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Diploma thesis

Effect of vegetation loss on freshwater diversity

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DIPLOMA THESIS ASSIGNMENT

Bc. Anna Marie Poskočilová

Engineering Ecology
Nature Conservation

Thesis title

Effect of vegetation loss on freshwater diversity

Objectives of thesis

We are observing a significant loss of biodiversity in general, all over the world, by the era of the 21th century. Research will be focused on freshwater invertebrates and amphibians ranging fish ponds at the area of the Czech republic.

It is observed that the fish pond management practices causes massive loss of littoral and aquatic vegetation. Strong fish predation has also negative impact on inhabitant populations. The consequence may be crucial for the presence of freshwater invertebrates and amphibians.

The main objective of diploma thesis is to investigate the effect of vegetation enrichment on the diversity of amphibians and freshwater invertebrates in fish ponds.

Methodology

Field research is focused on four areas in the Czech republic. Those areas are already under the long term research. The data will be gathered during the season in 2019. We will build 32 cages (2x2m) on 16 fish ponds. The cages should reduce or even stop the presence of fishes. We will add some water plants at the water level inside the cages to attract the animals.

Nymphs and imagoes are going to be captured by sweep net and live catch trap in the interval of 3 weeks.

Research schedule:

- March to May 2019: compilation of materials
- June to September 2019: data sampling
- October to December 2019: data preparation and analysis

• December 2019 to April 2020: creating of diploma thesis

The proposed extent of the thesis

30 – 40 pages + attachments

Keywords

freshwater invertebrates, diversity loss, fish predation

Recommended information sources

Dolný A., Bárta D., Waldhauser M., Holuša O., Hanel L., et al. (2007). Vážky České republiky: Ekologie, ochrana a rozšíření. Český svaz ochránců přírody Vlašim, 672 s.

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Dean

Prague on 20. 03. 2020

Declaration

I declare that I have a diploma thesis on the topic: Effect of vegetation loss on freshwater diversity I prepared and quoted all information sources that I used in the work and which I also listed at the end of the work in the list of used information sources.
I am aware that my diploma thesis is fully covered by Act No. 121/2000 Coll., On Copyright, on Rights Related to Copyright and on Amendments to Certain Acts, as amended, in particular the provisions of § 35 para. 3 of this Act, ie on the use of this work.
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By signing, I also declare that the electronic version of the work is identical to the printed version and that the data provided in the work was handled in connection with the GDPR.

In Prague, 30 March 2020 ....

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In Prague, 30 March 2020 …………………………

Abstract

Agricultural intensification is the main cause of loss of aquatic invertebrate species in farm ponds. The vast majority of our ponds are used for fish breeding, predominantly carps, and are often overfished. Homogenous fish stock and minimal cover of littoral vegetation negatively affects the population of aquatic invertebrates. The literature review summarizes the issue of intensive agricultural management and its impact on aquatic invertebrates. Furthermore, it analyses the influence of aquatic vegetation on aquatic invertebrates and equally summarizes the defense antipredation mechanisms of aquatic invertebrates. The experiment took place at twelve ponds used for intensive fish farming in three regions of the Czech Republic. The aim was to create alternative fish-free zones within homogeneous ponds (using fences) and plant vegetation artificially in half of them. Sampling took place throughout the season of 2019 (April-October). The experiment has demonstrated that only several invertebrate groups proved more abundant in cages without fish (Gerridae, Notonectidae, Noteridae), and that aquatic Hemiptera were attracted to microhabitats with natural vegetation. Moreover, the experiment has shown that species composition did not differ significantly between the researched localities. All the groups captured during the field experiment were pioneer species, which tend to quickly colonize new environments, and they were not particularly selective in their habitat preferences.

key words: freshwater invertebrates, diversity loss, fish predation, freshwater ponds, vegetation loss, intensive management

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1 Introduction

This diploma thesis responds to the current problem of intensive agricultural exploitation and habitat homogenization of our landscape. Specifically, it deals with the impact of intensive fish stock and minimum littoral vegetation in our ponds in relation to aquatic invertebrates.

In the past, the way we managed our ponds was significantly more extensive. The ponds were not so eutrophicated, the fish stock was more heterogeneous, the extraction of the pond was not so frequent. A turning point occurred in the second half of the 20th century, when greater intensification began (Andreska, 1987).

The vast majority of our ponds are breeding, often overfished, with carp predominating. Predatory pressure on the aquatic invertebrates is enormous, and because of the lack of littoral vegetation, the aquatic invertebrates have no shelters (Janda *et.al.,* 1996).

However, most of them have developed a number of antipredatory mechanisms. Good swimmers escape from fish by active swimming (Johnson, 1991), others hide in the mud (Thompson, 1987). Some can jump out of the water and fly away (Kovac and Maschwitz, 1991), others have a cryptic color and are almost invisible to fish (Del-Claro and Guillermo, 2019). Some aquatic invertebrates even excrete repulsive substances or are overgrown with spines that discourage fish (Cordoba-Aguilar, 2008). Despite these sophisticated defense mechanisms, the aquatic invertebrates are the main food source for carp in our ponds. And although they are equipped with various defense strategies against fish predators, they struggle in habitats where predation pressure level is so high.

The *in situ* experiment was established to analyze whether small isolated habitats with rich vegetation could be more attractive to aquatic invertebrates in comparison to open homogeneous ponds. Intraspecific and interspecific interactions between aquatic invertebrates are not well researched and I believe that this work could be beneficial in this area.

1.1 Main questions and goals

As far as we know the presence of the littoral vegetation is crucial for the abundance and diversity of freshwater invertebrates.

Are we able to reverse the decrease of freshwater invertebrates using the tools of specific management? Is it possible to increase the number of freshwater invertebrates in the artificial fishponds by planting vegetation and eliminating fish stock at just a small area of the pond? Do all of the taxonomical groups react the same way to the intensity of the fish stock or the presence of littoral vegetation? Or is there any diversification among the taxonomical groups?

