challenges of our time. There have been numerous studies on biodiversity in regions with dams and hydropower sites, many of which have focused on aquatic habitats or riparian vegetation. The aim of this study is to investigate how the dredging of rivers for dam construction affects faunistic biodiversity in the riparian zone. Two modified riparian zones, created by the construction of hydroelectric power plants through dredging of the riverbed in the 1960s, were selected as study sites. Furthermore, a protected area of a non-modified and restored free-flowing river was studied as a reference site. For this purpose, the terrestrial faunistic biodiversity of the habitats was estimated in order to provide information on the ecological potential of the modified sites. I used a variety of ecological methods and models to develop a picture of the current state of terrestrial fauna, such as camera traps, ultrasound audio recorder, insect traps and eDNA soil sampling combined with measurements of environmental factors and heterogeneity, such as topography (slope), vertical structure (vegetation height), horizontal structure (NDVI) and annual variation in flow.

1.1 Riparian zones

Riparian zones are diverse and complex ecosystems (Naiman et al., 1993; Naiman and Décamps, 1997; Nilsson et al., 2012). They serve as interfaces between the water and the surrounding landscape and are therefore important for both, terrestrial and the aquatic ecosystems. Riparian zones are defined as the part of terrestrial landscape, from high water mark to “uplands where vegetation may be influenced by elevated water tables or flooding, and by the ability of soils to hold water” and include the stream channel (Naiman et al. 1993). Therefore, all zones of the riparian areas are affected and at least temporally in direct contact with sessional flooding.

Consistent with the intermediate disturbance hypothesis (Connell, 1978), riparian zones provide a heterogeneous environment with temporal variation due to periodic flooding, resulting in diverse and complex habitats with high species richness (Naiman et al., 1993; Nilsson & Berggren, 2000). Due to the connection between

the channel and the terrestrial system, also the food web and abiotic factors are binding these two ecosystems (Standfort et al., 2005).

In particular, riparian vegetation performs a number of important key functions within the river system (Gregory et al., 1991; Naiman & Décamps, 1997; Standfort et al., 2005). Riparian vegetation provides shade over the river, thereby influencing light and temperature in the water. In addition, the decomposition of dead plant parts is a source of nutrients for both aquatic and terrestrial ecosystems. Growing vegetation directly influences water flow (Naiman et al., 1993). Riparian zones provide nutrient fluxes and function as "filters" between the river system and the adjacent land area (Naiman & Décamps, 1997, Nilsson & Berggren, 2000). Riparian zones have been shown to play an important role in maintaining regional biodiversity through their regulating effect on surrounding ecosystems and provision of important habitats (Naiman et al., 1993).

Typically, riparian zones host different groups of organisms such as large herbivores like deer (Brookshire et al., 2002) and keystone species such as the Eurasian beaver (Naiman et al., 1994; Nazarov et al., 2023). Studies have also shown the importance of riparian zones for bird biodiversity in Sweden (Darveau et al., 1995, Larue et al., 1994; Lind et al., 2019) and for *Carabidae* and *Staphylinidae* beetles (Andersen & Hanssen, 2003). Aquatic insect orders such as *Ephemeroptera*, *Plecoptera* and *Trichoptera* are found in rivers systems, are used as indicator species for environmental change in Sweden (Sandin et al., 2014, Tszedel et al., 2024), can be also found in their adult stage in the terrestrial riparian zones.

The width of riparian zones has been shown to have a major influence on ecosystem function in these systems. The width of a natural riparian zone depends on the size of the river, its location within the river basin system and local physical conditions (Naiman et al., 1993). For large rivers, it is typical for riparian zones to be large and to have a diverse flooding system, with parts regularly flooded and even forming temporary lakes (Naiman et al., 1993). Lind et al. (2019) have shown that the width of riparian zones has a significant effect on plant and bird biodiversity. They conclude that a 30 m wide buffer zone is beneficial for increasing floral diversity, while birds need wider buffers around 144 m in Swedish river systems (Lind et al., 2019).

Typically, riparian zones consist of different zones: regularly flooded areas covered with graminoids and amphibious plants, transition zones with shrubs and ending in riparian forests with moss covered grounds (Nilsson & Berggren, 2000; Nilsson et al., 2012) (see Figure 1). In typical Swedish riparian zones, the middle section of

the transition zone has the highest floral species richness and riparian forests are dominated by conifers (Nilsson et al., 2012), likely due to forestry practices.

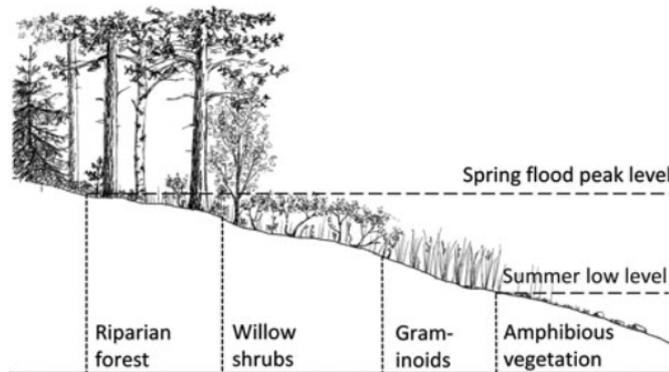


Figure 1. Vertical zones of riparian areas with water levels in low and high flood periods. Riparian zones are directly affected by seasonal floodings. Illustration from Nilsson et al., 2012

1.2 Effects from river regulation and dam construction on riparian zones

Sweden has a long history of river regulation. Since the 19th century, rivers have been modified to make it easier for the forestry industry to float timber. Stones and woody debris were removed, secondary channels were blocked and remained dry and rivers were channelized, dredged and widened. All these modifications have had a major negative impact on riparian areas (Helfield et al., 2007). Today, Sweden's major rivers are used to produce green energy, and hydropower plants are one of the largest sources of energy in Sweden. In 1994, Dynesius and Nilsson showed that 77% of rivers in the northern hemisphere are heavily to moderately affected by river regulation and the construction of dams and reservoirs, and only 12% of all large river systems in Europe are unaffected by human regulation (Nilsson et al., 2005). In Sweden alone, there are now more than 1800 hydropower plants, which together account for a difference of 19% from the natural distribution of the country's total annual discharge (Arheimer & Lindström, 2014). In short, hydropower is a great opportunity to produce green energy. In addition to all the benefits of hydropower, its construction has caused severe changes in river systems, which have a direct impact on ecosystems and biodiversity on a large scale. It is therefore important to monitor and understand the effects of these major impacts. Several studies have investigated the effects of river regulation on aquatic biodiversity and riparian vegetation (e.g. Belmar et al., 2013; Jansson et al., 2000; Nilsson & Berggren, 2000; Nilsson et al., 2012).

In general, the construction of dams and reservoirs is followed by a chain reaction of shifting ecological processes. Dams act directly as barriers, leading to fragmentation of the river. Migratory species that seasonally migrate downstream to their spawning grounds can be severely affected (Ammar et al., 2020; Nilsson & Bergren, 2000; Rendardy et al., 2021). If fish cannot pass these barriers due to the loss of connectivity, the result will be a reduced fish population. Studies have shown that not only the restriction of migration by dams, but also the risk of injury leads to high mortality rates and reduced fitness in the European eel population (Ammar et al., 2020). Other studies have investigated the negative impact of hydropower turbines on Atlantic salmon populations, due to the high mortality of smolts caused by turbines (Rendardy et al., 2021). Also, it is likely that the predation risk for fish is higher close to the turbines. In addition, reservoirs and dams prevent nutrients and deadwood from being distributed in the downstream river system. The connectivity of rivers also plays an important role in the long-distance dispersal of plant seed and can be restricted by barriers (Jansson et al., 2000; Jansson et al., 2005, Johansson et al., 1996). However, other studies have shown that stable flood management has allowed alien species to establish in riparian zones, such as non-native aquatic plants in South Africa or tree species in Nord America (Nilsson & Bergren, 2000).

The effects of hydropower plant construction on riparian zones have been explored in previous literature. Such effects can be divided into upstream effects (Effects around the reservoir) and downstream effects (Change in flow regime).

Upstream effects - effect around reservoir

The construction of reservoirs that result in immense amounts of stored water and long-term flooding of the surrounding riparian vegetation upstream (Nilsson & Bergren, 2000; Nilsson et al., 2005). This can lead to either river widening or lake enlargement. Shortly after the construction of the reservoir, plant mortality is high in this area, followed by loss of both habitat and reservoirs that absorb atmospheric carbon dioxide (CO₂). In addition, decomposition of the vegetation leads to the production of CO₂ and methane (CH₄) and takes time depending on the size of the flooded area (Nilsson & Bergren, 2000; St. Louis et al., 2000). Both processes contribute to global warming through the release of greenhouse gases, especially when areas that store large amounts of CO₂, such as peatlands, are flooded (St. Louis et al., 2000). In addition, the decomposition process produces the organic molecule methylmercury, which accumulates in the food chain and can remain in reservoirs for up to 20-30 years and could also get detected downstream (Rosenberg et al., 1997). In addition, nutrients stored in the soil are leached into the reservoir, and nitrogen and phosphorus in particular are known to cause problems in aquatic systems through increased algal blooms, which can lead to eutrophication of aquatic

systems and dead zones (Nilsson & Bergren, 2000). In addition, the newly created shoreline of the reservoir, which was originally more distant from the water level, forms the riparian zone after flooding, and studies have shown that these areas have reduced floral biodiversity compared to free-flowing rivers (Nilsson & Bergren, 2000; Nilsson et al., 1997).

Downstream effects - Change in flood regime

The construction of dams leads to a shift of flood regime from upstream to downstream, with major impacts on the downstream hydrology of rivers. Due to the controlled release of water, periodic flooding and regular flood plains are reduced or not existing. Natural system adapted to these circumstances and the period flooding is, like stated above, one of the main drivers of the high diversity of riparian zones. When these floodings are reduced, the seasonal succession of vegetation is prohibited and the riparian zones get to a stable stage, which leads to a reduced biodiversity of vegetation. Studies could show a change in the riparian vegetative communities after the reduced flood plains due to modifications (Decamps et al., 1988) or an increase of riparian vegetation after the construction of hydropower dams, which are nevertheless less diverse (Aguilar et al., 2016). A study by Belmar et al. (2013) showed that with increasing degree of river alternation, the complexity of riparian vegetation reduces, leading to a more uniform riparian vegetation. Former studies show that river regulations often negatively affect riparian vegetation in sense of reduced species richness and cover (Johnson, 2002; Nilsson et al., 1991).

The change in the flood regime is not only detectable for riparian vegetation, but animals are also affected by these changes. Studies could show a change in behaviour and reduced reproduction due to the change in flood regime, for example for amphibian, which using ponds for laying their eggs and might not existing when there are no high flood events (Attwell, 1970).

The storage and controlled release of water also causes lowering in the ground water level which effects the surrounding vegetation largely as well (Nilsson & Berggren, 2000). Due to the barriers to dams, sediment get trapped in the reservoirs and get prevented from traveling downstream (Nilsson & Berggren, 2000).

1.3 Dredging of riverbeds and environmental heterogeneity

Effects from dredging

There is widespread interest in river sediments for the construction industry, and studies have shown how sediment and gravel extraction from riverbeds affects hydrology (e.g. mean flood depth; velocity), groundwater levels, and causes river changes due to increased erosion in the riverbed and bank (Calle et al., 2017; Islam et al., 2018; Rovira et al., 2005).

However, no study investigated the effect of sediment placement from dredged riverbeds (due to hydropower construction) in riparian zones. During the construction of the hydropower plants in my study areas during the 1960s, sand was dredged from the riverbed downstream of the construction site to increase the head, improve efficiency and reduce bank erosion at the outlet. It is likely that this led to a redistribution of the river sediment and therefore the riverbed ecosystem. The removed sandy sediment was placed in the riparian zones, creating a new riparian zone where there used to be water, or reshaping the existing riparian zones. As a result of these changes, the riparian vegetation has been severely disturbed and is likely to end up in a more homogeneous vegetation. As a result of the dredging of the river and the subsequent removal of sandy soils, the riparian zones in the study areas are expected to have a less diverse structure than before the river modification and the typical zonation of the riparian zones into regularly flooded, transitional and forest zones may be disturbed or no longer exist. It is debatable whether the greater distance between water level and vegetation and the reduced flooding means that these areas can still be classified as riparian zones or whether they are more likely to be upland forests. In addition, the dredging of riverbeds results in the replacement of the nutrient-rich soil typical of riparian habitats with a sandy soil type. This is associated with changes in available nutrients and soil moisture. Reduced biodiversity is expected to be one of the consequences. With this study I want to investigate the biodiversity of these specific sand-filled areas.

Environmental heterogeneity and species diversity

The theory that a more heterogeneous habitat leads to a more diverse species community is a commonly used and well-studied ecological assumption (Renner et al., 2017; Ricklefs, 1977; Stein et al., 2014; Stirnemann et al., 2015). It is based on the assumption that greater habitat heterogeneity leads to a greater diversity of ecological niches, thus creating more space for the coexistence of species and greater species richness (Heidrich et al., 2020; Stein et al., 2014). Recently, this

relationship between environmental heterogeneity (EH) and biodiversity has been found to be more complex than a general positive trend due to the effect of stochastic events in different habitats (Heidrich et al., 2020; Stein et al., 2014). Nevertheless, Heidrich et al. (2020) showed a positive effect of vertical and horizontal EH on a variety of different species groups in temperate forests and a negative effect of increasing topographic EH.

1.4 Research question and hypothesis

In this study I investigated the differences in faunistic biodiversity and environmental factors of riparian zones, to find the drivers of these differences and to discuss ideas for how the sites could be improved to increase their ecological value. To do this, I studied three riparian sites during the summer of 2023 (data collection: June - August). Two of the sites had been modified, and I focused on the area where sand from river dredging had been used to create 'new' riparian zones and one site in a free-flowing river with partially restored riparian zones was used as a positive control. All three study sites are part of different large river systems in northern Sweden. Biodiversity data such as species richness, total abundance, presence of indicator species and biodiversity indicators (Shannon- and Simpson-Index) from different ecological field methods (insect traps, camera traps, audio recorder and eDNA soil sampling) were used to differentiate the diversity between sites. Additionally, measures of environmental factors were used to estimate EH. In this study, environmental factors such as Normalised Difference Vegetation Index (NDVI), slope, tree height and monthly streamflow were measured using drones, digital surface models and publicly available streamflow data. To estimate EH, I compared the variability of these factors under the assumption that greater variability (in standard deviation (SD)) provides a more heterogeneous habitat, leading to higher biodiversity (Ricklefs, 1977, Renner et al., 2017, Stirnemann et al., 2015, Stein et al., 2014).

Therefore, I tested the following hypothesis:

Table 1. Overview assumption, research question, hypothesis and null hypothesis for this study

	<i>Assumption</i>	<i>Research question</i>	<i>Hypothesis</i>	<i>Null hypothesis</i>
1)	Modified sites have undergone environmental alterations affecting EH	Is there a difference in EH and productivity between modified sites (GJ, RF) and natural riparian area (VF)? Is there	Modified sites (GJ and RF) have a reduced EH (SD NDVI, SD vegetation height, SD of flow) along with lower vegetation productivity (mean	There is no difference in EH and NDVI between modified and free-flowing river sites.

	and productivity.	a gradient in level of modification and EH?	NDVI) compared to natural site (VF), but a higher variability in topographic EH (SD slope). Expected gradient: VF – RF - GJ	
2)	Modified habitats have a reduced biodiversity.	Does the non-modified site (VF) have a higher biodiversity than modified (RF) and highly modified area (GJ)?	VF, as non-modified site, has the highest biodiversity of fauna , followed by RF and is the lowest in GJ.	There is no difference in biodiversity between the sites.
3)	Natural riparian zones have a higher amount of indicator species, such as <i>Carabidae</i> , <i>Staphylinidae</i> (Andersen & Hanssen, 2003) and <i>Trichoptera</i> , <i>Plecoptera</i> and <i>Ephemeroptera</i> (Sandin et al., 2014, Tszedel et al., 2024).	How does the modification status of riparian zones affect the presence of indicator species?	The natural VF site, will have a higher detection of indicator species such as <i>Carabidae</i> and <i>Staphylinidae</i> beetles as well as <i>Trichoptera</i> , <i>Plecoptera</i> and <i>Ephemeroptera</i> compared to modified zones (RF and GJ).	Riparian zone modification does not influence the presence of indicator species.
4)	Greater EH leads to more diverse habitats and therefore to an increased biodiversity.	Does EH influence biodiversity in the investigated riparian zones?	Sites with higher EH (SD NDVI, SD vegetation height, SD of flow) exhibit greater biodiversity . A higher variability in topography (SD slope) decreases biodiversity.	EH has no impact on biodiversity in the investigated riparian zones.

2. Methods

2.1 Study sites

The study was carried out on the riparian zones of three rivers in northern Sweden, the Skellefte River (site: Gallejaur - GJ), Ume River (site: Rusfors - RF), and Vindel River (site: Vormfors - VF), two of which are modified by dam construction. These locations are shown on a map of Sweden in Figure 2.

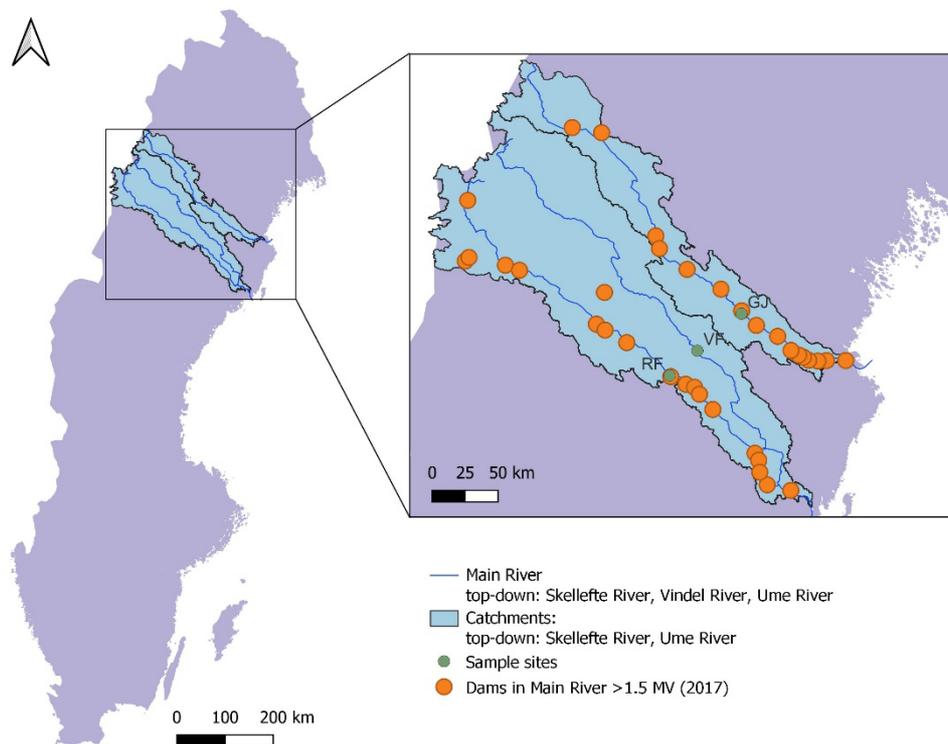


Figure 2. Map showing the locations of catchments and main river of study sites: Ume River, Vindel River and Skellefte River. Including locations of large dams (>1.5 MV) illustrated with orange dots and locations of study sites (GJ = Gallejaur, VF = Vormfors, RF = Rusfors).