2 Theory section

2.1 The importance of the freshwater ponds

Freshwater habitats make up only 0,01 % of the Earth's waters and 0,8 % of the Earth's surface. As far as we know, there are 100 000 species bound to the freshwater habitats. It is almost 6 % of all the described species in the world. The inland waters have also a great value for all humans. We are being attracted by those habitats not only for using water as a natural resource, but it also has a cultural, aesthetical, economical and scientific value (Dudgeon *et al.*, 2006)**.**

The proper management and conservation are crucial for the preservation of those precious spots and it should be in the interest of each human, nation and government to protect them. Yet the freshwater ecosystems are now being in a deep crisis. There is a massive loss of biodiversity and it is far greater than in the most affected terrestrial ecosystems (Céréghino *et al.*, 2014).

What are the main problems causing the global freshwater biodiversity threats? They are overexploitation, water pollution, flow modification, destruction or degradation of habitat and invasion by exotic species. Their effect causes population decline and reduction of freshwater biodiversity all over the world (Dudgeon *et al.*, 2006).

The inland waters constitute a complicated network that is dynamic and connected. That means that it is important to protect not only the downstream areas such as ponds, swamps etc. but also the upstream rivers and springs and their surroundings. The freshwater habitats are nowadays threatened by a reduction of connectivity that leads to the isolation and rupture of migration corridors (Oertli *et al.*, 2005). It is necessary to make a trade-off between the human use of ecosystems and the conservation of freshwater biodiversity (Dudgeon *et al.*, 2006).

To define ponds - they are small, artificial or natural-like, shallow waterbodies. They are strongly diversified (size, shape, vegetation and fish structure, trophism) by their location and usage, so it increases the beta diversity of the area. Water bodies such as ponds increase the biodiversity at a regional level, because they are able to host many unique, endangered and rare species unlike other water-body types (Oertli *et al.*, 2005).

They are key habitats for many taxa and they are considered to be biodiversity hotspots. Some taxa called ‘flagship groups’, connected uniquely to ponds, are known. They are mostly amphibians, dragonflies and aquatic plants. All of these groups include many critically endangered species. There is not much known about ecological processes in ponds and the lack of knowledge leads to the degradation of these habitats (Céréghino *et al.*, 2014).

Despite the fact, the temporary ponds have been recognized as a wetland type of the international importance by the Ramsar Convention, there is no substantial concern to perceive them as an important part of water ecosystems. Numbers of monitoring programs took place in European states, yet they failed to include ponds as well. To improve the recent situation concerning ponds, the research needs to be coordinated, public needs to be more educated, the pond management must be carried out gently and the environmental policy should be updated (Céréghino *et al.*, 2014).

Many ponds have been lost because of drainage due to the agricultural intensification or urbanization. As result we can now see an increasing fragmentation and isolation of freshwater ecosystems. Most of ponds are now being highly enriched by the nutrients. It causes a poor habitat and water quality which allows the invasive species to spread (Hill *et al.*, 2016).

2.2 History of pond management in the Czech Republic

2.2.1 Development of fish breeding

There is not much information concerning the fish species in farm ponds in the past. Ponds were formally used only as a storage of caught fish. The first indication of carp breeding is dated up to 1450. They placed adult fish that was suitable for reproduction to the pond, then they left it for 6 or more years and after that they finally fished it out. The largest fish were caught to consume and the smaller were relocated to other ponds (Pechar, 2000).

This process was later replaced by separated breeding of fingerlings and adult fish. The ponds were divided into three categories. The first one was to breed fingerlings, the second one was for young fish (2-3 y. o.) and the main category was formed by ponds with adult fish used for fishing and market. Until the early 16th century there were no other species but carp bred in our ponds then the pike was added. Thanks to frequent updates of the methods of breeding, carp breeding started to be very effective. The production of fish grew continuously and even some new species appeared in our ponds (*Tinca tinca, Perca fluviatilis, Abramis brama*) (Badinová, 2007).

Due to the historical consequences – Thirty Years' War, many people died and the rest of them were in poverty and misery. It negatively affected the pond management in the area. In 1620, people completely stopped fishing and taking care of ponds. It led to the pike overbreeding situation. The ponds overgrew with vegetation and the productivity of ponds continuously declined (Andreska, 1997).

In the 18th century a very important management practice - summer drainage - appeared for the first time. During the vegetation season the pond is drained. That makes an opportunity for the vegetation of muddy bottoms to grow, the littoral vegetation can regenerate (Francová *et al.*, 2019a). It positively effects waders and other birds, the sediments can mineralize and there are more nutrients that should increase a production of fish. It was common that pikes were added to the ponds. A great attention was paid to *Tinca tinca* and *Lota lota* breeding. The second half of the 18th century brought the first mention of using yard dung to make a pond more fruitful. Consequently, there were two ways to increase a productivity of ponds - summer drainage and yard dung addition. Later more of them appeared. It was for example removing forest edges off the ponds neighborhood and replacing them with grass fields. That should have allowed a nutrient runoff from fields into ponds (Andreska, 1997).

During the mentioned era fish breeding was mostly based on achieved skills and experience. But the results were often off balance. There was no basic information about the biology of fish, especially about the feeding requirements. No eligible agricultural interventions, that would positively effect breeding fish were done. The knowledge was based only on empirical local findings. As an example - a newly built pond was more productive than the old one (that is the reason why summer drainage took place) (Pechar, 2000).

The research of Josef Šusta (1835-1914) helped to make the pond management and the fish production more effective. His research showed the importance of freshwater organisms as a food source of carp. Šusta considered carp to be a suitable fish for breeding, because carp feed on natural food as well as food added to the pond (Šusta, 1884).

In the second half of the 20^{th} century there was a huge change in the pond ecosystem caused by the agricultural intensification. Until then the fish production was quite low. Most of the ponds were slightly acid, and the fish stock was more heterogenous. But the number of individuals was rather low. The water in ponds was more transparent and the coverage of macro vegetation was higher. This fact enabled the biodiversity of pond ecosystems to increase. The great agricultural intensification in the second half of 20^{th} century completely changed the conditions in ponds. More nutrients were being artificially added to the ponds. That caused a higher productivity of fish, and consequently it changed the water chemism. As a result, the biodiversity of water organisms decreased. Further, in 1952 duck breeding in ponds started. That led to even higher eutrophication of water and edges of ponds (Pechar, 2000).

2.2.2 Breeding situation nowadays

Ponds are usually extracted once a year or once in two years. The total fish production counts 17 000 tons of carp per year (500 kg/ha). It is 20 000 tons in total (carp, pike, silver carp, grass carp, tench). There are many technologies used to reach these high numbers. Young fish (up to 1 y. o.) are artificially reproduced in laboratories. Fishermen add extra nutrients and extra food for carp into ponds. But it causes that the ponds are overcrowded, the transparency of water is lower as well as the level of oxygen and amount of light (Badinová, 2007).

The technology of carp breeding and reproduction is well developed. Carp accepts grains as a source of energy, that is cheap and effective. It is tolerant to poor water quality (level of oxygen, transparency) and it survives well in overcrowded ponds. Most of the ponds are private and designed for business. The owner's duty is to take care of the pond but the outcomes on operating and maintaining ponds are high. There are several ways to make money running a pond: primarily it is selling fish, then receiving subsidies from the government and marginally providing recreation activities like sport fishing (Badinová, 2007).

2.3 Processes in planktonic and zoobentic communities

2.3.1 Stability disturbance in planktonic communities

Nowadays, the increasing trophic level in ponds affects the amount of phytoplankton, its species composition and biological activity. Photosynthesis of algae and cyanobacteria strongly influences two main factors determining the plankton stability. It is pH and the concentration of dissolved oxygen. The artificial nutrient income increases the intensity of respirating processes of the bottom and pelagic organisms (Pechar, 2000)

Today's fish breeding practice tries to retain the amount of fish individuals in the pond as high as possible. As the result, the predation pressure of fish causes the elimination of zooplankton, mostly *Daphnia*, especially the biggest individuals. Big *Daphnia* represent very effective filtrators, they are able to reduce the growth of small phytoplankton. If there were no *Daphnia*, there would be a lot of phytoplankton (Pechar, 1995). In today's ponds there are also lots of nutrients so algae and cyanobacteria can produce very dense biomass grow (Francová *et al.*, 2019a). The described processes are connected to March and April when the temperature does not exceed 12 degrees. That is the period during which the photosynthetic activity of phytoplankton can increase the water pH up to 10. This procedure realizes because of high phytoplankton biomass, sufficient nutrients available, good light conditions (day length, light intensity) along with lower respiration processes of the entire planktonic community and sediments. While the photosynthesis is not slowed down in low temperatures, the respiration is highly temperature dependent (Pechar, 1995). The respiration is basically lower in winter and early spring. During photosynthetic processes the alkalization of water occurs. While the respirating processes release CO_2 and cause acidification. The considerable prevalence of photosynthetic processes over respiratory in spring causes the frequent high level pH fluctuations. The fish population which is often weakened after hibernation, may be affected by gill necrosis due to an increased pH often up to 10 (Janda *et.al.,* 1996).

If the fish stock is able to survive this period successfully and its feeding pressure further eliminates a large filtering zooplankton (*Daphnia*), an intensive phytoplankton development occures in the summer. This time of the year the water temperature rises above 16 °C and the respiration processes increase in both planktonic and sediment communities (Janda *et.al*., 1996). In early summer the availability of nutrients, ammonia and phosphates increases and are apparently intensively released from sediments. What is more, the dung is usually added to the pond. This huge nutrient supply significantly stimulates the development of phytoplankton. At the same time, the intensity of the respiratory processes increases. It causes a paradoxical drop in pH values during the season, despite the fact that phytoplankton biomass increases rapidly. The intensive respiration in the bottom, littoral and open water causes the oxygen concentration drop to critical levels during just a short period of time (Hlaváč *et al.*, 2014).

During the summer high phytoplankton biomass causes a decrease in the concentration of available nutrients. Nitrate and often ammonia concentrations fall below the detection limit of commonly used analytical methods. In contrast, the phosphate concentration is generally maintained at a sufficient level. Due to the enormous development of phytoplankton, the level of underwater light is lower. The transparency of the water decreases up to 10 - 20 cm. Low light intensities and lack of inorganic nitrogen are very suitable conditions for the development of planktonic cyanobacteria. The presence of them causes hygienic and toxicological problems (Janda *et.al.,* 1996).

If fish are temporarily weakened or even killed as a result of gill necrosis, oxygen deficiency or infectious or parasitic diseases, then large species of *Daphnia* are rapidly prevalent in zooplankton. Large *Daphnia* are able to eliminate phytoplankton by their filtration activity. Achieving a condition when a large filtering zooplankton eliminates the development of phytoplankton brings the risk of oxygen deficit again. The oxygen consumption for the decomposition of large quantities of organic substances (supplied by fertilization) in the bottom is so great that the minimal photosynthetic production of suppressed phytoplankton is not big enough to cover this consumption. Equally, the oxygen diffusion from the atmosphere is not sufficient (Hlaváč *et al.*, 2014). It is a paradoxical situation when the stage of 'clean water' (water with higher transparency) is risky for fish stock. The most common way to change that is a usage of low concentrations of Soldep (insecticide - organophosphate), which is poisonous to *Daphnia.* It allows the development of phytoplankton and improves oxygen ratios (Janda *et.al.,* 1996).

Due to the ecological situation in ponds the reduction of fish stock is risky because eliminating the fish stock the planktonic communities with a predominance of large *Daphnia* grow and it causes oxygen deficits during the pure water stage. Maintaining intense vegetation coverage is not without risk either (Francová *et al.*, 2019b). They are mainly represented by planktonic cyanobacteria. They are poorly suited as food for zooplankton and they are able to maintain a very high pH level consistently. Many of them produce various types of toxins and, last but not least, their respiratory activity or collapse of the entire population means a risk of oxygen deficits (Janda *et.al.,* 1996).