The riverbeds of the two modified sites (GJ and RF) were straightened and dredged during the construction of hydropower plants and dams. The dredged, sandy

material was transported to the river sites and used to partially cover and extend the natural riparian zone on the river banks or to create an artificial island in the middle of the river at one of the sites (GJ). These river modifications took place in the 1960s, when the hydroelectric plants were built. To find suitable sites, I compared orthophotos from 1960 and 1975 to see where new habitat had been created. I marked the areas where I could visually see that sand was being transported from the riverbed to the riparian zone and used these areas as study sites (see Figure 3). Moreover, I compared the soil types of the areas based on a map (see Appendix - Figure 1) from *Lantmäteriet* (accessed in March 2024).

The orthophotos from around 1960 (taken between 1949 and 1970) were taken at a resolution of 0.5 m or 1.0 m in black and white and are open source data from *Lantmäteriet*. Current orthophotos with a resolution of 0.5 m, taken in 2018, and land use data are also available on *Lantmäteriet*. Orthophotos from 1975 were available on *kartbild.com*, provided by *Lantmäteriet*. The open-source software qGIS 3.22.16 (QGIS Development Team, 2023) was used to create the maps and process the geo data. The 1975 orthophotos were screenshotted and georeferenced using the georeferencing toolbar in qGIS, using both the dam and characteristic parts of the land surface as reference points (Aguiar et al., 2016). All photographs and other GIS data used in the project were transformed to the commonly used Swedish reference system (*SWEREF99*).

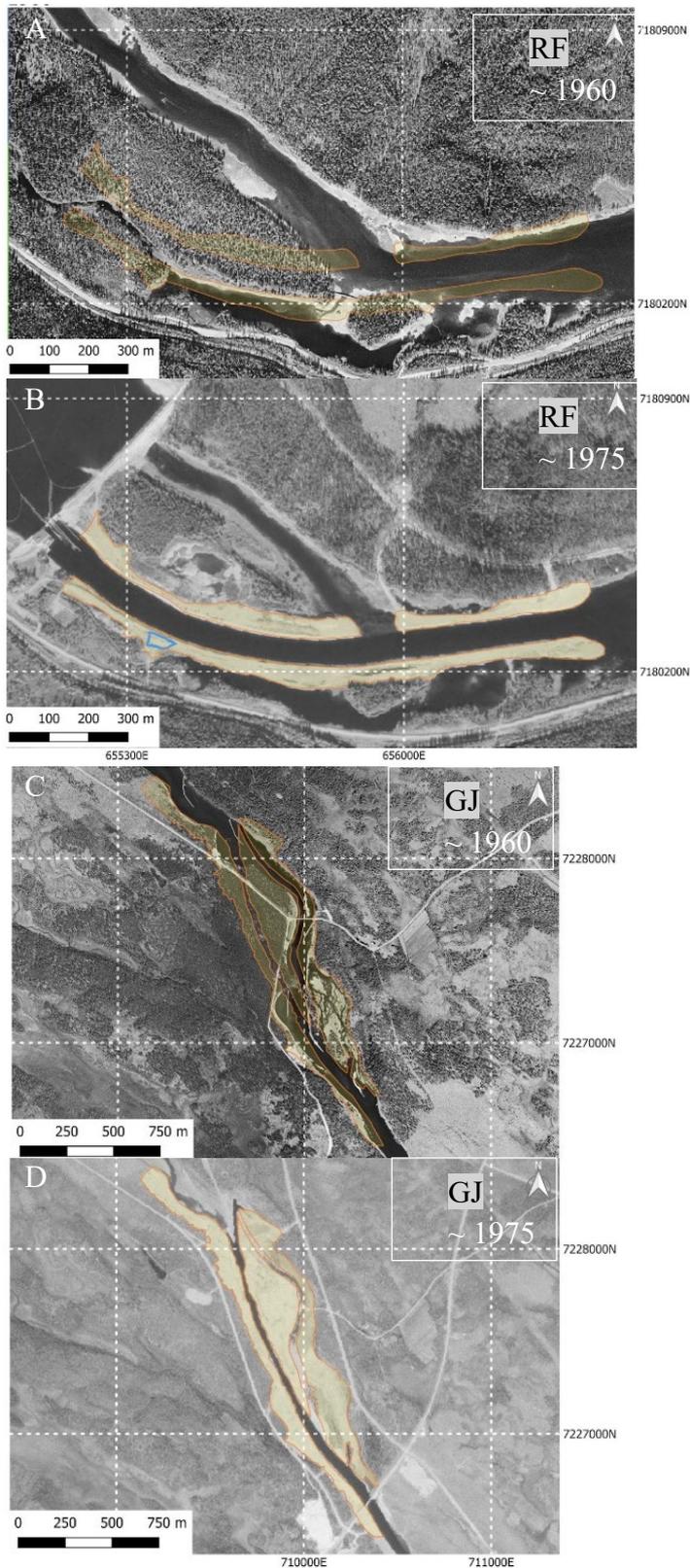


Figure 3. A: Rusfors hydropower station (RF) site before dam construction around 1960; B: RF after dam construction around 1975; C: Gallejaur hydropower plant outlet (GJ) before dam construction circa 1960; D: GJ after modification and construction around 1975. Yellow lined polygons indicating sand deposits in the riparian zone before and after the modification. RF site = 0.,158 km². GJ site = 0.479 km².

One study site was located in the Skellefte River, 2 km downstream of the Gallejaur hydropower plant, and includes the plant's outlet (GJ). The Gallejaur reservoir and the modified riverbank area were developed in 1964 and have an electrical capacity of 220 MWe with a total head of 80 m (*Vattenfall.com*). The elevation of the study site is about 276 m above sea level. Both plants are built with large storage reservoirs. The total area of the area is 0.479 km² and the modification of the riparian area is very large. Therefore, this site will get described as heavily modified in this study.

The other modified was located close to the water outlet of Rusfors hydropower plant (RF). This plant is located in the Ume River, about 30 km upstream from Lycksele. Construction began in 1962 and today the plant has an output of 41 MWe with a head of 12 m (*Vattenfall.com*). The approximate altitude of the site is 334 m above sea level. The total area of filled sediment is 0.158 km². Also, the dam is smaller, then the heavily modified site GJ and while the soil on the GJ site on the map from *Lantmäteriet* is even categorized as “filling”, in the RF site the filled areas are marked as “glacial sand” soil at the RF site (see Appendix - Figure 1). This could be, because the amount of sediment was less, but it is nevertheless visible on the historical orthophotos. The filled riparian areas were to large parts constantly connected to the surrounding forest after the construction. Moreover, the site is near to a small meander, which is unconnected to the main river and is likely to have a positive ecological impact. The RF site is close by the nature reserves “Stor-Skorvliiden” and “Lill-Skorvliiden” (see Appendix - Figure 2). Therefore, this site will be classified as modified in this study.

To compare the natural biodiversity of a free-flowing river riparian zone, I chose the Vormfors rapid in the Vindel River (VF). In the 1960s there were plans to build hydroelectric power stations in the Vindel River. However, for reasons of nature conservation, the Vindel River remained untouched by hydroelectric power stations. But the river has not been completely untouched, as the river was shaped by floating timber around the 19th century (Helfield et al., 2007). Stones were removed and the river was widened and straightened. In recent years, a lot of restoration work has been done on the Vindel River, especially the riparian zones (*Lansstyrelsen, Västerbotten website*). To restoration work within the Vindel River, stones, gravel, large wood debris were placed, along with dam removal, creation of fish paths and riparian vegetation management (*Vindel River LIFE project*).

The Vormfors rapids are over 9 m high, 500 m long and about 315 m above sea level. For comparison, Gallejaur is 267 metres above sea level and Rusfors 334 m, so Vormfors is quite similar and a good place to compare as it is a natural fall and many of the other natural rapids in Vindel River are at lower elevations. Moreover, VF is a nature reserve and a protected area. Endangered lichen species can be found

here. The area also has a rich riparian forest with large amounts of dead wood (*Lansstyrelsen*). Also, the site is easy to reach and not directly disturbed to a highway like many other suitable sites in Vindel River. As a study area, we marked an area with similar size and width to the study areas in RF and GJ, including parts of the protected area and parts upstream of it (see Figure 4) so that the total area of the study area in VF is 0.158 km². Due the absence of hydropower, performed restoration work and the free-flowing river dynamic, this site will be described as non-modified in this study.

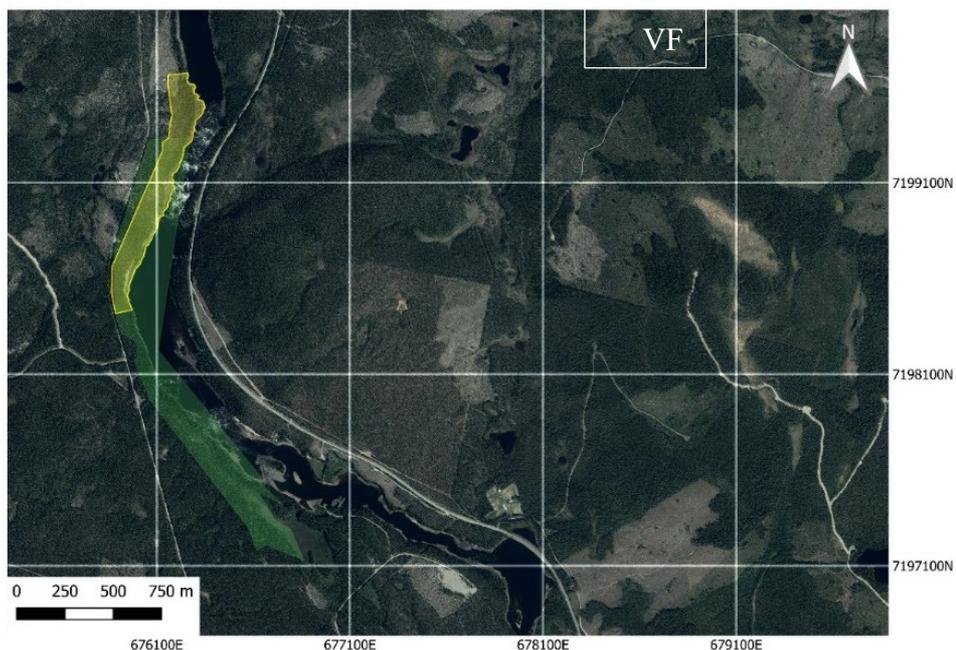


Figure 4. Vormfors (VF) site at the Vindel River with a current orthophoto; Green polygon: Protected area - Nature reserve Vormforsen; Yellow polygon: defined study site (0.158 km²)

To effectively monitor terrestrial biodiversity across the entire sites, I conducted field sampling at three randomly selected locations within each, resulting in three sub-sampling plots per site. This strategy ensured efficient fieldwork while maintaining a comprehensive representation of each area. The locations of these sub-sites were determined from the previously marked study areas using the QGIS Random Points in Polygons tool close to the rivers (see Appendix - Figure 3). All traps were placed within a 20 m radius of these randomly generated points.

2.2 Overview of methods

To assess the biodiversity of insects, bats and mammals, I used three pitfall traps, one window trap (IBL type-II) and one camera trap at each sub-site. Due to the

limited availability of audio recorders, with only six in total, I only placed them at two of the three subsites in each area. The insect traps at the VF site had to be concentrated in a smaller area because it was not allowed to collect insects inside the nature reserve (see Figure 4). I also collected a soil sample from each subsite for eDNA analysis. Further details of each method are given in the following paragraphs. I also measured environmental factors with open-source data, drone surveys and digital surface models. An overview of the scale (temporal and spatial) from each method is given in Table 2. Moreover, a table with the time schedule and exact days of field work is attached in the Appendix (see Appendix - Table 7). The exact location of each field method is visualised in the Appendix (see Appendix - Figure 4).

In summary, I want to investigate and connect the biodiversity with EH factors in the three riparian zones.

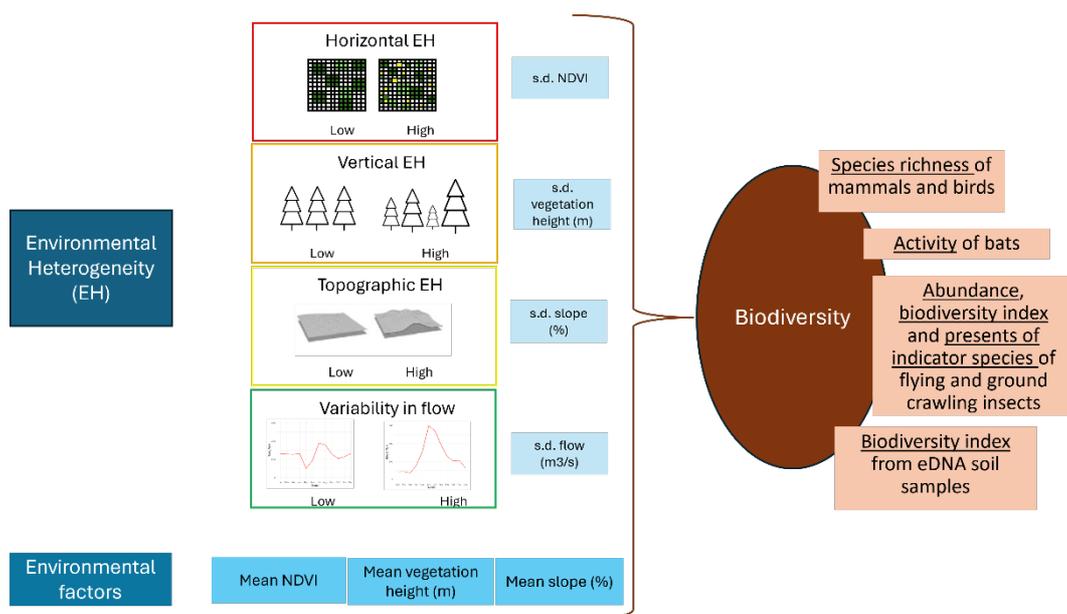


Figure 5. Flowchart overview for combined methods: Environmental heterogeneity (EH) here used with variability in horizontal vegetation, vertical vegetation, topography and flow combined with Environmental factors to determine drivers in differences in biodiversity along the sites.

Table 2. Overview over field methods and measured scale

Method	Variable	Measured scale	
		Time	Space
NDVI	SD, mean	Single measurement	5 places within site (radius ca. 30 m)
Vegetation height	SD, mean	Single measurement	5 places within site (radius ca. 30 m, close to river)
Slope	SD, mean	Single measurement	Entire sites (10 m)
Flow data	SD	Monthly measurement from SMHI (2022)	One per site (outlet point)
Camera trap	Species Richness	52, 48, 45 days	1 camera per subsite (3 per site)
Audio recorder	Total abundance	14 days	2 recorder per site (1 per most distant subsite)
Insect trap	Species Richness, Total abundance	17, 13, 10 days	3 pitfall traps per subsite, 1 IBL trap per subsite (9 pitfall, 3 IBL per site)
eDNA (soil sampling)	diversity indices	Single measurement	1 per subsampling site (pooled from 4 places around) (3 per site)

2.3 Vegetation analysis

To determine the vegetation structure, I used a combination of drone data, available elevation data and field observations.

I determined the environmental heterogeneity (EH) according to Stein and Kreft (2005) and Heidrich et al. (2020) with some adaptations. I used a selection of facets of EH: horizontal EH (here: SD NDVI), vertical EH (SD vegetation height) and microscale topography (SD slope). As the study areas are riparian zones, which are known to be affected by the flood regime, I also included variation in annual flow (SD flow) as measurement for EH. Although the literature suggests using gap edge length as a measure of horizontal EH, because our plots are much smaller, I decided to use NDVI as better suited measurement for this facet of EH.

To further identify the quality of the areas, I also investigated the mean NDVI as index for productivity, the mean slope and the maximum height of vegetation.

Drone flight settings and data

To determine NDVI and vegetation height, I flew the drone over 5 randomly assigned squares over each study area and placed them close to the river. To get a true random distribution on these, I used the qGIS function Vector > Research tools > Random points in polygons and set the number to 5 before going to the field. On site, I set up 30x30m squares around these random generated points for the drone. I used a DJI Phantom 4 multi-spectral drone in the field. I had previously trained to obtain an official licence from the EASA (European Union Aviation Safety Agency) in the open subcategory A1/A3 to fly a drone. I used a tablet connected to the drone to set up the flight using the *DJI Fly* app. I chose a high overlap (85%) to get a good resolution for the following analysis and a flight altitude of 50m above ground. I flew the drone on two consecutive days (19 and 20 July 2023) with similar weather conditions to have the same initial conditions, which is especially important for the NDVI calculation.

I processed the data using *pix4Dmapper* software to align the photos and GPS information. Again, I first had to set the coordinate system to SWEREF99 for further processing in qGIS. The result was a merged digital surface model (DSM) of each plot, which I used for the vertical EH (vegetation height), and a merged ortho mosaic “.tif” file for each spectral band (red, near infrared, blue, green), which I used for the horizontal EH (NDVI). For further calculation of the horizontal and vertical vegetation structure I used qGIS. I cut off the edges of each file to avoid fuzzy data, as the edges may contain inaccurate information due to a reduced overlay of the captured images.