2.3.2 Changes in zoobenthos groups

In the late 19^{th} century, zoobenthos communities were much richer in species composition than they are in the contemporary ponds (Pechar, 2000). This results from a relatively large list of larvae of caddisflies (Trichoptera), but also larvae of mayflies (Ephemeroptera), stoneflies (Plecoptera) and dragonflies (Odonata) and also bugs (Heteroptera), found in more closely monitored ponds. This fact is evidenced by the regular occurrence of larvae of caddisflies found in the digestive tract of carp (Šusta, 1884). Several species of Gastropoda, Bivalvia and Bryozoa were regularly found. Surprisingly, the occurrence of freshwater mushrooms (Porifera) was observed as well. It is worth mentioning the relatively frequent occurrence of silver spider (*Argyroneta aquatica*), regular ladybug (*Asellus aquaticus*) and occasionally the crayfish (*Astacus astacus*). It can be assumed that the midges (Chironomidae) were also represented by numerous species, but the determination difficulties obscured this fact. (Šusta, 1984).

The increase of carp stock led to the elimination of the regular occurrence of most of these zoobenthic groups and the relatively abundant occurrence of the larvae of Chironomidae and Tubificidae. Those two groups are connected to a relatively high nutrient level. There are both organically richer mud formed by the decomposition of farmyard dung and many times more planktonic organisms falling to the bottom in the pond. The disappearance of many species of zoobenthos from ponds is also contributed by the regular annual drainage of many ponds and their wintering (*Anodonta, Unio*). That leads to loss of shelters and disappearance of macro vegetation. The changes in water chemistry caused by the higher content of suspended organic substances and the regularly unfavorable oxygen conditions at the bottom may also play a role. It is not possible to compare biomass of zoobenthos in the past and present on the basis of literature, because the oldest data are only qualitative. However, it was probably very low, similar to zooplankton biomass (Petrovici *et al.*, 2010).

2.4 Development and formation of macrophyte in ponds

One of the basic characteristics of the water and wetland macro vegetation of natural habitats is the considerable adaptability to water column fluctuations, to the complete exposure of the bottom and also to a wider range of nutrients. It is precisely in these parameters that ponds often undergo frequent and significant changes as a result of economic activity. The intensity of economic interventions, which has changed several times over the centuries of ponds, significantly modifies the natural processes of aquatic vegetation development. These processes are well described from lake littorals (Whigham *et.al*., 1990).

In the ponds with low farming intensity, littoral macro vegetation communities exhibit considerably similar features of natural successive stages of development as in shallow lakes. In the intensively managed ponds, it is possible to document the specific development of certain types of vegetation, decisively formed by economic interventions (vegetation of muddy bottoms during summer drainage, changes in the composition of submerged vegetation depending on the size of fish stock, etc.). Overall, the pond littoral vegetation shows a higher degree of instability than the lake littoral ecosystems (Eiseltova, 2010).

Intensification of pond management has led to changes in the periodicity and dynamics of the water regime, an overall wider range of changes in the height of the water column and changes in the frequency of drying. The nutrient supply burden of the entire pond ecosystem was fundamentally changed. Gradually, the influence of fish stock (higher fish stock directly affects mainly submerged vegetation), as well as a change in the structure of phytoplankton, which is manifested by a decrease in transparency, became essential. Finally, herbivorous fish were introduced (Šusta, 1898).

2.4.1 Submerged macrophytes and fish production

Submerged littoral vegetation is an important habitat for fish. It provides them a shelter for reproduction and protects them from predator attacks. As many species of aquatic invertebrates are hidden in vegetation, it is also an important food supply. The relationship between macrophytes and fish depends on the composition of each species of fish, plants, water body type and geographical area (Francová *et al.*, 2019b).

Macrophytes generally increase the productivity of the aquatic surroundings, however, the amount of vegetation and the amount of fish correlate negatively. R. Randall et al. (1996) deals with the relationships between submerged vegetation and the abundance and diversity of fish found in the Great Lakes in North America. They found that species diversity and density of aquatic invertebrates are high close to aquatic macrophytes, which attracts many fish. Macrophytes diversify the aquatic environment and thus increase the heterogeneity of habitats. This diversification increases fish species diversity and affects the predator – prey interactions. The species richness of fish was demonstrably smaller in lakes without vegetation. It was also found that the larger density of water macrophytes, the smaller fish were in the lake (Randall *et al.*, 1996).

2.4.2 Influence of macrophytes on aquatic invertebrate populations

Aquatic bodies with dense stands of aquatic macrophytes generally contain greater densities of aquatic invertebrates. The more structured the habitat is, the higher species richness it represents. Diversification of habitat could be due to the architectural complexity of aquatic macrophytes (Francová *et al.*, 2019b). This relationship has not been explored in detail, yet. Habitat architecture has been shown to play an essential role in colonization, species richness and overall abundance of aquatic invertebrates. The results of study of Hinojosa-Garro et al. (2010) shows that the structural complexity of plants is an important factor affecting the abundance of individuals of invertebrates (Hinojosa-Garro *et al.*, 2010).

Dense vegetation decreases fish predation on notonectids and other invertebrates. Invertebrates that inhabit aquatic vegetation reduce the risk of fish predation, but at the same time increase the risk of predation by other invertebrates (odonates, nepids etc.) who live in vegetation. It means that a strong association with vegetation can be beneficial in fish ponds or harmful in ponds without fish but with a high population density of predatory invertebrates living in vegetation (Bennett and Streams, 1986).

Vegetation dependence has been shown to be essential for the occurrence of two species of water bugs: *Notonecta lunata* and *Notonecta undulata*. *N. undulata* is the most common backswimmer of medium and small ponds in North America. Where there were no fish present in the pond, *N. undulata* was the most numerous and dominant. However, *N. undulata* shows low density in ponds with fish. At the same time, *N. lunata* is dominant in fish ponds. The basic difference in the behavior of these two bugs is that *N. lunata* is strongly connected to vegetation throughout its development. *N. undulata* lives in vegetation only initially, later leaves the vegetation and lives in open water and thus becomes an easy prey for fish. *N. lunata* can coexist with fish because it lives hidden in vegetation, it is also smaller, and lighter, and it makes it less visible to fish. This study shows that differences between species in life strategies and behavior can explain the characteristics of local distribution and helps to explain the differences in species composition of bugs in different habitats. Thus, bugs exposed to predation can be eliminated in fish ponds. However, predation is only one of the factors influencing the structure of aquatic invertebrate groups (Bennett and Streams, 1986).