Elevation data was available from Lantmäteriet.se. For the analysis I used the digital elevation model (DEM) from the National Elevation Model in a 2+ grid to get the highest resolution available. The data is from 2019 (file name: Höjddata, Grid 2+ 2019 CLIP (tif)). The DEM was used not only for the topographic EH, but also to obtain the vegetation height.

NDVI

The Normalized Difference Vegetation Index (NDVI) is calculated using the following formula: $NDVI = (NIR - R) / (NIR + R)$. Where NIR is the near infrared band and R is the red band of the light spectrum. The NDVI index is used for vegetation productivity (Assmann et al., 2019) and can be used for horizontal EH. The NDVI ranges between -1 to +1, whereby a high value indicates a higher

productivity of vegetation and a value around 0 indicates areas with no vegetation (like sand, rocks, or water bodies).

I applied the NDVI formular in the “Raster Calculation” function in qGIS, where I used the two raster files (one for NIR, one for R) generated for each plot to create a raster layer with the NDVI information. I performed further steps in R using the R packages “raster” and “tiff” to calculate the mean and SD from the NDVI files.

Vegetation height

To determine the height of the vegetation, I used the DSM from the drone flights, a total of 5 per site. As this data also included the general ground elevation (above sea level), I had to subtract the elevation from the generated DSM files. To do this, I first had to cut the DEMs from the National Elevation Model into the same shape as the drone generated DSMs. This resulted in a reduced number of pixels, as the DEM model is created on a 2m grid. I then used the “Raster Calculation” function to subtract them from each other. This gave me files with vegetation height information, and I used R to calculate the SD and maximum hight from the vegetation height of each plot.

Slope

To get information about the slope I used the DEM with 2m grid. As I wanted to determine the slope along a gradient away from the river, I created zones of 10m width away from the river until 100m or the end of the modified site (=area of dredged sediment) was reached, resulting in 10 zones away from the river in each location. I then cut the DEM into each of the zones and afterwards used the raster analysis function “slope” and set the output in percent to get information about the slope of each zone. I then used the “Zonal Statistics” option to create a file containing the mean and SD of the slope in each zone. For further analysis, I decided to use only the values from the first and second zones (0-10m from the river) as predictor variables for the biodiversity analysis. I used the "Zonal statics" file in R to create plots and for further analysis.

Flow data

I downloaded streamflow data from the *Sveriges meteorologiska och hydrologiska institute* (SMHI) vattenweb platform and used the S-HYPE model information. They have monitoring stations for streamflow (and other) data all over Sweden, and data was available in all three of my study sites. I chose to use the monthly corrected measured discharge (=stationskorrigerad vattenföring in m³/s) from one year (2022) to reduce the amount of data. I loaded this file into R to create plots and calculate

the SD of the annual flow. The mean flow is not taken into account within this, since the mean is depending on a lot of the size, shape, water depth and sediment of the river and is therefore not useful to compare in this study.

2.4 Camera traps

Camera traps were installed at the RF site from 12 June to 3 August 2023, at the GJ site from 16 June to 3 August 2023, and within the VF nature reserve from 18 June to 2 August 2023, for a total of 52 days at RF, 48 days at GJ, and 45 days at VF. We used one camera trap per sub-site (see Figure 6), resulting in a total of three cameras across the sample sites for species detection.

Cameras were attached with straps to trees 1 metre above the ground in semi-open areas selected on site. All cameras were positioned 5-10 metres from the river. I used DOERR SnapShot (Mobile Black 5.1) cameras configured to take two high quality photos when motion was detected and set to medium sensitivity. In addition, one photo a day was sent to my mobile phone via MMS to monitor battery status and ensure the cameras were working properly. The model has an infrared flash, so also detection at darkness was possible.



Figure 6. Set up of Camera trap (DOERR SnapShot), here at Rusfors subsampling site 1 at a birch tree (Picture from set-up day – 12. June 2023) A: set up B: set up-close up

After retrieval, all photographs were manually reviewed and the date, time and species of all positive detections were catalogued. This data was then analysed using R to visualise the total number of species detected and their abundance at each site.

2.5 Audio recorder

Settings

In order to assess the presence and absence of bat species (order *Chiroptera*) at the study sites, I implemented passive acoustic monitoring (PAM) using autonomous recording units (ARUs), in this case the Wildlife Acoustics Song Meter Mini Bat Ultrasonic Recorders (see Figure 7). These units were configured using the *Wildlife Acoustics Song Meter Configurator* app, following the bat survey guidelines recommended by Länsstyrelsen (contact: Åsa Granberg). These guidelines included setting the device mode to “ultrasound recording” and scheduling recordings from one hour before sunset to one hour after sunrise, with the recording frequency set to “always” to ensure continuous monitoring when ultrasound signals were detected.

Six audio recorders were used for this study, two per site. The selection of two of the three subsampling sites for recorder placement prioritised maximum spatial separation (see Appendix - Figure 4). In the field, I identified a (semi-)open area and selected a nearby tree for the setup. The ARUs were attached to the tree with a rope at a height of 1.70 m, and all were installed approximately 10 to 20 m from the rivers. The deployment dates were 19 July 2023 for VF and 20 July 2023 for GJ and RF, with all recorders collected after 14 nights of operation, ending on 2 and 3 August respectively.



B



Figure 7. A: Set up of audio recorder at GJ site in late July 2023; B: Product photo of Wildlife Song Meter Mini Bat Ultrasonic recorder (Picture from www.wildlifeacoustics.com)

uncertainty, we grouped calls at the family level. I developed a decision flowchart (see Figure 9) to aid this differentiation process, particularly for *Myotis* species where only *Myotis daubentonii* could be confidently identified. This species is distinguished by its unique S-shaped call and a frequency range between 30 and 80 kHz, which is unique compared to other *Myotis* species (Russ, 2021).

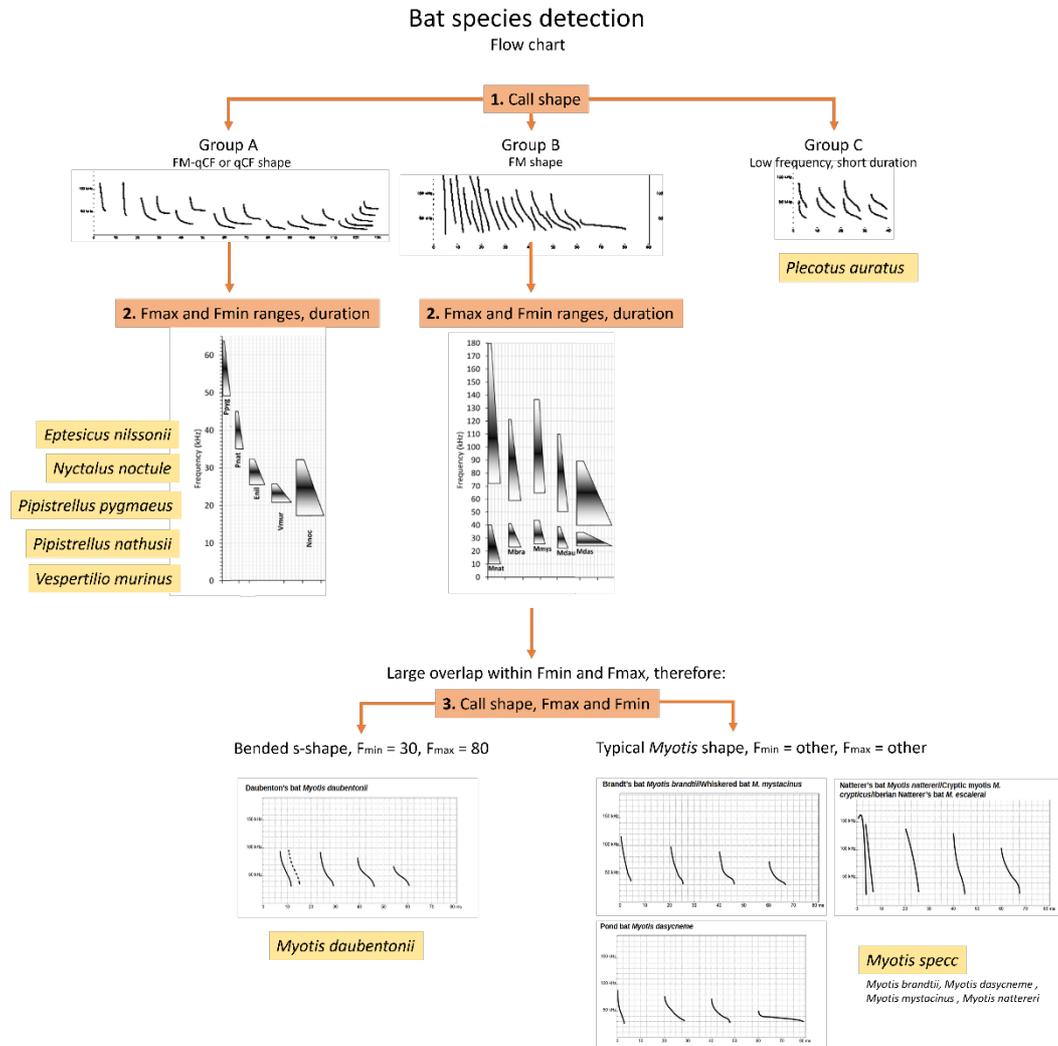


Figure 9. Decision flowchart for the identification of the 11 bat species occurring in northern Sweden used in this study; all diagrams from: Russ, 2021. 1. Step: Distinguish call shape based on shape and frequency. 2. Step: Differences in and duration of call. 3. Step: Distinguish between *Myotis daubentonii* and other *Myotis* species based on call shape, min and max frequency

2.6 Insect traps

In order to assess arthropod biodiversity within the study sites, I used pitfall traps to assess ground-living insects and IBL type-II traps for flying insects.

I equipped each randomly sampled subsite (see *Methods - Study sites*) with 1 IBL trap and 3 pitfall traps, resulting in 3 IBL and 9 pitfall traps per site. The location of the traps within the subsite was randomly chosen on site. Glycol was used as the preservice fluid for both types of traps.

At all locations, I hung the IBL trap between two trees about 2-3m apart, facing an open area. The IBL traps were fixed with a rope at a height of about 2 m, so that the capture funnel hung at 1 m height. The coordinates were recorded, as well as the direction the IBL was facing when sampling. The IBL traps were equipped with a 0.75L bottle filled to about 1/3 with glycol. The pitfall traps have a diameter from ca. 10 cm and were filled to about 1/4 with glycol. The pitfall traps were buried in the ground until the edge of the cup reached the surrounding soil and then covered with metal lids, leaving a small gap of 2 cm between the lid and the cup to protect it both, from rain and to prevent larger animals (such as shrews or amphibians) from falling in.

The insect traps were deployed from mid to the end of June and the sampling period varied between 10, 13 and 17 days between sampling sites (see Appendix - Table 7 for details). In the following evaluation of biodiversity, the different sampling times was taken into account.

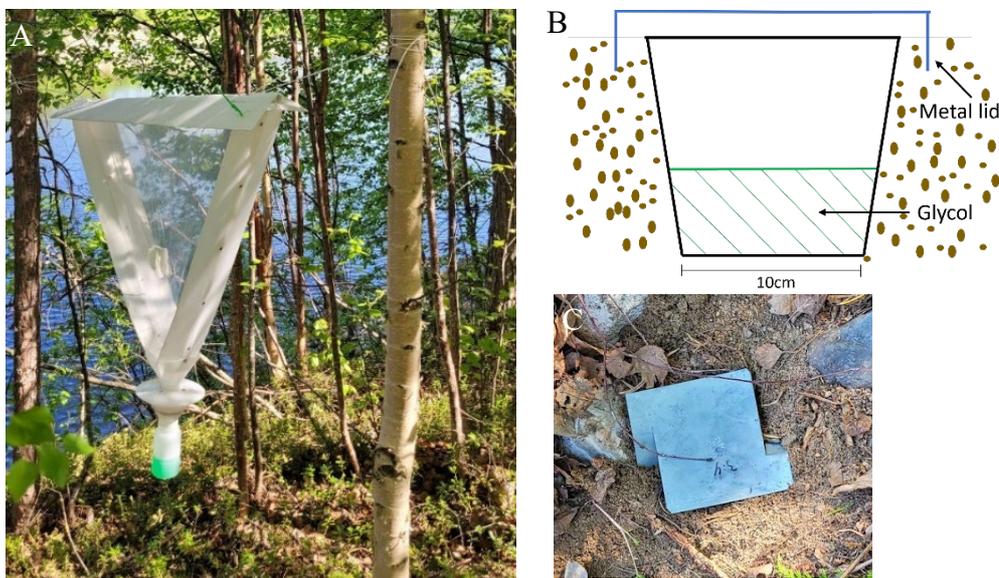


Figure 10. Insect traps set up. A. IBL type-II window trap for flying insects, here at RF subsampling site 1 (date of installation: 29 June 2023), B. Schematic structure of a pitfall trap cross-section to capture ground crawling insects, C. Pitfall trap from above with visible metal lid.

Classification and analysis

After collecting the traps in the field, I sorted them into different groups in the laboratory. Due to the high probability of making errors, I decided to sort and count the insects either in their systemic class or, if possible (when the change of error is low), in their order or family. The sorting resulted in the following list of taxonomic groups:

Table 3. Classification of taxonomic groups used for this study, all belong to the Phylum Arthropoda. Black: Taxonomic group I used in this study, Grey: Guidance to differentiate the phylogenetic order

<i>Subphyla</i>	<i>Class</i>	<i>Order</i>	<i>Family</i>
<i>Chelicerata</i>	<i>Arachnida</i>	<i>Acari</i>	
		<i>Araneae</i>	
		<i>Opiliones</i>	
<i>Tetraconata</i>	<i>Insecta</i>	<i>Coleoptera</i>	<i>Carabidae</i>
			<i>Cerambycidae</i>
			<i>Chrysomeliadae</i>
			<i>Coccinellidae</i>
			<i>Curculionidae</i>
			<i>Scarabeidae</i>
			<i>Scolytidae</i> (Subfamily
			<i>Curculionidae)</i>
			<i>Silphidae</i>
			<i>Staphylinidae</i>
		<i>Dermaptera</i>	
		<i>Hymenoptera</i>	<i>Apoidea</i>
			<i>Diprionidae</i>
			<i>Formicidae</i>
		<i>Diptera</i>	
		<i>Ephemeroptera</i>	
		<i>Hemiptera</i>	<i>Sternorrhyncha</i>
		<i>Heteroptera</i>	
		<i>Homoptera</i>	
		<i>Lepidoptera</i>	
		<i>Neuroptera</i>	
		<i>Orthoptera</i>	
		<i>Plecoptera</i>	
		<i>Trichoptera</i>	
	<i>Collembola</i>		
	<i>Diplura</i>		
<i>Myriapoda</i>	<i>Chilopoda</i>		
	<i>Diplopoda</i>		

Before calculating species richness, abundance and biodiversity indexes and graphical display, I filtered out the columns, where the count was 0 in the beginning of the analysis.

2.7 eDNA analysis (soil samples)

Sampling in the field

To get a picture of riparian terrestrial biodiversity, soil samples were taken from the sites at the beginning of August 2023 (02. and 03. August) and were analysed on eDNA (environmental DNA). Soil eDNA is a good way to get information about the present taxa, without being an invasive for the local ecosystem (Edwards et al., 2018).

I took about 500g of soil from each subsampling site, resulting in 3 samples per site. I filled a 1 litre plastic bag from IKEA and pooled soil from 3 different nearby locations at each subsampling site, to minimise sampling bias. I wore plastic gloves between each subsampling site to reduce contamination. The bags were immediately sealed and stored at -80°C until shipping.

The samples were then shipped on dry ice to the laboratory at the end of August 2023 for analysis. I chose to work with the laboratory SE Analytics AB (<https://seanalytics.se/>) in Gothenburg. My contacts there were Per Sundberg and Marina Panova, who carried out the laboratory processing and bioinformatic analysis. I chose to use the COI primer, which is an all-metazoan primer, because it detects a mitochondrial gene found in all metazoans (Folmer et al., 1994, Kirse et al., 20-21, Geller et al., 2013). Bioinformatics was performed using the BOLD reference system, which is widely used (e.g. Geller et al., 2013). I received a detailed protocol of the laboratory processing and analysis. The full report, written by Marina Panova, Marty Breidenbach and Per Sundberg, is attached as Appendix II. The data were received at the end of January and analysed using R.

Filtering of data

Since some of the detected ASVs (Amplicon Sequence Variant) did not match any entry in the reference database, nor could they be assigned to any phylum, class, order, species or genus, I decided to remove them from the following analysis. In addition, in the control (which ran alongside my samples and contained a known species community), it was found that 1% of the total reads were contaminated with species not present in the sample due to barcode jumping in the laboratory process. I therefore decided to remove 5% of the total reads to be conservative with the

following data analysis. This led me to remove all positive detections where the total read of ASV across all sites was less than 33 reads, to ASVs which just occurred in an exceedingly small amount and are likely to got detected due to contamination. To further provide an indication of the species present at each site, I filtered out all ASVs that could not be assigned to a *family*, *genus*, or *species*. The following flowchart illustrates the data filtering process visually and with numbers from the data:

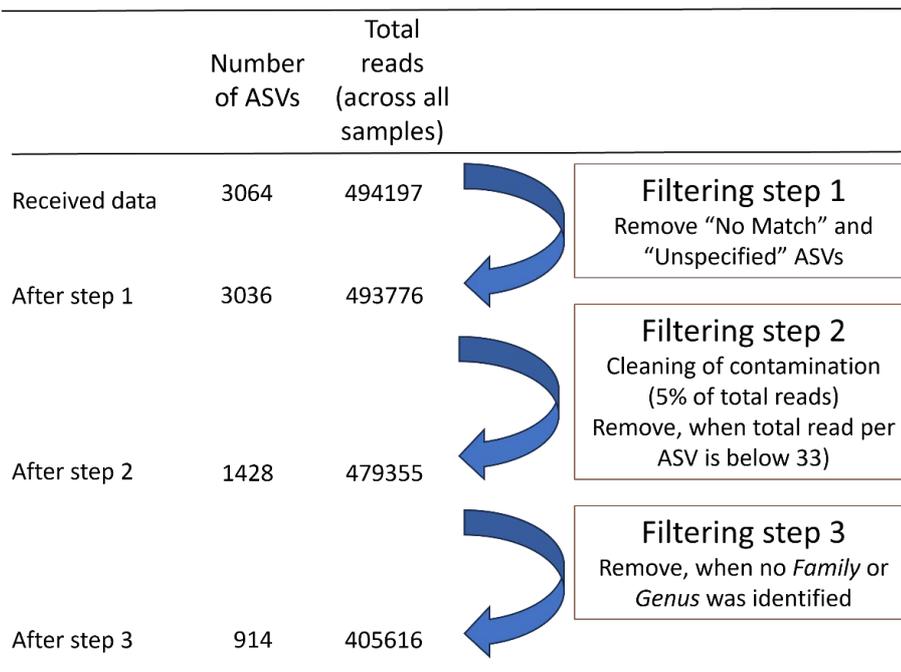


Figure 11. Flow chart for filtering process of eDNA data with according steps and numbers from the dataset to visualise filter process.