2.4.3 Macrophyte in fishponds under different fish farming management

Macrophyte assemblages are widely diversified in different types of ponds, but environmental factors forming their composition and abundance *in situ* are not well understood. Fish farm management in ponds is influenced by many factors - fish size, fish age, type of feed, summer or winter drainage. Fish stock seems to inhibit macrophyte growth - water turbidity, plucking, feeding. Nowadays, most ponds in Central Europe are eutrophic to hypertrophic, which can also negatively affect the growth of macrophytes (Francová *et al.*, 2019a).

A large diversity of aquatic macrophytes was found in ponds of Central Europe. The study of Francová *et al*., 2019b compares the differences in macrophyte composition in nursery and main ponds. In nursery ponds there is a lower fish population and higher water transparency, especially in the first year in spring. During the season, the growing influence of carp on pond development was recorded. There was an extreme increase in chlorophyll *a* and at the same time the transparency of water decreased rapidly. During its development, carp changed its food source from plankton to zoobenthos, and that caused increased turbulence and damage to plant communities. Nursery ponds contained a large number of aquatic macrophytes, as the water was transparent for a long time (Francová *et al.*, 2019b).

The situation in the main ponds was different. Due to the extreme conditions caused by the high fish stock, the total number of macrophyte species and their abundance was lower. Main ponds were not suitable habitats even for free floating macrophytes, due to water movements caused by carp. Macrophytes may have been eaten by fish or birds. Thus, carp was not the only reason of macrophytes loss. In spite of unsuitable conditions, dynamics in the development of aquatic macrophytes were recorded, especially in shallow waters where fish do not float. However, as soon as the water level increased, the water macrophytes disappeared again quickly (Francová *et al.*, 2019b).

2.5 Effect of fish predation on aquatic invertebrates

Fish predation pressure is one of the main influences on the structure, abundance and species composition of aquatic invertebrates in natural freshwater habitats. Many behavioral and morphological adaptations of invertebrates are a consequence of fish predation pressure. The presence or absence of fish can have a greater impact on invertebrate communities than environmental factors themselves. In the present the intensive fish pressure is part of most ponds, but the relationships between organisms are not well understood (Schilling *et al.*, 2009).

There are two types of searching for food – 'active' model and 'sit and wait' model. Optimum prey search time varies depending on prey density, prey movement and predation risk. Since it is common for prey to be close to the predator when foraging, and since prey movements can attract the predator, trade - off between feeding and predation risk should exist. Animals either choose less effective mechanisms of foraging which is safer from predators, or vice versa. The suppression of prey activity reduces the reproductive success and successful foraging, which can affect population densities (Johansson, 1993).

2.5.1 Predator-prey interactions

To be able to live and reproduce in the presence of fish, the prey must develop mechanisms to prevent predation. It can be either to avoid fish in time and space, or to develop tools to reduce vulnerability to predation (Williamson C. E. *et al.*, 1989).

Prey reactions to predators vary widely and may affect the population and species composition of the entire pond. Some studies show that the decline in total prey biomass is usually accompanied by a decrease in the average prey size and thus often an increase in the number of prey. Predators influence the prey's behavior and morphology, thus creating competitive interactions between different kinds of prey. The prey compete for hiding from a predator. Mittelbach (1988) shows that with the increase in fish numbers, the average size of the invertebrate body decreases significantly, which is associated with a decrease in the number of large invertebrates. It also shows that the rate of growth of fish population correlates positively with the availability of prey (Mittelbach, 1988).

2.5.2 Defenses of aquatic invertebrates

Adult and larval stages of aquatic insects are targets for many predators. For this reason, the aquatic invertebrates have developed many strategies to defend themselves against the predators. These are often very energy efficient mechanisms that make the aquatic invertebrates more likely to survive the predator attack (Corbet, 1999).

There are various defense mechanisms against predators known concerning Odonates. They have no types of chemical defense in larvae or adults (Corbet, 1999). The risk of fish predation is reduced by the various behavioral and morphological features of the odonate larvae. As demonstrated by Wohlfahrt et al. (2006) these features occur in the presence and absence of predator (Wohlfahrt *et al.*, 2006). Larvae of some species reduce their activity in the presence of fish, while others escape from predators by swimming (Johnson, 1991). Many Odonata larvae remain hidden in dense vegetation or are buried in bottom substrates (Thompson, 1987). Some Anisoptera larvae have extremely flexible abdomen with lateral and dorsal spines on it, which intimidate many fish predators (Cordoba-Aguilar, 2008).

Gerridae move quickly across the water membrane and can even jump. They often have countershading to reduce their shadows. To record surrounding movement they use their extremely long midlegs (Scrimshaw and Kerfoot, 1987). Corixidae are able to jump out of the water and fly away, they have defensive glands all over their bodies (Kovac and Maschwitz, 1991). Notonectidae swim on their backs, and often have reverse countershading. If the fish try to catch them, they can inject painful poisonous saliva (Schmidt, 1982). Nepidae are well camouflaged due to their brown color. They are dorsoventrally flattened and thus resemble a dead leaf. They use poisonous secretions as active defense (Del-Claro and Guillermo, 2019).

Many predators (vertebrates, fish, amphibians, Odonata, Hemiptera) target Dytiscidae. They also have various defense mechanisms. Usage of them usually depends on the size and type of habitat where they occur. Many Dytiscidae are able to swim or hide quickly. In our ponds Dytiscidae tend to be cryptically colored and they also use an extended chemical defense. Noteridae are mainly found in muddy bottom substrates and their glandular secretion is used to defend (Dettner, 2014).

The vast majority of aquatic invertebrates have a cryptic coloration. Some proficient swimmers escape the predators actively by swimming, or they can jump out of the water and fly away, others hide in the vegetation or are buried in the bottom. Some groups have glands with repulsive to toxic secretions. A single group (Anisoptera) has spines on the body that intimidate or repel fish predators. Types of antipredatory defense of individual groups usually depend on the body size, properties and habitat types of individual groups of aquatic invertebrates (Del-Claro and Guillermo, 2019).

2.5.3 Low fish stock ponds

A few studies that focused on ponds with no fish have shown that the introduced fish can have a great impact on the size structure of benthic invertebrate communities. Hall et al. (1970) in his study showed the effect of bluegill prediction on invertebrates. As a result, there was a significant negative correlation between populations of all benthic invertebrate taxa that weighed >0.01 mg dry weight and those of the introduced bluegill fish. On the other hand, there was no influence recorded of fish on the total biomass of benthic invertebrates. The conclusion is that the strongest effect of fish was to reduce the number of big invertebrates and that body size was more determining than tax liability in relation to the invertebrate response (Hall *et al.*, 1970).

Crowder and Cooper (1982) had similar findings. When bluegills were added to the pond with no fish, the average size of invertebrates decreased compared to fish-free control ponds. This reduction in the mean prey size is connected with reduction in the number of big invertebrates (for ex. Hyalella, Zygoptera) in the presence of fish. The total biomass of invertebrates decreased by the presence of fish, despite the increase in the number of small invertebrates in the presence of bluegill fish (Crowder and Cooper, 1982).

Study of Post and Cucin (1984) explored the littoral areas of Little Minnow Lake in Ontario and its invertebrates. It was investigated before and after the accidental introduction of *Perca flavescens*. They studied nine invertebrate taxa, five significantly decreased in total biomass and eight significantly decreased in average weight, but none significantly reduced density (Post and Cucin, 1984).

The study of Schilling (2009) found several differences between invertebrate composition in fish-free lakes and in fish-introduced lakes. Numerous differences were found in the structure of invertebrate communities, abundance and species richness. The presence or absence of fish was a key determinant of community structures rather than different environmental characteristics. The taxa associated with the absence of fish were: Hemiptera - Notonectidae and Corixidae, thriving in fish-free environments. It also applies to *Buenoa spp*., *Notonecta spp*., *Hesperocorixa spp*., *Callicorixa spp*. and *Sigara*. Although habitat characteristics affect the distribution of notonectid and corixid species, the presence or absence of fish is the primary factor explaining the observed distribution of species between lakes. The abundance of Notonectidae and Corixidae species decreased considerably following the introduction of fish, due to their susceptibility to predation. These are relatively large taxa, they must float regularly on the water surface to be able to breathe, that makes them visible and often exposed to predation. The largest notonectids (*N. insulata*) and corixids (*Hesperocorixa spp*.) have been identified as bioindicators of fish-free lakes (Schilling *et al.*, 2009).

The abundance and distribution of Dytiscidae was more influenced by the presence of fish than by environmental variables. The abundance of dragonflies and the distribution of all species strongly correlated with the presence or absence of fish. Species associated with fish-free habitats are often large, visually active predators. In contrast, in fish ponds, dragonflies are smaller, slow-moving and cryptic. In fish-free lakes, dragonflies of the genus Aeshnidae are more abundant. It is a group that includes the largest dragonflies in North America. In this study, the Gerridae group was the only taxa common in fish lakes and, on the contrary, did not occur in fish-free lakes (Schilling *et al.*, 2009).

2.5.4 High fish stock ponds

The studies mentioned above were carried out on ponds without fish or on ponds with few fish. It can be argued that the impact of introduced fish was particularly dramatic. However, studies conducted on fish ponds showed the same general effect of fish predation on invertebrate populations. For example, the occurrence of fish had a significant negative effect on the abundance of large invertebrates (often decreasing average invertebrate size) but appeared to have no effect on the total number of invertebrates.

Morin (1984) found a significant effect of fish predation on the size composition of dragonfly larvae in a pond in North Carolina. The exclusion of fish resulted changes in their communities. Originally dominant small communities were replaced by medium size communities (Morin, 1984).

Gilinsky (1984) also investigated the invertebrate community in ponds using experimental cages with and without fish. This study showed that fish significantly influenced the occurrence of some benthic invertebrates. However, the results were rather difficult to interpret because the abundance of some invertebrate species increased and others decreased and the effect of fish predation varied according to the season. Gilinsky examined the densities of species, but did not monitor the distribution of different body sizes (Gilinsky, 1984).

Hanson and Leggett (1986) also examined only the total number of invertebrates and their biomass. However, the invertebrates were not affected by density of fish (Hanson and Leggett, 1986).

The effect of fish on the density of large invertebrates is masked if we focus only on the total number of invertebrates. It includes classes of small body size, which are generally much more abundant in the community (Mittelbach, 1988). Mittelbach's study showed that fish had a very negative effect on densities of large invertebrates but no effect on densities of small invertebrates. The lack of influence on the abundance of small invertebrates contradicts the results of Crowder and Cooper (1982) who found that the abundance of small invertebrates increased in the presence of fish (Crowder and Cooper, 1982).

Gilinsky (1984) found that the density of small chironomids increased in the presence of fish. That is probably the response to the reduction of larger types of chironomids (Gilinsky, 1984).

The study of Mittelbach (1988) experimentally investigated the effect of fish on benthic groups of invertebrates in lakes. Based on the findings in this study, fish in lentic systems have: a strong influence on the size structure of benthic invertebrates, little or no effect on species diversity of benthic invertebrates, and variable effects on overall invertebrate density (Mittelbach, 1988).

The importance of predation that regulates the distribution and abundance of Chironomidae larvae is controversial as there is insufficient information. Many authors have speculated or tried to prove the importance of vertebrate predators in the regulation of larvae of midges. A study by Gurzeda (1960) shows that carp changed the size distribution of midges larvae by feeding most on small larvae. According to study of Thorp and Bergey (1981) the mean total number of midges was not significantly affected by the exclusion of vertebrate predators (Thorp and Bergey, 1981).

2.5.5 The effect of fish predation on *Notonecta sp.*

Habitat structure is crucial for prey to survive. Invertebrates are very sensitive to predation when they have no possibility of shelter (Walls *et al.*, 1990).