2.8 Statistical Analysis and biodiversity indexes

Statistical analysis was carried with the software R Studio 4.3.1 (R Core Team, 2018). I used the R package “dplyr” (Wickham et al., 2023) to clean and filter the data. Before using a parametric test such as ANOVA or t-test, I checked for homogeneity of variance with the package “car” (Weisberg, 2019) and normality of error. Otherwise, I used the non-parametric Kruskal-Wallis test or Mann-Whitney U test. For ordination visualisations (NDMS and CCA) and calculated biodiversity indexes I used the R package “vegan” (Okasen et al., 2019). Plots were created with the R package “ggplot2”. To test differences in species communities, I performed PERMANOVA with the *adonis2* function from the R package “vegan” (Okasen et al., 2019).

As measurements for biodiversity, I used the following indexes in this study: Species richness, Shannon-index, Simpson-Index and Whittaker beta diversity.

Species richness is the number of present species.

The Shannon-Index (or: Shannon-Wiener-Index) is based on relative abundance data and takes both, species richness and evenness into account. However, it is more sensitive to richness (than evenness) and therefore focus more on rare species. The Shannon index is calculated with this formular: $H' = -\sum_{i=1}^s p_i (\log * p_i)$. Whereby p_i the relative abundance from each species $p_i = p(\text{abundance species } i) - \text{total}(\text{abundance all species})$. A higher Shannon-Index indicates a higher biodiversity, so that no species is dominating the community.

The Simpson-index, invented by Simpson (1949), is also based on relative abundance data of richness and evenness and calculates the “probability that two individuals chosen at random and independently from the population will be found to belong to the same group” (Simpson, 1949). It is therefore more sensitive to evenness (than on richness) and therefore for common species. In the R-package “vegan” the Simpson index is calculate with this formular: $D = \sum * p_i^2$. There is the option to use “*simpson*” in the function, which configurate the value into $1 - D$ or the option “*invsimpson*” with configurate the index into $D/1$ (Okasen et al., 2019). In this study all Simpson Indexes are converted into $1 - D$, ranging between 0 and 1, and can be interpretated that low values suggest the community is dominated by a single or a few species and a high value suggest a more even distribution and a greater biodiversity.

The Beta-diversity, calculated with the gamma diversity (diversity of all population) and the alpha-diversity of each individual population, to investigate pairwise similarities between population communities (Whittaker, 1960). In R, I used the function *betadiv = “w”* from the R-package “vegan” package. A value close to 0 indicated a large similarity between species communities, whereby a value close to 1 a large dissimilarly between two populations.

3. Results

The different methods I used to determine the three sites implied that the modified and non-modified sites differ in some of the tested facets of EH and environmental factors. The results for biodiversity have been mixed and were different between the methods.

3.1 Vegetation analysis

I tested the difference in EH between the sites with the SD of NDVI, vegetation height, slope and flow of each area. Furthermore, I compared the mean NDVI, mean vegetation height and slope as other environmental factors between the sites.

NDVI and vegetation height

There is a graphical presentation of all NDVI and vegetation height in the appendix (see Appendix I - Figure 5 for NDVI and Figure 6 for vegetation height).

For site GJ, the mean NDVI is -0.01, with a SD of 0.19, indicating a low vegetation productivity and some variability in these conditions across the site. NDVI values around 0 indicate that there is mostly non-vegetated area. The mean height of vegetation at GJ is 30.23 m, with a SD of 4.15 m, suggesting moderate vegetation height with relatively low variability at the site.

RF shows a mean NDVI of 0.08 with a standard deviation of 0.23, representing higher productivity than GJ, along with a higher degree of variability. Across all sites, the variability in horizontal EH (SD NDVI) was highest in RF. The mean height is 32.63 m with a SD of 3.87 m, indicating a slightly higher average vegetation height than GJ and with lower variability.

Lastly, site VF exhibits the highest mean NDVI of 0.13 with a SD of 0.20, suggesting the highest productivity among the three sites, with a moderate level of variability. The mean height at VF is 32.82 m, the highest among the sites, with a standard deviation of 6.19 m, indicating the greatest variability vegetation height and therefore the highest EH in the vertical facet.

I was able to show that the **SD of NDVI** does not significantly differ between sites (ANOVA: $F=2.725$, $p=0.244$) nor between modified (RF, GJ) and unmodified (VF) riparian areas (t-test: $p=0.590$).

However, the **SD of vegetation height** was significantly different between sites (ANOVA: $F=6.688$, $p=0.0126$ *). With a Tukey post-hoc test based on the test, I could prove that the difference between RF and GJ is not significant ($p=0.897$), but between VF and GJ it is ($p=0.0304$ *) and also between VF and RF ($p=0.0148$ *) (see Figure 12). This indicates that there is a significant difference between modified (GJ, RF) and free flowing (VF), which was also found to be significant in a separate t-test ($p=0.003007$ **).

There was no statistically significant difference for the **mean vegetation height** between sites (ANOVA: $F=1.609$, $p=0.244$) or between modified and non-modified (t-test: $p=0.250$).

In contrast to the **mean NDVI**, that differed significantly from each other (ANOVA: $F=9.647$, $p=0.0038$ **). With the Tukey post-hoc test based on the ANOVA, I prove that the difference between RF and GJ ($p=0.0400$ *) and between VF and GJ ($p=0.0033$ **) is significant, while between VF and RF there is no significant difference ($p=0.276$). This leads to the conclusion, that in sense of mean NDVI, as vegetation index, VF and RF are closer to each other than both with GJ.

The results indicate that not only does productivity vary significantly across the sites, as shown by the mean NDVI values, but there are also significant differences in the variability of the vegetation height, with site VF showing both the highest mean values and the greatest variability in height. The results are additionally represented in Figure 12.

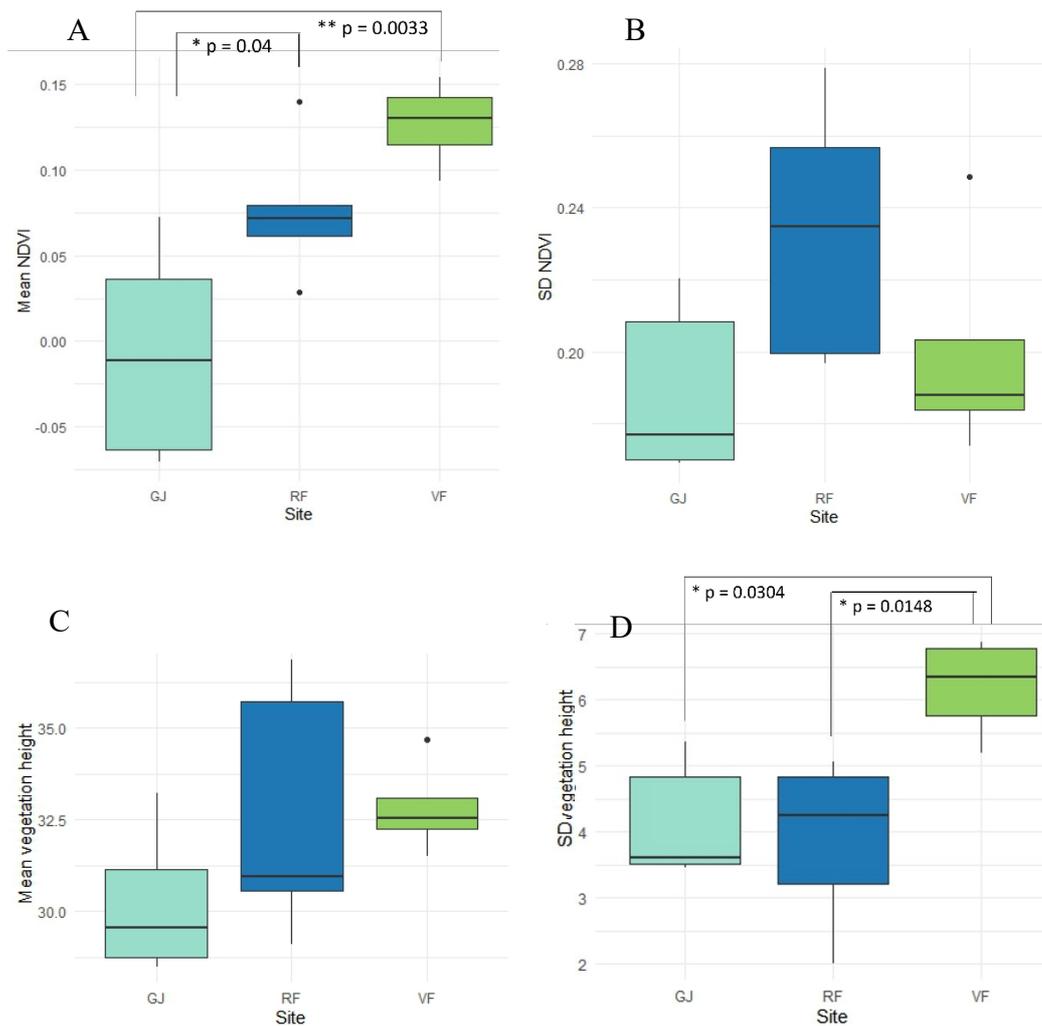


Figure 12. Results from vegetation height and NDVI measurements in form of boxplots. Light blue: modified site GJ, Dark blue: modified site RF, Green: non-modified site VF. A: Mean NDVI as index for productivity, significant difference between sites and highest in non-modified site VF. B: SD of NDVI as measurement for horizontal environmental heterogeneity (EH) with non-significant differences and highest value in modified site RF. C: Mean vegetation height with no significant differences between sites and highest mean value at non-modified site VF. D: SD of vegetation height as measurement for vertical EH with significant differences between sites and highest value in VF.

Slope

The following graph (see Figure 13) shows the slope in all study areas. Green indicates a high percentage of slope. It can be seen that in particular the slope immediately adjacent to the waterway is much steeper in RF and GJ, where the dredged sand has been deposited than in the free-flowing and unmodified site VF.

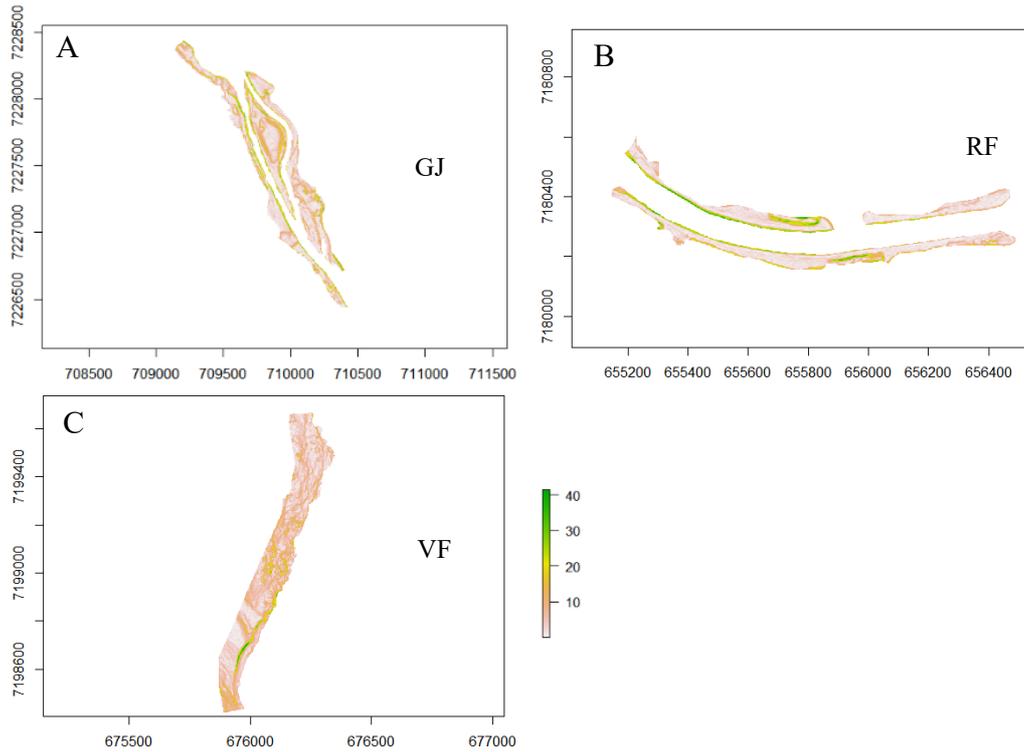


Figure 13. Percentage of slopes in all three study areas. Green indicates a high percentage of slope, light pink a low percentage. Legend for all three plots. A: Modified site GJ with high slope close to the river line. B: Modified site RF also shows steep slope along to the river. C: Non-modified site VF with moderate slope along the entire area.

Based on the distribution of steep slopes near the water at the modified sites, I decided to create Figure 14 to visualise the difference in slope in 10m blocks from the waterline. The results suggested that the slope of the first 10m of the terrestrial riparian zones seemed to differ greatly between modified and unmodified sites. This part is likely to have a large impact on flooding, as a high slope indicates that the water has to rise a lot to get in contact with the vegetation. Therefore, I used the mean and SD of the slope between 0 and 10 m from the waterway as a measure of EH in this study.

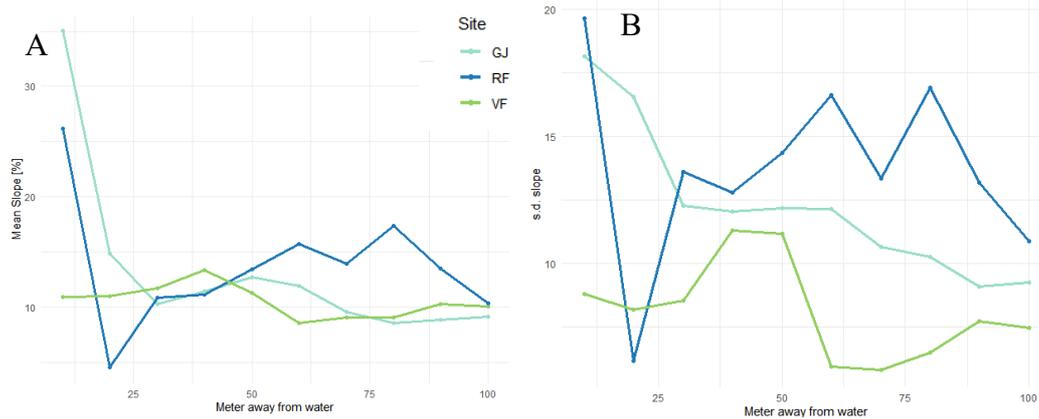


Figure 14. Slope (in percent) over a distance from 0-100 m away from water in 10m blocks. A: Mean slope. B: SD of slope. Pointing out that the mean and SD of slope shows large variation in the first 10m away from the water between modified sites in blue (light blue GJ, dark blue: RF) and the natural area in green (VF).

For site GJ, the mean slope between 0 and 10m from the water is 35.01%, with a SD of 18.14%, indicating a relatively steep incline and considerable variability in slope steepness across the site.

Site RF has a mean slope of 26.19% with a SD of 19.63%. This suggests that, while the average slope is less steep than at GJ, the variability in slope steepness at RF is slightly higher, pointing to a more diverse topography.

Lastly, site VF shows a mean slope percentage of 10.97% with a SD of 8.79%. This site has the gentlest average slope of the three and the lowest variability from meter 0 to 10 from the river, indicating a more uniform and moderate slope across this area. These values provide insights into the topographical diversity and slope steepness across the three sites, ranging from relatively steep and variable slopes at GJ and RF to a more uniform and gentler slope at VF.

Flow

To see a difference in the flood regime, I have created Figure 15 to show the difference in flow events between the different sites. It is clear that RF and GJ are very different from their natural flow regime (pink line indicates corrected measured flow, green line estimated natural flow). In VF, the measured peak of the spring flood in July is clearly visible, which is absent in the hydropower influenced sites GJ and RF. Furthermore, the flow regime in GJ and RF is more or less stable throughout the year compared to VF due to the free-flowing Vindel River, which comes to a halt during the winter months.

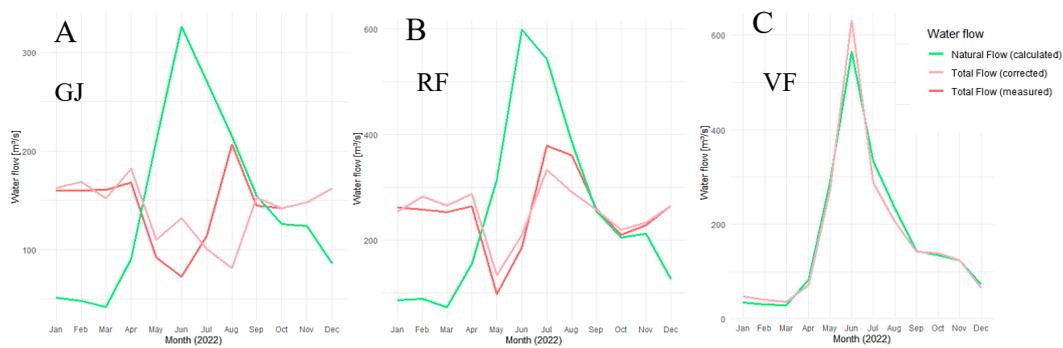


Figure 15. Monthly flow data (2022) from SMHI data for all three sites. Green line indicates natural flow without hydropower, pink line indicates corrected flow data, red line is actual measured flow. A: modified site GJ with hydropower shows large difference from natural flow with no spring flood and moving water in the winter months. B: Modified site RF with hydropower plant, shows similar pattern than GJ. C: Non-modified site VF along free-flowing river shows large overlap with estimated natural flow including spring flood and non-flow events during winter months.