Based on the study of Cook and Streams (1984) the presence of vegetation as a potential shelter reduces the risk of predation of six *Notonecta* species. The smallest species usually try not to move in the presence of fish. Some species are able to climb out of the water. Their foraging behavior, the relatively large body size, and frequent trips to the water surface for air make *Notonecta* potentially exposed to a high risk of fish predation. The smallest fish (11cm) apart from larger fish has more difficulty handling *Notonecta*. When fish is looking for food, it tends to detect large prey species (*N.irrorata* and *N. insulata*) significantly earlier than small species (*N. lunata* and *N. petrunkevitchi*). During the field research in tanks without vegetation, the search time for large individuals was less than 5 seconds, while for small species the time was more than one minute. Many notonects survived the initial attack of the fish and saved themselves from their clutches. In several cases, the prey was able to escape the fish by climbing out of the water. The ability to avoid an initial attack does not seem to be related to the size of *Notonecta*. The study was based on the Ware prey risk model (1973). The main factors included in his model are: the width of the predator search range, the proportion of prey in that range, the prey that is recognized, the probability that the prey detected can be caught, the time consuming prey handling, the predator search speed and the prey density. Presence of vegetation increased predator search times, probably by reducing prey exposure. The presence of vegetation also enabled *Notonecta* avoid frequent attack by fish (Cook and Streams, 1984).

N. lunata and *N. petrunkevitchi* have the greatest chance of avoiding predator and surviving in a fish pond. They are linked by the following: a narrower search range for fish due to the small size of the prey, they are light colored and thus have less contrast in open water, a tendency to remain motionless in the presence of fish. It gives them a great ability to prevent catches when the fish attacks. The interaction of fish with *Notonecta* adults can be a major factor in determining the habitats of these species (Cook and Streams, 1984).

2.5.6 The effect of fish predation on Odonata

Studies on the dragonfly larvae of the Coenagrionidae family show that their foraging behavior varies in the presence or absence of a predator. However, there are no studies focused on how the behavior of prey varies depending on the characteristics of prey and its density in the presence or absence of a predator. Study of Johansson (1993) shows that the *C. hastulatum* larva become motionless in the presence of a predator. Conversely, higher activity without predator was reported. The author suggests that if the larvae of *C. hastulatum* starve for a long time, the activity will probably increase even in the presence of a predator. That will increase the risk of predation (Johansson, 1993).

Study of Morin, 1984 dealing with dragonfly larvae and their predators shows that the experimental exclusion of fish dramatically alters the abundance and dominance of species in dragonfly larvae (Anisoptera). A hypothesis was tested, based on the following observation: a small species of dragonfly (*Perithemis tenera*) dominated the group of dragonflies. However, this species has competitive disadvantage compared to larger species (*Tramea lacerata*) inhabiting the same area (South-East Europe). The author assumed that dense fish stocks that are selective for the prey size can cause the growth of small species. The fish stock suppress the abundance of larger dragonfly species that could otherwise compete with or catch smaller dragonfly species. This short-term experiment confirms the effect of vertebrate predator on the structure and composition of invertebrates. This study shows that predation may be an important regulatory mechanism during the larval phase of the dragonfly life cycle. The presence of fish greatly reduces the number of larvae, allowing a successful transition from a water larvae to a terrestrial adult. These interactions underline the potential of competitive interactions between fish and terrestrial species sharing the aquatic environment. It encourages for further study and are partly supported by further observations. A negative relationship between the reproductive success of some water birds and the presence of fish was observed. In the presence of fish, there were fewer invertebrates in the pond, leaving little food for birds (Morin, 1984).

The linkage between the abundance of adults and dragonfly larvae may be variable. The composition of the larval population is influenced by predation (especially species susceptible to predation). Larval communities are also affected by predatory pressure among dragonflies, competition and possible physiological constraints. The composition of adult dragonfly populations is affected by the composition of pond larvae, adult migration, competitive and predatory interactions among adults. To address the importance of these potentially complex interactions in determining the distribution and abundance of species with complex life cycles, integrated studies of the water and adult phases of dragonfly life need to be initiated (Morin, 1984).

2.6 Fish pond as a monoculture

Freshwater habitats support some of the most biologically rich and diverse habitats, but at the same time they are the most endangered ecosystems worldwide (Gioria *et al.*, 2010). There are many activities that endanger these ecosystems - overexploitation of resources (water, fish), eutrophication, pollution, hydrological regime adjustments, deterioration of habitats of ponds and their surroundings and colonization by non-native species. Some studies have shown that in anthropogenically modified landscapes and intensive agricultural areas, ponds represent hot spots of biodiversity and work as habitat islands in otherwise ecologically poor habitats (Céréghino *et al.*, 2008).

Although the pond ecosystems are used intensively and they are so important for society and biological communities, they are not given enough attention (Williams *et al.*, 2004). Most ponds provide poor habitats and poor water quality due to nutrient enrichment (chemical and organic), as well as induction of non-native species (Biggs *et al.*, 2007).

While special protection areas are important for the conservation of species and habitats, the conservation of biodiversity beyond these protected areas should be considered (Le Viol *et al.*, 2009).

The structure of pond communities is changing rapidly, mainly due to the intensification of pond management. There is a high predation pressure in fish ponds because of the excessive number of fish stock. Furthermore, littoral vegetation is gradually reduced. Not only the combination of these two factors makes aquatic invertebrates difficult to survive (Baudron and Giller, 2014). There is a total loss of invertebrate species and individuals. However, interspecific and intraspecific interactions structuring the ponds communities are not well understood (Walls *et al.*, 1990). The pond should provide a heterogeneous environment for the occurrence of many plant species, invertebrates and vertebrates. In our intense landscape, however, it becomes a monoculture and it allows only selected, resistant species to survive (Hill *et al.*, 2016).