I calculated the mean flow during the year 2022 based on the data from SMHI each month for all the sites. GJ has the lowest mean annual flow of $141.28 \text{ m}^3/\text{s}$, RF has the highest mean annual flow of $252.58 \text{ m}^3/\text{s}$ and VF has an annual flow of $171.78 \text{ m}^3/\text{s}$. The mean flow will not be discussed further, since it is depending on a variety of different factors as well (see Methods – EH – Flow data). However, due to the controlled release of water by the hydroelectric turbines, the constant water flow throughout the year and the absence of spring flood, the SD of the annual flow is lowest in GJ with a SD of $30.02 \text{ m}^3/\text{s}$, followed by RF with $50.35 \text{ m}^3/\text{s}$ and considerably higher in VF with a SD of $168.74 \text{ m}^3/\text{s}$.

Summary of vegetation analysis

To address **hypothesis 1** I tested if modified sites (GJ and RF) have less variation in the vertical, horizontal EH and flow than the non-modified VF site, but more variation in topographic EH, and also to see if this pattern follows the degree of modification (highly-modified GJ → modified RF → non-modified VF). I also compared mean NDVI, vegetation height and slope. For the NDVI and vegetation height measurements I was able to perform a statistical test, but the slope and flow data were compared based on their values due to a lack of statistical power.

The findings of the environmental analysis of the three sites can be concluded as:

- Horizontal facet of EH:
Statistically, I could not prove that the variation in NDVI differs between the sites. Findings indicating however, that the modified site RF shows the highest variation in horizontal EH, flowed by the natural site VF and is lowest in GJ.

- Vertical facet of EH:
The variation in tree height differs significantly between the sites. The SD of the vegetation height is the highest in VF, followed by GJ and is the lowest in RF. I could prove that there is a difference in modified and non-modified site, but the order does not follow the expected degree of modification.
- Topographic facet of EH:
The variation of slope is highest in the first 10 m in RF, followed by GJ and the lowest in VF. The values indicate a difference between modified and unmodified sites but did not follow the expected degree of modification.
- Variation in flow:
The SD of flow is the highest in the non-modified site VF, and lowest at GJ, followed by RF. The values indicate a difference in non- and modified sites, as well in the expected level of modification.
- Mean NDVI:
Mean NDVI is highest in VF, followed by RF and lowest in GJ. Significant differences between the three are highlighting these findings and also the gradient of modification as expected is visible.
- Mean vegetation height:
Mean vegetation height is highest in VF, followed by RF and lowest in GJ. However, the differences between the sites are not significant. Also, these results follow the expected level of modification.
- Mean slope:
Mean slope is highest in GJ, followed by RF and lowest in VF. These findings follow the expected level of modification.

In order to visualise whether or not the expected pattern of environmental factors follows the degree of change, I have created Figure 16.

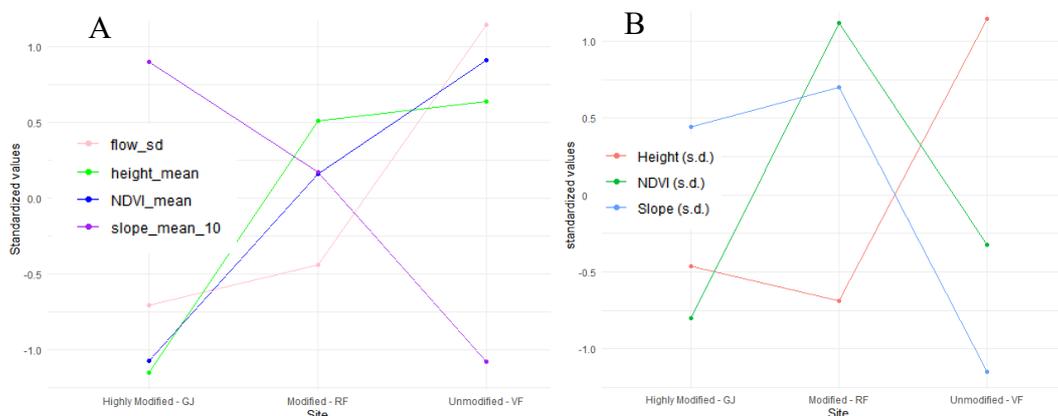


Figure 16. Standardized environmental factors and level of modification A: Variables do follow the pattern of modification. B: Variables which not follow the expected pattern of modification.

3.2 Camera traps

Across all sites, the camera traps detected a total of 11 distinct species. Species richness varied, ranging from zero to nine species identified per subsampling site. A selection of positive detection visualises the picture frame of each camera trap in Figure 17:



Figure 17. Selection of positive detection from camera traps in all three sites. GJ1 Group of reindeers, GJ2 Red fox, GJ3 no species detected during sampling period, RF1 Whooper swan, RF2 Eurasian Woodcock, RF3 Mountain hare, VF1 Roe deer, VF2 Reindeer, VF3 Reindeer

Because of the difficulties in distinguishing between individuals, I decided not to use abundance data to estimate biodiversity measures and focus more on the species richness (Detected species in each site). For example, when one individual was recorded multiple times at on site, it is likely that these detections represent repeated sightings of the same individual. A full list of all positive detections is given in the Appendix (see Appendix I - Table 9).

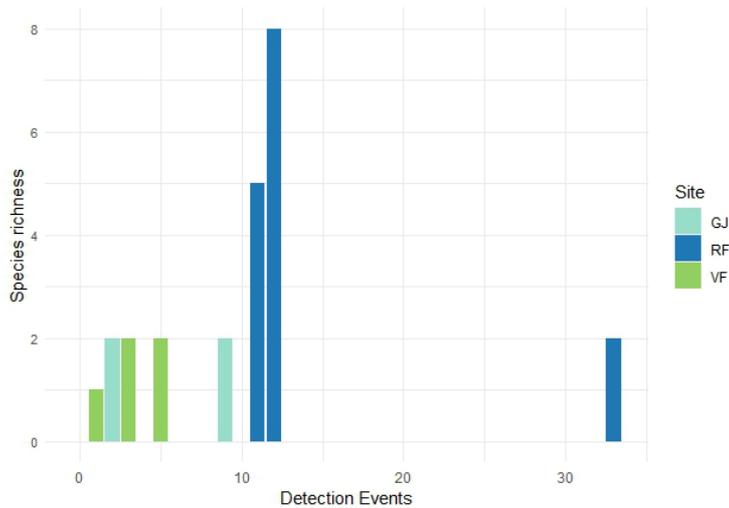


Figure 18. Results from camera traps: Detection Events and Species Richness for each camera trap. Pointing out that the site with highest detection events has only a low species richness due to multiple recordings of the same species or individual.

RF had the highest species richness (=10) and number of positive detections (= 56) of all the sites. However, as mentioned above, it is likely that some detections represent multiple records of the same individuals. Specifically, subsampling site 2 at RF, where Eurasian Woodcock were 23 times observed and field fare 10 times, recorded the highest number of detection events. The first subsampling site at RF, which covered a large part of the river surface (see Figure 171, RF1), facilitated the detection of several aquatic species, including Whooper Swan, Great Crested Grebe and Canada Goose. In addition, terrestrial detections at this site included fieldfare, red fox, roe deer, woodcock, common sandpiper and red squirrel, making it the site with the most diverse species detections of all camera trap setups. Subsampling site RF 3 also showed significant species richness, with the presence of mountain hare, fieldfare, red fox and common sandpiper.

The open habitat of this site is likely to have contributed to the wider range of species recorded, in contrast to the denser vegetation of the VF sites, where only roe deer and reindeer were consistently observed (Richness = 2). The dense undergrowth in VF probably hindered the detection of smaller mammals and birds (Detections = 11).

In contrast, despite the open habitats, only a few species were detected at all GJ subsampling sites (Richness = 3, Detections = 11). At GJ 1, roe deer and groups of reindeer were detected. Reindeer and red fox were observed at sampling site 2. Notably, GJ3 did not yield any positive detections throughout the study period.

The detection probability (calculated with: Detection events / Sampling time (in days)) is lower in VF than in RF (VF = 6.7 %, 13.3 %, 4.4%, total=8.1%) compared to RF (RF= 23.1 %, 63.5%, 21.5 %, total = 35.9%). I expected to see this difference

due to the dense vegetation in VF. However, due to the small sample size of camera traps, it is not possible to conduct an occupancy model, to see if the difference in detected species is due to the location or the vegetation. GJ had the lowest detection probability overall (GJ= 18.8%, 4.2%, 0.0%, total=7.6%), but with also a low vegetation understory.

3.3 Audio recorder

From the 11 different species occurring in the North of Sweden, this study was able to detect only 4 distinct species/families across all sites with the PAM. There were *Eptesicus nilssonii* (Northern Bat), *Myotis specc.* (*Myotis brandtii*, *Myotis dasycneme*, *Myotis mystacinus* or *Myotis nattereri*), *Myotis daubentonii* (Daubenton's bat) and *Vespertilio murinus* (Particoloured bat).



Figure 19. Overview of detections and species distribution over all subsites. EPTNIL = Northern bat (*Eptesicus nilssonii*), MYO specc. = *Myotis* species, MYODAU = Daubenton's Bat (*Myotis daubentonii*), VESMUR = Particoloured bat (*Vespertilio murinus*). The natural site VF has overall the highest detection of calls, followed by the highly modified site GJ and was lowest in RF.

Figure 19, shows a graph with detected bat calls, as well as the distribution of detected species in each sample plot. Values, like richness and biodiversity indexes can be found additionally in the Table 4.

The ARUs at the GJ detected 239 calls at GJ1 and 187 at GJ2 (total = 426).

At RF, there was a notable difference in detection numbers between subsites: RF1 recorded only 23 bat calls, while RF2 recorded 129 calls, making it the site with the lowest number of bat calls overall (total = 125).

The PAM highlighted VF as the site with the most bat detections. The ARU at VFs recorded the highest levels of bat activity, with VF1 recording 314 individual calls and VF2 the highest with 436 calls (total = 750).

The Particoloured bat (*Vespertilio murinus*) call was detected in 14 sampling days only rarely with five times in total (within 20 min in one night) at RF2 and one single time at VF2. In all the sampled points, ARUs recorded three different species/families: Northern bat (*Eptesicus nilssonii*), *Myotis* species and Daubenton's Bat (*Myotis daubentonii*). According to *Artdatabanken.se*. the most common species in northern Sweden is *Eptesicus nilssonii* (Northern Bat). According to this, over all sites, the Northern bat was the species with the highest detection (total = 795), followed by Daubenton's bat (total = 361), *Myotis spec.* (total = 174).

I observed considerable variation in detected numbers and species diversity between subsites, suggesting that the placement of ARUs likely influenced detection probabilities due to different site-specific characteristics.

Table 4. Biodiversity indexes from audio recorder for bat detection

	GJ		RF		VF	
	1	2	1	2	1	2
Total count	426		152		750	
	239	187	23	129	314	436
Species Richness	3		4		4	
	3	3	3	4	3	4
Shannon Diversity (H')	0.987		0.833		0.928	
	0.054	0.780	0.615	0.862	0.820	0.151
Simpson Diversity (1/D)	0.588		0.466		0.550	
	0.017	0.492	0.329	0.490	0.489	0.054

Taking the biodiversity indexes into account, indicated that the biodiversity is highest in GJ site, in both, Simpson- and Shannon- Index ($H' = 0.987$, $1/D = 0.588$), since the distribution of different species is more variable. However, GJ1. The second largest diversity in bat species could get detected in the VF site, both in sense of Simpson- and Shannon-Index ($H' = 0.928$, $1/D = 0.550$). The lowest diversity of bats could get detected in RF ($H' = 0.833$, $1/D = 0.466$). However, since only 4 different species/families got detection throughout the sampled time, I decided to focus more on the amount of detection as indicator for bat diversity. The activity of bats is often used as indicator for bat diversity.

Since I detected just a small species community within bats, it was impossible to calculate a meaningful beta diversity or PERMANOVA to find if there is a statistically significant difference between the sites.

3.4 Insect traps

Overall, all samples from both trap types and all sites, I detected 27 different taxonomic *Arthropoda* groups.

Even though the traps in VF were only out for 10 days, the sampling period in GJ was 17 days and in RF 13 days (see Appendix - Table 7). VF was the site with the highest number of detections, with a total of 1006 individuals in both IBL and pitfall traps (IBL = 628, pitfall = 378). At the RF site, a total of 755 arthropod individuals were found and it was the site with the highest number of individuals in both trap types after the VF site (IBL = 518, pitfall = 237). The site with the lowest number of individuals found in both trap types (IBL = 143, pitfall = 412), with a total of 555 individuals, is the GJ site. Figure 20 shows the number of the different taxonomic groups per subsampling site.

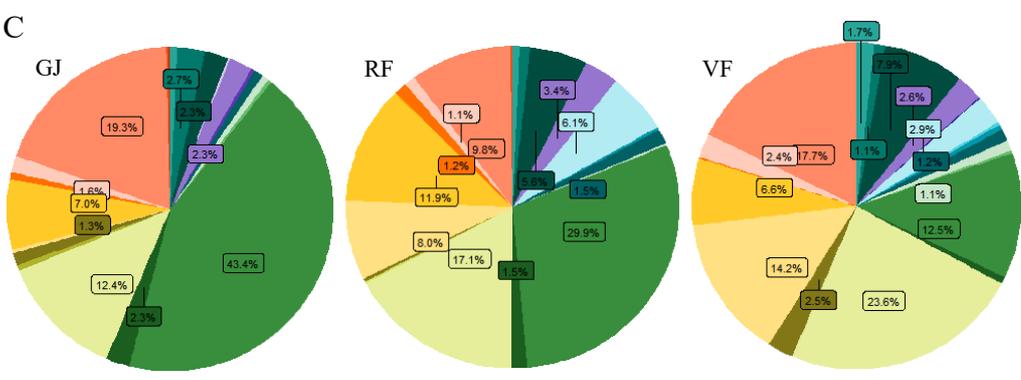
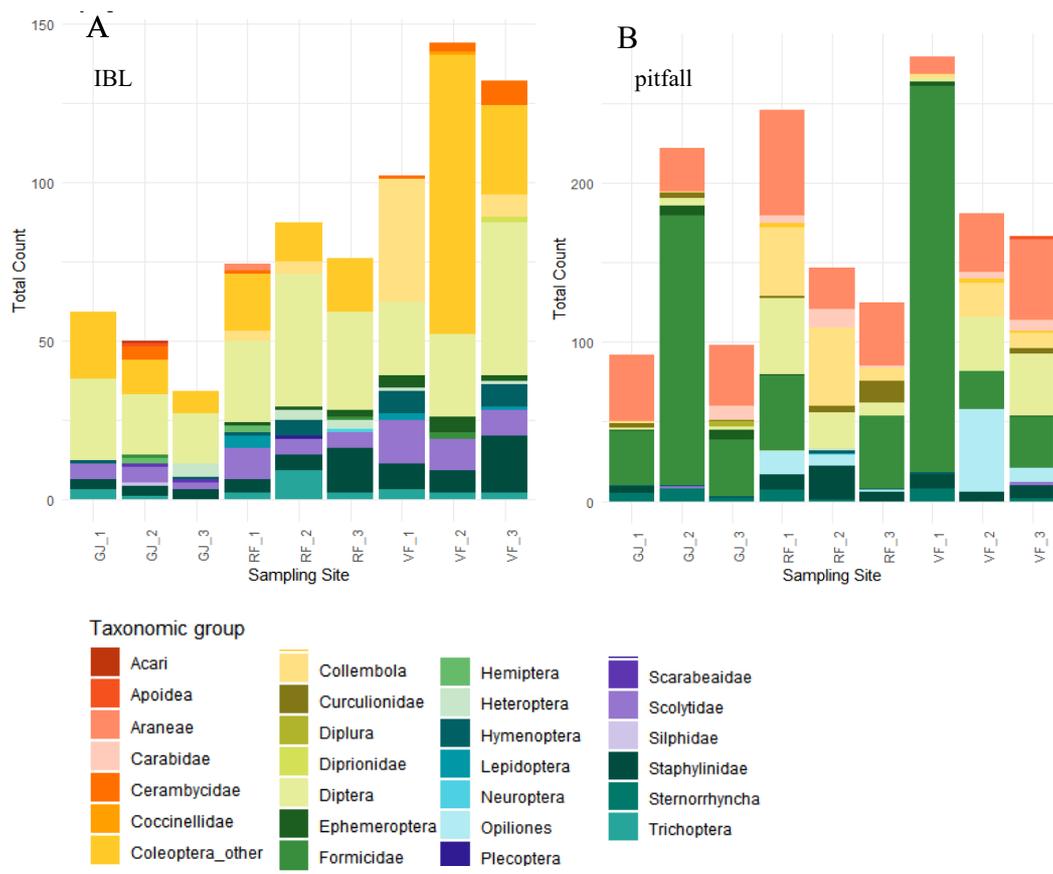


Figure 20. A & B: Numbers of detected arthropod taxonomic groups per subsampling site, with each group distinguished by colour. A: Flying insects captured with IBL trap, B: Ground crawling insects captured with pitfall trap, C: Distribution of taxonomic groups within a site across all traps, Legend represents taxonomic groups for all shown plots.

Since the detected communities showing a large overlap in the detected groups, I decided to pool the subsites (1, 2 and 3 together) and will continue the discussion with the results, seeing the site as total.

The biodiversity indexes (Shannon- and Simpson-Index) are the lowest in GJ for both traps (IBL: $H'=1.865$, $1/D = 0.773$; Pitfall: $H'=1.374$, $1/D = 0.614$) as well as for the traps combined ($H'=1.969$, $1/D = 0.769$) in comparison with the other sites. The species richness with both trap types combined had site GJ, but also showed the lowest biodiversity indexes and lowest total abundance. Concluding from that, GJ is the site the lowest arthropod biodiversity, despite the highest richness.

The site RF shows a higher species richness in the IBL trap (16 groups) then GJ or VF (both each 14 groups). In both measured indexes for biodiversity, the diversity in arthropods detected with pitfall traps ($H'=2.060$, $1/D = 0.870$) and for both traps combined ($H'=2.334$, $1/D = 2.334$), is the highest across all sites.

In VF the biodiversity is the most diverse in IBL trap across all sites ($H'=2.020$, $1/D = 0.821$). All the values for abundance, species richness and biodiversity indexes for each site can be additionally found in Table 5.

To measure the difference between the site, I calculated the beta diversity between the sites. It became clear, that the sites RF and VF are more similar with their species composition (Beta-diversity = 0.130), in comparison to the similarity of RF and GJ (Beta-diversity = 0.191) or VF and GJ (Beta-diversity = 0.191).

To address **hypothesis 2**) if there is a difference in species communities between the sites is also statistically proven, I performed a PERMANOVA with the `adonis2` function in R, based on the distance matrix of all subsampling sites. This function takes the total abundances of each detected group into account and compares the differences across the entire community. Approximately 34.6 % of the differences in the insect community can be explained due to the differences between sites ($R^2 = 0.346$). However, this difference is not statistically significant ($F=1.589$, $p=0.194$).

Table 5. Abundance, Richness and Biodiversity indexed from insect traps. Results from subsites pooled together into a site for a better overview about the total area.

	GJ	RF	VF
Total abundances			
IBL traps	143	237	378
Pitfall traps	412	518	628
Combined trap types	555	755	1006
Species richness			
IBL traps	14	16	14
Pitfall traps	12	14	14
Combined trap types	21	20	20

Shannon-index (H')			
IBL traps	1.865	1.997	2.020
Pitfall traps	1.374	2.060	1.746
Combined trap types	1.969	2.334	2.232
Simpson index (1/D)			
IBL traps	0.773	0.792	0.821
Pitfall traps	0.614	0.842	0.733
Combined trap types	0.769	0.870	0.849

To address **hypothesis 3**) to gain more information about the biodiversity between the sites, I also focused on indicator species for riparian zones and compared the occurrence in the sites. The most *Carabidae* individuals were found in RF (=18), followed by 11 detected individuals in VF and the lowest abundance was found in GJ with 9 detections. Also, the amount of *Staphylinidae* was the highest in RF (60), closely followed by VF (56) and the lowest detections of individuals was found in GJ (13).

However, the indicator species *Ephemeroptera* was most present in VF (15), closely followed by GJ (13) and had the least counts in RF (5).

There was only 1 *Plecoptera* individual found in total, at the RF site.

The most *Trichoptera* individuals were found in RF (13), followed by VF (7) and has the last detections in GJ (4).

These findings are additionally presented in a table in the Appendix (see Appendix I - Table 10) along with counts of all detected taxonomic group in each site.

It can be concluded from the arthropod analysis:

- GJ shows the lowest biodiversity in arthropod taxonomic groups (lowest total abundance, biodiversity indexes)
- RF has the highest total biodiversity (based on Shannon- and Simpson-Index) and in ground-crawling insects.
- VF has the highest diversity in flying insects.
- VF site has the highest numbers of arthropods overall, even with the lowest sampling time. Followed by RF and the is the lowest in GJ.
- Indicator species present in all sites, but general trend to be more abundant in RF, followed by VF and least present in GJ.

3.5 eDNA analysis (soil samples)

After filtering, the eDNA samples contained 914 different species and a total amount of 405,616 reads. The majority of the reads came from fungi and clitellate worms, which reflects the biomass and DNA composition in the soil samples.

First, I created a non-metric multidimensional scaling (NMDS) ordination, to see similarities in the species communities of all subsampling sites (see Figure 21). The ordination plot displays the community composition of three different sites: GJ (blue), RF (red), and VF (green). The positioning of the points represents the similarity of species communities based on the NMDS analysis. A low stress value under 0.05 (here: $k=2$, stress=0.015), indicates a very good match of the NMDS presentation and the actual ecological distance in the data. I can therefore interpret the NMDS plot.

The plot indicates that the species community at site VF (green) is positioned closer on the ordination to site RF (red), suggesting more similarity between VF and RF than with site GJ (blue). Meanwhile, RF and GJ, despite being distinct, appear to be closer to each other than either is to VF, implying that the species communities at RF and GJ share more similarities with each other than with VF. However, the overall pattern shows clear differentiation of species communities between all three sites. The distribution of sites follows the gradient of modification (whereby GJ is the highest modification, followed by RF and VF as non-modified) I expected.

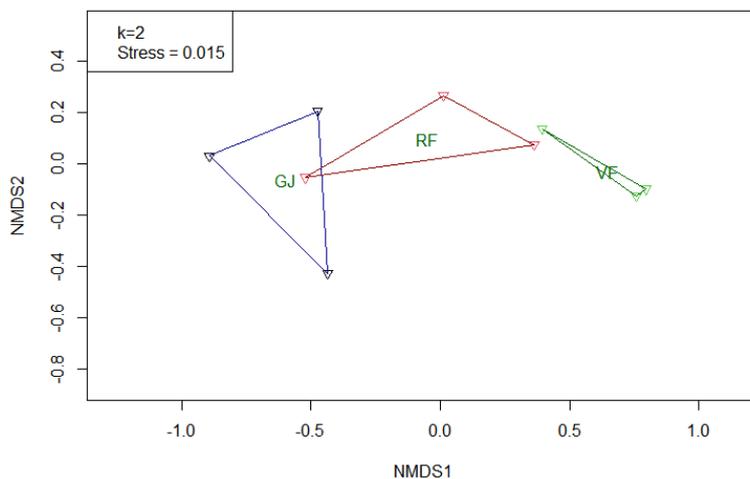


Figure 21. Non-metric multidimensional scaling (NMDS) ordination of species communities from soil sampled eDNA at the three different sites. Triangles are pointing out the locations of species community within the ordination space. RF site is located between the two other sites, indicating similarities to both of the sites.

I also generated a Venn diagram (see Figure 22) to visualise the common and unique species communities between the three sites. As all three are riparian zones in northern Sweden, there was a considerable overlap of 114 species (36.5%). The RF shows a remarkable similarity with the other sites: it shares 62 species (19.9%) exclusively with GJ and 77 species (24.7%) exclusively with VF. In contrast, VF and GJ share only 7 species (2.2%) that are absent from RF. This pattern confirms the results of the ordination analysis, where the species community of RF was positioned between those of VF and GJ. Specifically, GJ has 17 unique species (5.4%), RF has 8 (2.6%), and VF has the most unique species with 27 (8.7%).

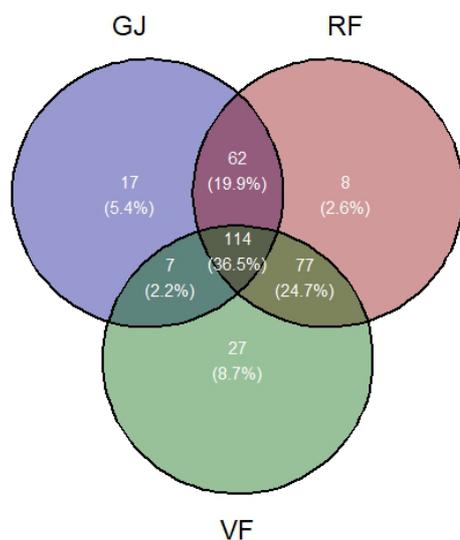


Figure 22. Venn Diagram, showing shared and unique species across all three sites.

To address **hypothesis 2)** and determine whether there is a statistical difference in species communities among the sites, I conducted a PERMANOVA using the dissimilarity matrix. The results revealed a significant difference in species communities between the sites ($F=2.667$, $p=0.005^{**}$). Approximately 47.1% of the variation in species communities ($R^2=0.471$) can be attributed to differences between the sites.

To test **hypothesis 4)** and to investigate the environmental factors (standardised mean and SD of NDVI, vegetation height, slope and flow) that influence the differences in species communities, I performed a Canonical Correspondence Analysis (CCA) (see Figure 23). The model explains 38.81% (proportion of constrained = 0.3881) of the variation in species communities. This proportion is represented visually on two axes in the ordination space: CCA1 (x-axis) with an eigenvalue of 0.7309 and CCA2 (y-axis) with an eigenvalue of 0.3699. The site labels indicate the locations of the species communities of each sample. The blue

vectors indicate the direction of influence of the environmental factors on the distribution of the species communities along the two axes, with the length indicating their relative influence. However, as there was only one environmental factor value per site (not for each subsite), it was not possible to statistically determine the most influential factor. Nevertheless, the vector orientations give an indication of possible correlations. Environmental factors with parallel vectors may have similar influences on species distribution. For example, NDVI mean and SD of flow are aligned with the CCA1 axis, suggesting that higher values of these factors may correspond to the observed gradient of species communities from VF to RF and then to GJ. Similarly, the mean and SD of vegetation height appear to follow this pattern. In contrast, the mean and SD of slope pointing towards GJ on the CCA1 axis suggest that lower values are associated with this site compared to RF and VF. As the mean and SD of NDVI are associated with CCA2, which explains less variance than CCA1, they describe the distribution along a gradient from RF to VF that becomes negative towards GJ.

PERMANOVA confirmed that CCA1 significantly accounts for variation in species communities ($df = 1$, $\text{ChiSquare} = 0.731$, $F = 2.527$, $p = 0.001^{***}$). CCA2, however, does not significantly influence species diversity ($df = 1$, $\text{ChiSquare} = 0.370$, $F = 1.2787$, $p = 0.206$). I could not test the environmental factors for correlation since I only had 3 data points per factor.

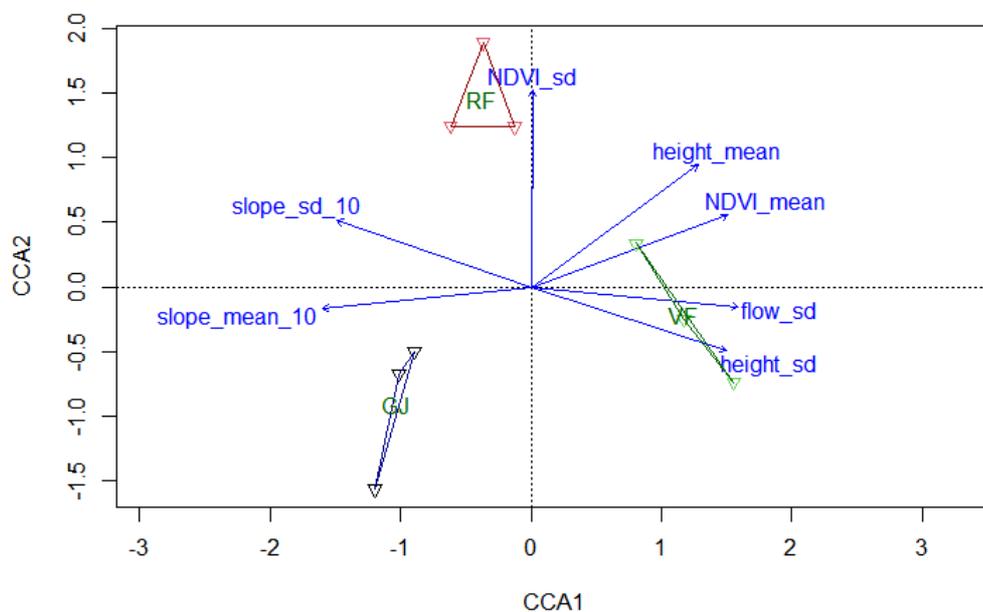


Figure 23. Canonical Correspondence Analysis (CCA) including difference in species communities and influence from environmental factors. Model explains 38.81% of species distribution with the environmental factors. Triangles representing a species community per subsampling site in the

ordination model. Blue vectors indicating the environmental factor which are influencing the species distribution.

The RF site has the highest biodiversity index from soil sampling ($H' = 3.519$, $1/D = 0.941$). GJ has the second highest biodiversity index ($H' = 2.587$, $1/D = 0.833$) and VF has the lowest ($H' = 2.466$, $1/D = 0.813$). To assess the differences in species communities between the three sites, I calculated Whittaker's beta diversity. VF and GJ show the greatest disparity in their species communities among the sites studied (beta diversity = 0.431), while VF and RF have the most similar species composition (beta diversity = 0.214). The species community diversity between GJ and RF also shows a relatively large overlap (beta diversity = 0.236). These values, also for each sample site, are additionally shown in Table 10:

Table 6. Species richness and Biodiversity indexes from soil sample eDNA analysis

	GJ			RF			VF		
	1	2	3	1	2	3	1	2	3
Total reads	133255			124279			148082		
	42694	38703	51858	37008	40991	46280	74827	35805	37450
Species richness	200			261			225		
	166	125	111	159	183	153	135	171	164
Shannon-index (H')	2.587			3.519			2.466		
	2.718	2.661	1.736	3.085	3.273	2.870	1.790	3.047	2.151
Simpson index ($1/D$)	0.833			0.941			0.813		
	0.869	0.861	0.686	0.900	0.958	0.877	0.491	0.903	0.700

4. Discussion

The aim of this study was to investigate riparian zones with varying levels of modification on their faunistic biodiversity, using various ecological field methods and environmental factors and to compare these with. The goal was to understand how modifications influence these ecosystems and to assess the potential of these sites for biodiversity.

4.1 Research questions and study results

The first hypothesis **(1)** aimed to explore the differences in environmental factors across riparian areas with different levels of modification.

My findings indicate that certain environmental factors indeed followed the anticipated gradient of modification, including the SD of flow, mean vegetation height, mean NDVI, and mean slope. Conversely, other factors, such as SD of height, SD of NDVI, and SD of slope, did not conform to the expected gradient. Notably, the SD of slope and vegetation height showed variations between modified and non-modified sites, whereas the SD of NDVI deviated from the predicted pattern and shows the greatest horizontal EH in the RF site.

The study revealed significant variances in the SD of vegetation height between the sites, highlighting the role of vertical environmental heterogeneity (EH) in distinguishing between modified and non-modified riparian zones in this study. As hypothesized, the natural site (VF) exhibited the highest SD of vegetation height, implying a more complex and potentially diverse habitat conducive to a variety of species. However, the absence of statistical significance in NDVI variability and low statistical power, to detect a significant difference for flow and slope SD suggest a complex interaction between EH and riparian zone modifications, deviating from the hypothesized gradient of modification.

Overall, the examined facets of EH (vertical, topographic, flow) proved effective in differentiating between modified and non-modified sites, yet they did not affirm the expected gradient in the degree of modification. While certain environmental factors, such as the mean vegetation height and NDVI, confirmed this gradient, the

horizontal aspects of EH did not support the hypothesis and the measurement EH was greater in modified site RF compared to the non-modified ones VF. One possible explanation for the higher variation in horizontal EH in RF is that the construction of roads in the riparian forest created zones with really low NDVI indexes with result in a higher SD taking the entire vegetation into account.

The second hypothesis **(2)** aimed to find a difference in faunistic diversity, expecting natural areas to exhibit higher diversity than modified areas. And the third hypothesis **(3)** should add the presence of indicator species for as additional evaluation of biodiversity. The fourth hypothesis **(4)** aimed to determine if these differences could be explained by EH and environmental factors.

The findings on biodiversity were mixed, indicating that modifications generally have a negative impact on faunistic diversity. The VF site showed the highest insect abundance and bat activity compared to the modified sites GJ and RF, supporting the assumption, that non-modified areas have a greater biodiversity.

While the abundance of insects followed also the degree of modifications (with VF as highest and GJ as lowest), for bats, the highly modified site GJ showed higher activity than the moderately modified site RF, contradicting expectations of the modification gradient.

The biodiversity index of bats suggested a more diverse bat community in the highly modified site than in the natural site, while the abundance was way higher in VF. I decided to highlight the bat activity as a better indicator, than the biodiversity indexes, due to the general low variation of detected species. This is consistent with a study by Renner et al. (2017), which found that increased vertical and horizontal EH positively affects bat diversity. Whereby I could only prove that the vertical EH is significantly higher in the non-modified site VF.

In contrast to the hypothesis that modified sites have a lower biodiversity, the biodiversity indexes for ground-crawling insects, insect traps as total, presence of indicator species as well as results from the eDNA analysis highlighted the site RF as most diverse, followed by VF and least diverse in GJ.

Identifying GJ as least diverse site, align with literature suggesting that habitat complexity fosters greater species diversity. Since for instance the horizontal EH was (even when not statistically significant) the highest in RF, before VF and the least in GJ.

With the addition of the environmental factors in the species community ordination, a trend that an increased variability of height and annual flow, together with an increased mean NDVI and vegetation height, combined with a decrease in mean

and variation in slope, organise the species communities from VF to RF to GJ, along the expected gradient. Whereby an increased variability of NDVI orders the communities from RF to VF to GJ, contrary to the expected gradient.

Combining the findings from the research questions, I can conclude that GJ shows the least EH and the least quality of habitat (e.g. productivity), together with the lowest abundances and biodiversity indexes in most of the biodiversity measurements. Pointing out GJ as the most modified sites and the least diversity.

Site RF however showed in some measurements a higher biodiversity than the natural site VF, together with a large variation in horizontal EH. Indicating that the modification still comes with a relatively high diversity but lower abundance. This is likely due to a more moderate modification compared to GJ and, that the with sand filled area was smaller and in contact with the surrounding forest. Moreover, the surrounding habitat might also influence these findings. Close to the study site are two protected nature reserves, a meander and peat lands.

4.2 Restoration potential of modified riparian zones

As I also found high insect biodiversity and several indicator species in RF, it is clear that the ecological restoration potential of the modified sites is high.

In the next section I will describe possible restoration measures that could be implemented on the sites. I will combine the main findings of my study with the restoration potential based on the restoration work carried out in previous studies for the RF and GJ sites. The methods mentioned could show an improvement for the local biodiversity.

The results suggest that EH is reduced in modified areas, and as it is likely that higher EH is correlated with higher biodiversity (Renner et al., 2017; Ricklefs, 1977; Stein et al., 2014; Stirnemann et al., 2015), it would be a good approach to target this. Planting native tree species and riparian plants could improve their productivity and diversity (González et al., 2015). Removing slopes and creating less steep slopes could be beneficial to increase contact with vegetation and water. Previous studies of restoration works have included the creation of floodplains to recreate the natural flooding of riparian zones. This was done by widening the channel and lowering the elevation of the surrounding riparian zones (Gumiero et al., 2013). This practice has had a positive impact on the nutrient cycling of river systems and riparian zones.

However, as riparian zones are driven by succession and disturbance, it could be beneficial to create artificial disturbances such as a spring flood, which would establish a greater diversity of smaller shrubs close to the river. Other disturbance methods used in forest systems such as forest fires, soil erosion or selective thinning could also be used.

In addition, soil preparation through the introduction of soil organic matter could be beneficial in creating a more natural state of the riparian zone. Deposition of dredged material has altered the soil and it is likely to be less nutrient rich than before modification. The incorporation of dead wood to improve and provide substrate for saproxylic insects and fungi could also be a good method in the study areas (Gumiero et al., 2013).

As the width of the riparian buffer has been shown to have a direct influence on vegetation, but also on faunistic diversity, it can be used to increase its size (Marczak et al., 2010; Lind et al., 2019).

Furthermore, I was able to show that the modified site RF has a high biodiversity, and I concluded that it is likely to have an impact on the surrounding environment, it could be a goal to increase connectivity to surrounding systems and improve the surrounding environment. This could be done by restoring meandering parts of the river with site arms and pools and riffles (Gumiero et al., 2013; Jähning et al., 2010), restoring nearby wetlands and peatlands (Gumiero et al., 2013), restoring secondary channels (Helfield et al., 2012) or generally improving water quality (Palmer et al., 2010).

It could also be actively attempted to improve faunistic diversity by installing bat and bird nest boxes to facilitate the establishment of species and their activity in the areas (Kennebecasis Watershed Restoration Committee, 2020). In addition, improving pollinator insects by planting flower meadows and placing "bee boxes" near the riparian zone can help increase faunistic diversity (Kennebecasis Watershed Restoration Committee, 2020).

4.3 Strengths, Limitations and Future Work

The practice of dredging riverbeds during dam construction and filling riparian zones with the dredged material was very common in northern Sweden about 60 years ago, and there are many of these modified riparian zones in the vicinity of hydropower plants. This is a largely unexplored research area with potential for improving these modified habitats.

The strengths of the study lie in its comparative approach and the inclusion of multiple environmental factors and biodiversity measurements. A great deal of time and effort has gone into making this possible.

In particular, the different approaches to measuring the faunistic diversity of different groups of organisms using different methods, the inclusion of indicator species and the discussion of different measures of biodiversity (abundance or indices) were good for getting a broad picture of the biodiversity potential of the sites. It was a strength and good to see that most of the biodiversity indicators measured pointed in the same direction.

Combining different biodiversity indices together with richness and abundance data and discussing them also highlighted the difficulty of measuring a complex system. Combining biodiversity results with environmental factors, rather than just measuring biodiversity, made this study more holistic and may reveal general patterns for riparian zones.

However, limitations such as the particular scale and resolution of different measures affected the statistical power for certain facets of EH and would have been beneficial to fully capture the complexity of these environments. The choice of metrics, while informative and appropriate in terms of time and resources, suggests that future research could benefit from a wider range of EH indicators and therefore more robust statistical analyses. Future studies should consider incorporating additional EH facets, such as soil type and moisture or dead wood, to provide a more holistic view of habitat complexity (Stein et al., 2014; Heidrichs et al., 2020). Alternatively, the use of alternative assessment methods such as HHS could also enrich the understanding of riparian habitat heterogeneity and its impact on biodiversity (Hekkala et al., 2023).

As river systems are generally highly variable and influenced by a wide range of factors, it would be extremely important to increase the number of study sites to identify common environmental drivers. In addition, experimental and long-term monitored studies could be conducted to see how riparian restoration from modified zones performs over time and which are the most effective.

The methods used to measure biodiversity may also have influenced my results. For example, camera trap data was difficult to validate because the ground in VF was covered with bushes and shrubs, whereas the ground in RF and GJ was more open. It would have been impossible to detect animals smaller than 30 cm in these covered habitats, and these accounted for most of the species detected at the RF site (e.g. red squirrel, mountain hare, various bird species). In addition, one of the cameras at RF was oriented towards the river and therefore captured (semi-)aquatic species, a perspective that was completely lacking at the other two sites.

In addition, the different sampling times of the insect traps could give a false picture of the biodiversity of the insect communities. Even when the traps were set for the shortest time in the VF, the abundance of insects was the highest. With an equally long sampling period, the biodiversity indices might have shown a different picture if other species had been detected.

4.4 Conclusions

In summary, this study reveals that riparian zone modifications impact environmental heterogeneity and biodiversity in complex and variable ways. Results pointing out that highly modified riparian zones show a reduced biodiversity and less complex environmental heterogeneity. However, modified sites do have potential for restoration work. Further work can build on these findings.

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Popular science summary

How bad is construction in riparian zones for terrestrial animals?

The short answer is that it depends on the scale of the change!

Scientists already know that the construction of dams and the dredging of rivers cause major problems for rivers, but also for riparian plants and animals. But what has not been studied so far, is how the construction of a new riparian zone affects environmental factors and surrounding biodiversity.

In this study, I compared the biodiversity of three different riparian zones:

- One without hydroelectric power plants or river modifications, and part of the Vormfors nature reserve as an "unmodified site",
- Another site where the riverbed was dredged, and the riparian zone filled with sand on a small scale near the dam of the Rusfors hydropower plant as "modified site" and
- One site where the dredging and filling took place on a large scale below the hydropower plant in Gallejaur as "highly modified site".

I compared vegetation height, plant productivity, flow and slope between the sites and was able to show that greater variation in plant height, less steep slopes, greater average vegetation height and vegetation productivity are more common in the natural site, slightly less in the modified site and are lowest in the highly modified site.

For biodiversity, I used camera traps to detect large animals, audio recorders to detect bats, insect traps and eDNA soil sampling. I found that the highly modified site had the lowest biodiversity overall, with both the natural and modified sites having clearly higher biodiversity (depending on the method used, it was higher in the natural or modified one). It seems that the surrounding environment also plays an important role, as the modified site is better connected with the surrounding forest and has some important ecological features (such as nature reserves, peat land and other water bodies) close to the studied area in comparison with the highly modified site.

In conclusion, the study shows that large modifications have a negative impact on ecosystems - maintaining a condition as close as possible to the natural state is the most effective for an improvement of modified areas.

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Appendix I

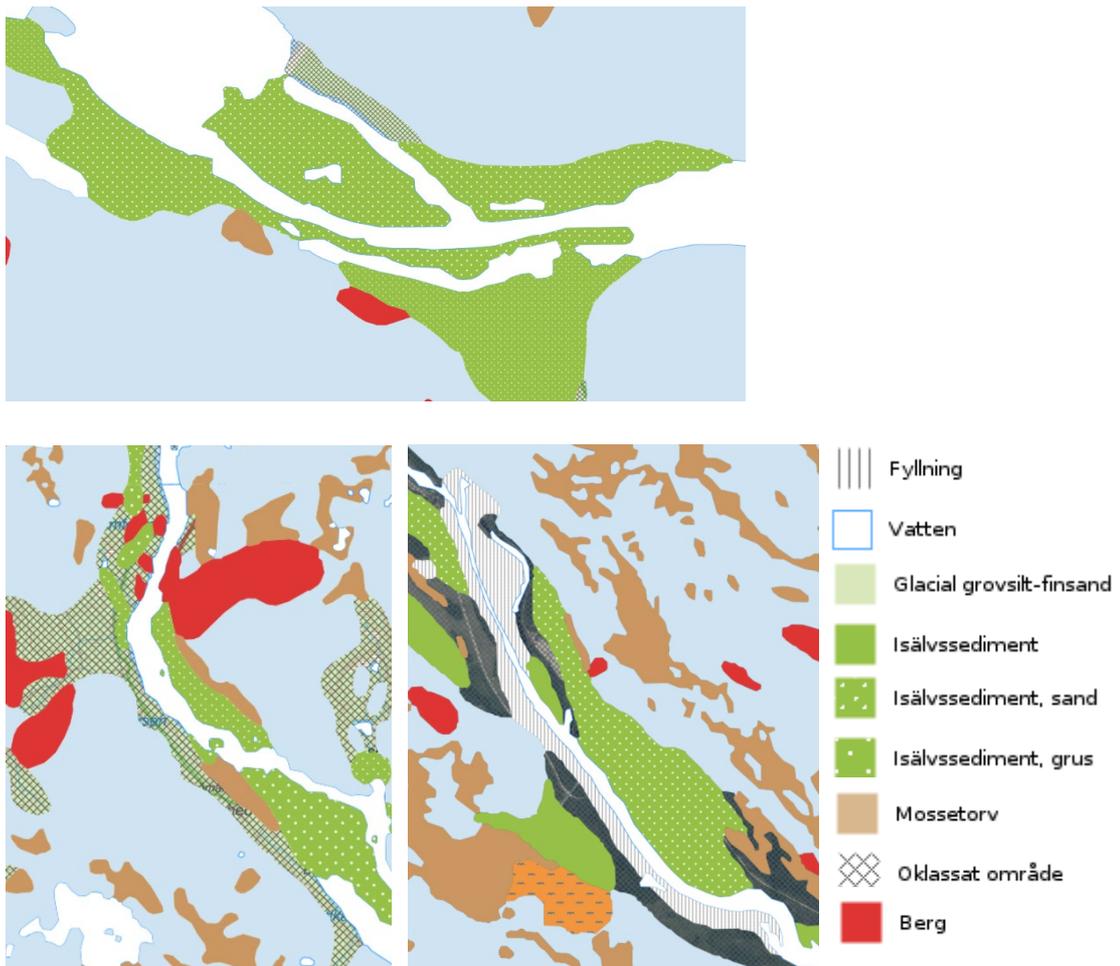


Figure 1. Map of soil types from Länsmäteriet (March 2024). Important: All maps have different scales.



Figure 2. Nature reserves close by RF site. Screenshot from Lantmäteriet: <https://minkarta.lantmateriet.se/> (07.03.2023)

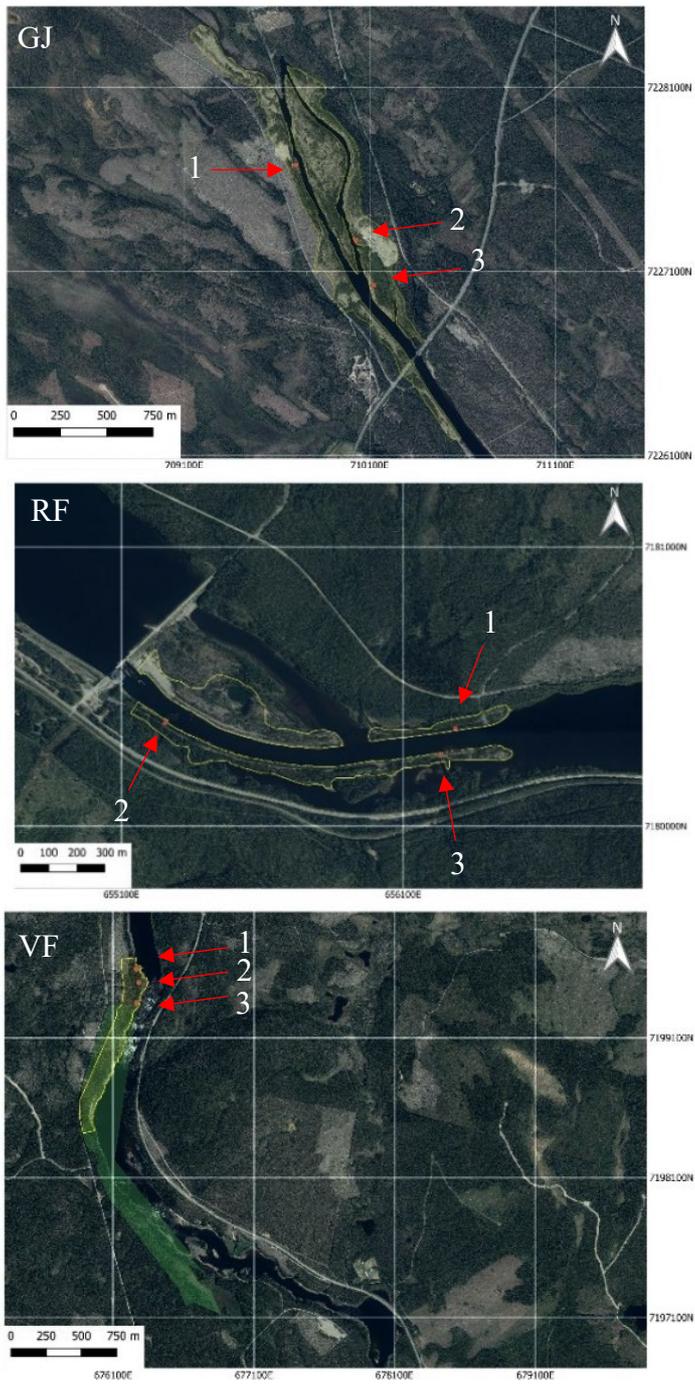


Figure 3. Subsampling plots with numeration used in the study (orange dots, marked with red arrows) at each study site.

Table 7. Overview: Time schedule fieldwork methods in summer 2023

		Start date	Date of removal	Sampling time (in days)
Camera traps	RF	12.06.2023	03.08.2023	52
	GJ	16.06.2023	03.08.2023	48
	VF	18.06.2023	02.08.2023	45
Insect traps	RF	12.06.2023	29.06.2023	17
	GJ	16.06.2023	29.06.2023	13
	VF	18.06.2023	28.06.2023	10
Audio recorders	RF	20.07.2023	03.08.2023	14
	GJ	20.07.2023	03.08.2023	14
	VF	19.07.2023	02.08.2023	14
Drone	RF	20.07.2023	/	1
	GJ	20.07.2023	/	1
	VF	19.07.2023	/	1
Soil sampling	RF	03.08.2023	/	1
	GJ	03.08.2023	/	1
	VF	02.08.2023	/	1

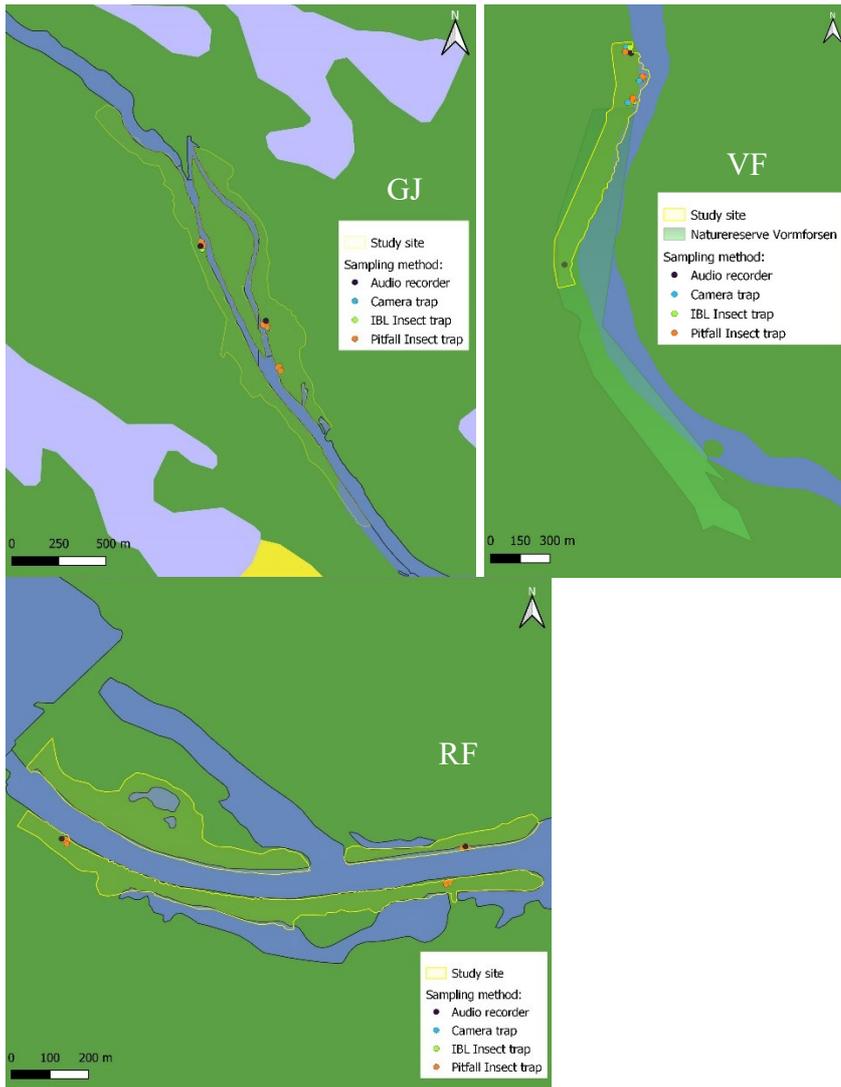
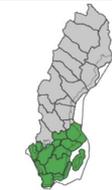


Figure 4. Locations of different sampling method within the study sites

Table 8. Bat Species in Sweden. Distribution maps from Artbanken.se, accessed in October 2023

Species name	English name	In Northern Sweden?	Distribution map
<i>Barbastella barbastellus</i>	Western barbastelle	no	
<i>Eptesicus nilssonii</i>	Northern bat	yes	

<i>Eptesicus serotinus</i>	Common serotine	no	
<i>Myotis alcathoe</i>	Alcathoe myotis	no	
<i>Myotis bechsteinii</i>	Bechstein's Bat	no	
<i>Myotis brandtii</i>	Brandt's myotis	yes	
<i>Myotis dasycneme</i>	Pond myotis	yes	
<i>Myotis daubentonii</i>	Daubenton's Bat	yes	
<i>Myotis myotis</i>	Mouse-eared myotis	no	
<i>Myotis mystacinus</i>	Whiskered Bat	yes	
<i>Myotis nattereri</i>	Natterer's Bat	yes	

<i>Nyctalus leisleri</i>	Leislers Bat		no	
<i>Nyctalus noctula</i>	Noctule		yes	
<i>Pipistrellus nathusii</i>	Nathusis' pipistrelle		yes	
<i>Pipistrellus pipistrellus</i>	Common Pipistrelle		no	
<i>Pipistrellus pygmaeus</i>	Soprano pipistrelle		yes	
<i>Plecotus auritus</i>	Brown Long-eared Bat		yes	
<i>Plecotus austriacus</i>	Grey Long-eared bat		no	
<i>Vespertilio murinus</i>	Particoloured bat		yes	

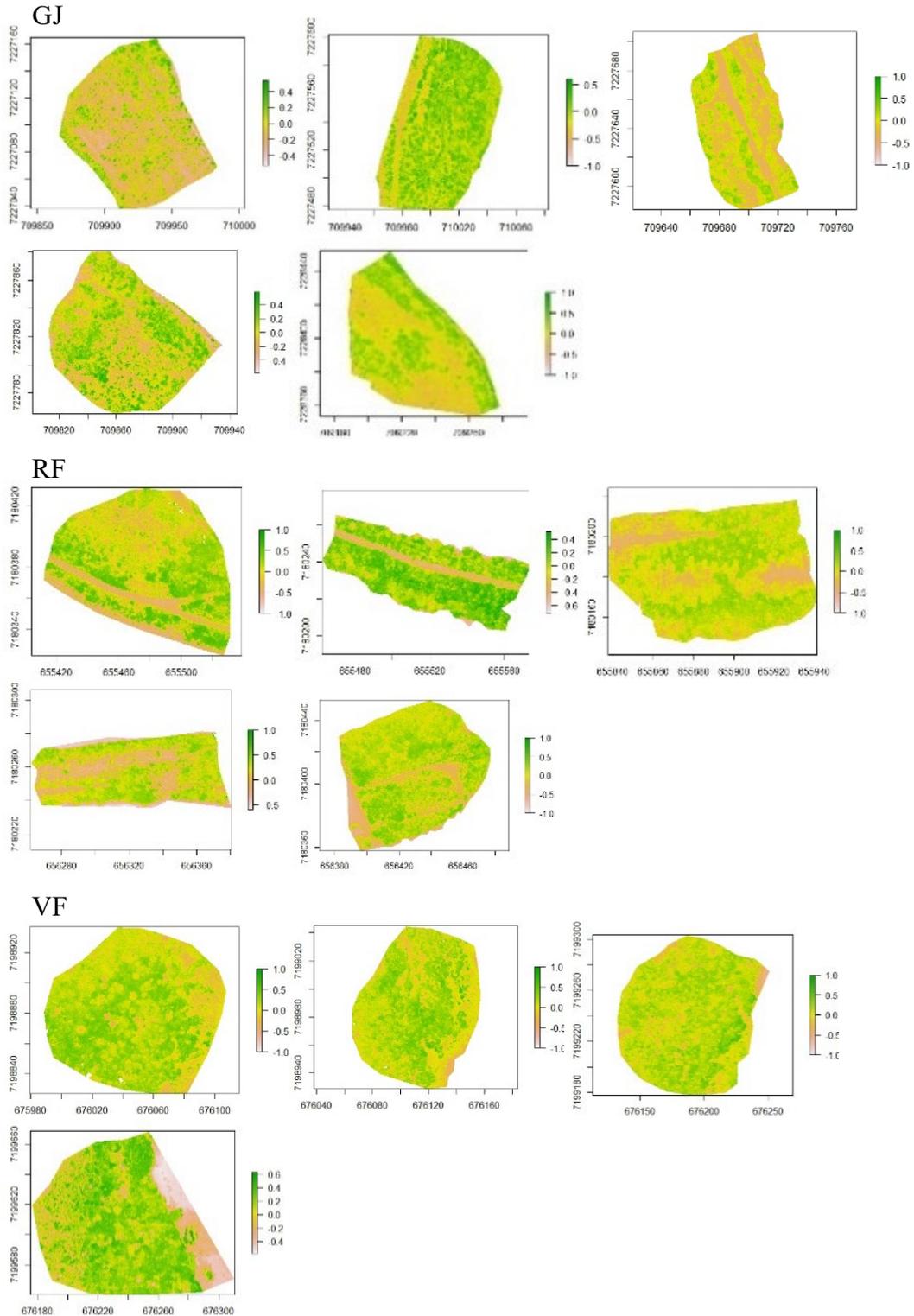


Figure 5. NDVI of each site (from top to bottom, left to right: GJ1, GJ2, GJ3, GJ4, GJ5, RF1, RF2, RF3, RF4, RF5, VF1, VF2, VF3, VF4)

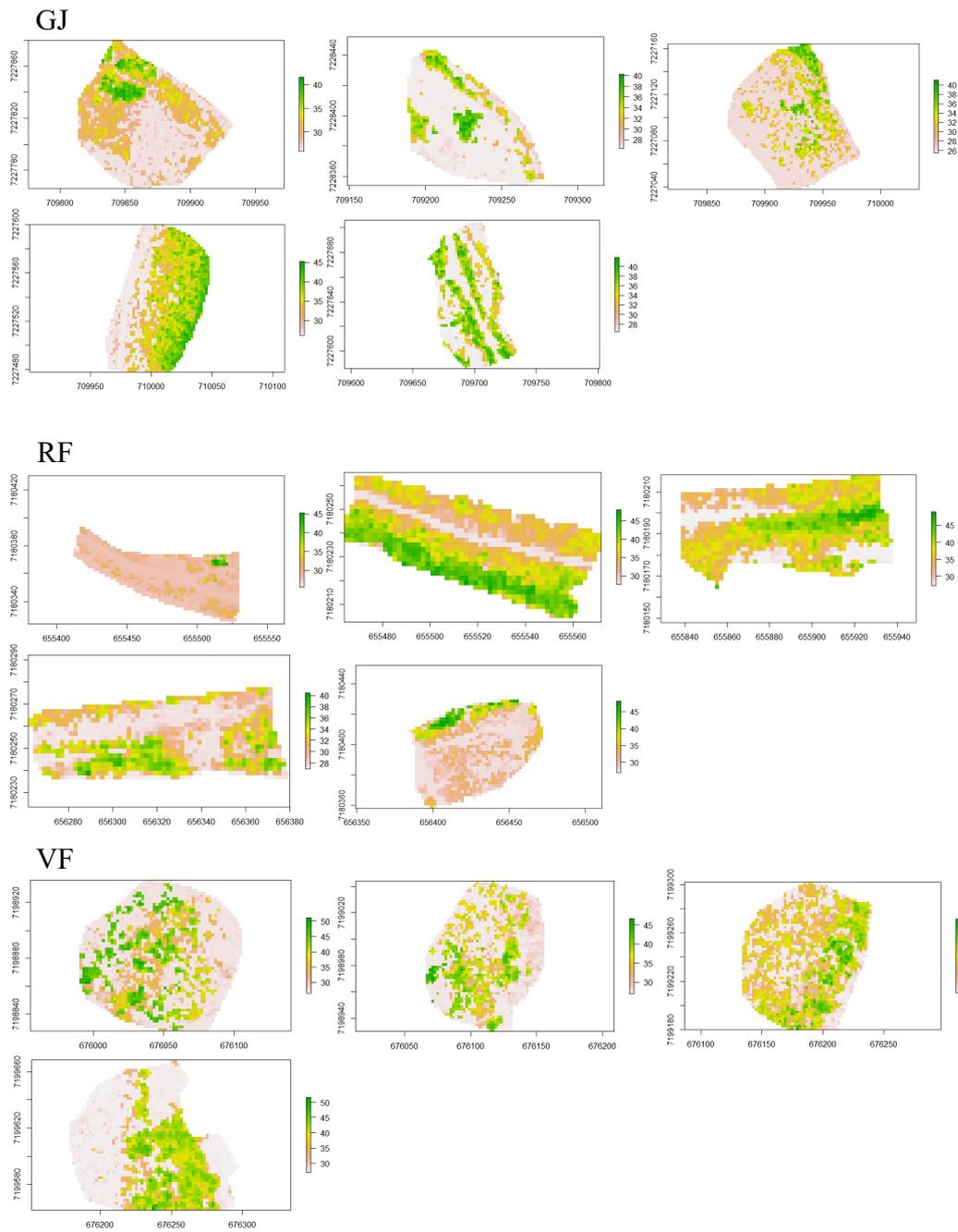


Figure 6. Vegetation height (from top to bottom, left to right: GJ1, GJ2, GJ3, GJ4, GJ5, RF1, RF2, RF3, RF4, RF5, VF1, VF2, VF3, VF4)

Table 9. Full list of detected animals from camera traps

Site	No	<i>Actitis hypoleucos</i>	<i>Branta canadensis</i>	<i>Capreolus capreolus</i>	<i>Cygnus cygnus</i>	<i>Lepus timidus</i>	<i>Podiceps cristatus</i>
		Common sandpiper	Canada goose	Roe deer	Whooper swan	Mountain hare	Great crested grebe
GJ	1	0	0	1	0	0	0
	2	0	0	0	0	0	0
	3	0	0	0	0	0	0
RF	1	1	4	1	1	0	1
	2	0	0	0	0	0	0
	3	2	0	0	0	1	0
VF	1	0	0	1	0	0	0
	2	0	0	3	0	0	0
	3	0	0	0	0	0	0

		<i>Rangifer tarandus</i>	<i>Sciurus vulgaris</i>	<i>Scolopax rusticola</i>	<i>Turdus pilaris</i>	<i>Vulpes vulpes</i>	Species Richness
		Reindeer	Red squirrel	Eurasian woodcock	Fieldfare	Red fox	
GJ	1	10	0	0	0	0	2
	2	1	0	0	0	1	2
	3	0	0	0	0	0	0
RF	1	0	2	1	1	1	9
	2	0	0	23	10	0	2
	3	2	0	0	4	2	5
VF	1	2	0	0	0	0	2
	2	3	0	0	0	0	2
	3	2	0	0	0	0	1

Table 10. Full overview of detected taxonomic arthropod groups at each site. * marks group, which was investigated as indicator species for riparian zones or ecosystem functioning

Species	GJ	RF	VF
<i>Acari</i>	1	0	0
<i>Apoidea</i>	1	0	2
<i>Araneae</i>	107	134	99
<i>Carabidae*</i>	9	18	11
<i>Cerambycidae</i>	4	1	12
<i>Coccinellidae</i>	0	0	1
<i>Coleoptera_other</i>	39	50	120
<i>Collembola</i>	2	107	80
<i>Curculionidae</i>	7	19	3
<i>Diplura</i>	3	0	0
<i>Diprionidae</i>	0	0	2
<i>Diptera</i>	69	178	172
<i>Ephemeroptera*</i>	13	5	15
<i>Formicidae</i>	241	94	301
<i>Hemiptera</i>	2	2	0
<i>Heteroptera</i>	4	8	2
<i>Hymenoptera</i>	5	9	15
<i>Lepidoptera</i>	0	4	3
<i>Neuroptera</i>	0	2	0
<i>Opiliones</i>	0	22	61
<i>Plecoptera*</i>	0	1	0
<i>Scarabeaidae</i>	2	0	0
<i>Scolytidae</i>	13	20	34
<i>Silphidae</i>	1	0	0
<i>Staphylinidae*</i>	13	60	56
<i>Sternorrhyncha</i>	15	8	10
<i>Trichoptera*</i>	4	13	7

Appendix II



Project report *DNA metabarcoding of soil samples*

Contractor

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Client

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Pia Knobloch, Navinder Singh

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Summary

Metabarcoding of DNA from environmental samples is broadly used for assessment of biodiversity. In this project invertebrate communities were analysed in soil samples, provided by the client. This technical report describes the methodology and provides the overview of the results, delivered in separate files.

DNA extraction

Soil samples were frozen and shipped on the dry ice, 9 samples in total.

DNA was extracted from 10 g starting material with QIAGEN DNeasy Power Maxi Soil kit, following the manufacturer's protocol. A blank extraction (without any soil material) was carried along with samples to control for contamination. DNA concentration was measured using Qubit fluorometer and ranged from 0.9 to 15.2 ng/µl (Excel Table 1). No measurable DNA was detected in the blank extraction.

Library preparation

Fragments of the mitochondrial COI gene is the most common marker for metabarcoding of invertebrates. We intended to use two pairs of primers, targeting the COI barcoding region: BF3 and BR2, recommended for terrestrial arthropods by Elbrecht et al. (2019), and universal metazoan primers mICOIntF (Leray et al. 2013) and jgHCOI2198 (Geller et al. 2013). The latter primer pair amplifies a broad range of metazoans and is used in the ARMS-MBON program for metabarcoding various marine invertebrates, including Arthropoda (Obst et al. 2020). They have been also used for metabarcoding arthropods in soil samples (Kirse et al. 2021). The primers were ordered from Eurofins Genomics with extensions (overhangs) complementary to the primers used in the index PCR.

The PCR amplifications with the BF3-BR2 primers was performed in 30 µl volume, containing 15 µl KAPA HiFi HotStart ReadyMix (Roche), 1 µl of each primer at 10 µM concentration, 0.5 µl of bovine serum albumin 20 mg/ml, 7.5 µl PCR-grade water and 5 µl of template DNA. PCR cycling consisted of initial denaturation 95 °C for 5 min followed by 30 cycles with 95 °C for 20 s, 46 °C for 30 s and 72 °C for 1 min, and a final extension at 72 °C for 7 min (based on the protocol in Elbrecht et al. 2019).

The PCR amplifications with the mICOIntF - jgHCOI2198 was performed following the protocols developed in the ARMS-MBON program. The reverse

primer contains inosine bases that interfere with some polymerases; it is recommended to use PCR reagents from KAPA Taq PCR kit with dNTPs (Roche). PCR reactions had 30 μ l volume, containing 3 μ l KAPA Buffer A, 0.6 μ l of $MgCl_2$ 25 mM, 0.8 μ l of dNTPs 10 mM each, 0.9 μ l of KAPA Taq 5U/ μ l, 1.8 μ l of each primer 10 μ M, 0.6 μ l of bovine serum albumin 20 mg/ml, 15.5 μ l PCR-grade water and 5 μ l of template DNA.

Touchdown PCR cycling consisted of initial denaturation 95 °C for 5 min followed by 16 cycles with 95 °C for 10 s, 62 °C (-1° per cycle) for 30 s and 72 °C for 1 min, then 24 cycles with 95 °C for 10 s, 46 °C for 30 s and 72 °C for 1 min, and a final extension at 72 °C for 7 min (based on the ARMS-MBON protocols at https://github.com/armsmbon/documentation/tree/main/standard_operating_procedures).

After amplifications PCR products were visualized using gel electrophoresis. While the universal COI primers produced a clear band of the expected size, the band produced by BF3-BR2 was visible but faint, indicating low efficiency of PCR. The attempts to optimize the PCR by changing the PCR mix and adjusting primers and template concentrations did not improve it.

To reduce random variation in the amplification, two PCR reactions were performed for each primer pair and combined. To control for contamination during the PCR stage, negative controls (a PCR reaction where template DNA is replaced by water) were included in each PCR. PCR were also performed for the blank extraction. For the universal COI primers, we included a positive control - a samples containing DNA from eight marine invertebrate species.

The PCR products were purified with AMPure XP beads (Beckman Coulter) using the Illumina protocol (https://support.illumina.com/documents/documentation/chemistry_documentation/16s/16s-metagenomic-library-prep-guide-15044223-b.pdf). The DNA concentrations were measured using Qubit fluorometer and the PCR products were sent to Eurofins Genomics, who performed the index PCR and sequencing.

Sequencing

Index PCR was done by Eurofins Genomics according to their protocols. For the PCR fragments with the BF3-BR2 arthropod primers the index PCR failed for 8/9 samples, most likely due to too low DNA concentrations of the PCR products. Index PCR also failed for the blank extraction and negative controls, confirming no contaminations.

For the PCR fragments with the COI universal primers index PCR worked well for all nine samples as well as for the positive control, and they have been sequenced at the Illumina MiSeq 2 x 300 bp platform with minimum 60 000 reads per sample.

Bioinformatic analyses

Sequence reads were demultiplexed by the sequence center. The quality of the raw sequences was accessed using *FastQC* tool. Remaining adapters and low-quality bases were removed with the *cutadapt* tool (paired-end mode `cutadapt -b CTGTCTCTTATA -B CTGTCTCTTATA -q 20,20 -j 0 -m 200`) and quality of the filtered reads was controlled with *FastQC*.

Trimmed sequences were further processed using DADA2 R package (Callahan et al. 2016) to remove the COI primers and reads shorter than 177 bp, to correct sequencing errors and to merge forward and reverse reads. Finally, chimeric sequences and sequences shorter than 310 bp and longer than 318 bp were removed (for most of the species the length of the amplified COI fragment is expected to be 313 bp).

For all unique sequences, or ASVs (amplicon sequence variants), taxonomy was assigned using the BOLD database (www.boldsystems.org/) and BOLDigger tool (Buchner & Leese 2020). BOLDigger compares each ASVs to the database using *blastn* algorithm and summarize the results of 20 hits per ASV.

Summary of the results

Number of the raw reads per sample was between 68 600 and 87 500. After all filtering steps 43 500 - 77 700 clean reads per sample remained (survival rate 52.3 – 88.8 %). Similar number of reads (50 600) was obtained for the positive control. In the positive control, 99.1% of the sequences came from the control species and all eight species were correctly identified. Overall, this indicate that metabarcoding and sequencing worked well.

In the samples, only 0.5 – 5.9% of reads were assigned to insects. The majority of the reads came from fungi and clitellate worms, which is likely reflects the biomass and DNA composition in the soil samples. Low insect DNA content in the present soil samples might also explain low amplification efficiency with the arthropod primers. In the original paper by Elbrecht et al. (2019) the performance of primers was evaluated for the mock community (sample of mixed insects), and these primers may be more suitable for the Malaise trap samples. Nonetheless, 1 382 ASVs came from phylum Arthropoda; of them 377 from Insecta, 222 from Arachnida and 132 from Collembola.

Many ASVs were assigned to Arthropoda with no further classification available. Those predominantly come from private records in the BOLD database, when the authors were not able to provide any further information. These unidentified sequences may also come from fungi and other organisms associated with arthropods.

Finally, some sequences in the samples were identified as marine algae: brown alga *Chorda asiatica*, red alga *Acanthosiphonia echinata*, raphidophyte alga *Chattonella minima* and brown alga *Halosiphon tomentosus*. *Chorda asiatica* sequences were also found in the control sample. The first three species are native to Asia or Northwest Atlantic, and none of these algae ever been found in any samples previously processed in our lab. Therefore, a likely source of contamination is “index jumping” during the index PCR when the samples were processed together with other metabarcoding samples, and some could be marine algae.

Data delivery

Technical information about the samples (DNA concentration, number of raw and filtered reads etc.) are in the Excel Table 1.

Excel Table 2 contains the main results of metabarcoding analyses: taxonomic information for each ASVs and its occurrence (number of reads) per sample. The presented taxonomic information is based on the first-hit summary function in the BOLDigger.

Excel Table 3 contains the information of all hits (20 per sample) in the BOLD database. It can be used to check similarity between different hits for ASVs of interest, whether they come from the private or public collections etc.

ASV sequences in the fasta format are provided in a separate file. They can be used for *blastn* searches in the NCBI-nt database etc.

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