The study of Buczynska (2007) examines a pond complex in Poland. There are some ponds with intensive farming and some ponds where farming is prohibited. This relatively small area contains running water (stream), canals and several types of stagnant water. It creates ideal conditions for the occurrence of species with very different ecological preferences. The forest is also important, because it serves as a buffer zone and thus ensures the isolation of habitat from adverse environmental influences. Buczynska (2007) found that the fauna of ponds and adjacent water bodies is even richer than the fauna of ponds in the protected reserves. That is why more emphasis should be paid on activities in unprotected areas in the nature conservation. A Pond is a special type of habitat that cannot exist without human intervention, and a specific management should be selected with regard to its structure (Buczynska *et al.*, 2007).

3 Field research

3.1 Brief characterization of the groups of the interest

3.1.1 Order Odonata

Dragonflies belong to the superorder Odonatoptera, it is one of the oldest insect radiations. Their existence dates back at least to early carbon. This radiation includes the largest insect that has ever lived. The unique properties of dragonflies are the strongly modified larval labium and the mechanism of indirect sperm transfer in adults (Thorp and Rogers, 2015).

Habitat

Odonata inhabit almost all freshwater habitats. Many species are associated to specific habitats such as forest streams or acid waters. One of the most important factors influencing the occurrence of dragonflies is the presence or absence of predators, especially fish. While a sedentary larva is usually limited by a specific aquatic habitat, a moving adult is active and fly to places very distant from the water (forests, mountain peaks). Their oviposition is associated to vegetation, that is why most dragonflies need it to be present in their habitat. Dragonfly larvae inhabit different types of microhabitats depending on the species. For example, most Gomphidae larvae are burrowers, their occurrence varies based on different types of substrate (sand, mud). Others hide among submerged vegetation or tree roots (Thorp and Rogers, 2015).

Dispersal

Since dragonflies are excellent flyers, they are expected to disperse well. However, it is difficult to track them over longer distances. There are not many records of dragonfly movements over longer distances. (Corbet, 1999). Migration of dragonflies is defined as a spatial displacement, in which part or all of the population is usually involved. The place where the dragonflies develop is usually not the same as the one where they reproduce (Corbet, 1999). Recently, the impact of climate change on dragonfly populations has been shown. In Europe, there has been an increase in temperature, and it has caused the expansion of ubiquitous stagnant water species from south to north (Ott, 2010).

Foraging behavior

Dragonfly larvae are exclusively carnivorous predators. Larvae feeds on different kinds of organisms, usually it depends on what prey they can handle and what habitat the larva inhabits. Many Zygoptera and Libellulidae larvae feed on zooplankton, Aeshnidae feed on larger prey, including other odonates, toads and small fish. Burrowing species, such as Gomphidae, mainly feed on other bottom-living species (Chironomidae and Oligochaeta larvae). There are different ways to search for food: species can be active, or sedentary, visual or tactile. These different strategies can be combined in different ways (Corbet, 1999). The active larva seeks prey by crawling or even swimming. The sedentary larva remains stationary in 'sit and wait' mode until suitable prey is within range. The visual larva is dependent on the eye signal to capture the prey. The tactile larva identifies the presence of prey based on a stimulus from the mechanoreceptors on the legs, antennas and mouth. Again, these modes can be combined. Factors that affect feeding rate are prey size, prey abundance, presence of other predators, prey handling time and temperature (Gresens *et al.*, 1982).

Adult dragonflies are visual predators, using eyesight to catch the prey. However, they have several different strategies. While hunting, Zygoptera prefer to catch sitting prey, most of Anisoptera catch only flying prey. Dragonflies are probably highly selective for prey size. They prefer prey in the species-specific size range (Olberg *et al*., 2005).

Biotic interactions

Odonate larvae occupy medium to highest positions in the food net of freshwater habitats. It mainly depends on the presence or absence of fish as predator. In lakes and rivers where fish are common, dragonfly larvae occupy a central position in the food net, but in fish-free habitats there may be the top predators. The main predators of dragonfly larvae are, among other dragonflies, probably fish. It influences the structure of dragonfly communities (Knight *et al.*, 2005).

Dragonfly larvae have different strategies to prevent a predator attack. These are behavioral and morphological adaptations. Behavioral adaptation is an overall decreased activity in the presence of a predator or an increased use of shelters in vegetation (Edmunds, 1974). Morphological adaptations are, for example, crypsis or abdominal spines. The length of spines varies considerably between species. These spines provide defenses against fish predators by prolonging the prey handling time (Johansson and Samuelsson, 1994). Several studies have shown that the composition of the dragonfly community is strongly influenced by intraguild predation, especially in habitats where larger predators are missing (Johansson, 1993).

3.1.2 Order Coleoptera

Aquatic beetles can be defined as those that are connected to freshwater or saltwater habitats. At the same time they have at least one phase of life which completely depends on the aquatic environment. The order Coleoptera contains more than 12 000 described aquatic species so it is considered to be one of the biggest groups of aquatic insects (Jäch and Balke, 2008).

Habitat

Aquatic beetles inhabit almost all types of aquatic habitats - large freshwater lakes, but also small periodic pools. It appears that more species of aquatic beetles are connected to stagnant waters than to running waters, unlike other groups of aquatic invertebrates (Ephemeroptera, Diptera). Most aquatic beetles are found in freely accessible water bodies (lakes, ponds, streams), but a few species live on subterranean habitats (Dytiscidae) (Thorp and Rogers, 2015).

Aquatic beetles also use different spatial habitats of individual water body. Many beetle species are related to the bottom surface (detritus, rocks, mud), and others are primarily pelagic (adults Dytiscidae and Hydrofilidae) (Hilsenhoff, 1987).

Abiotic factors affecting the occurrence of aquatic beetles are: habitat structure (cover, habitat stability), hydrological parameters (water flow, substrate type including macrophytes), water chemistry (salinity, nutrients, acidity, conductivity), and others (pH, temperature, dissolved oxygen). Most aquatic beetles have strict microhabitat requirements and often respond to small scale changes (Thorp and Rogers, 2015).

Aquatic beetles often occur in large numbers in newly formed habitats. It allows adults and larvae to escape fish or dragonfly predation or restricts competition with other groups. As a result there are large differences in the composition of groups of aquatic beetles in habitats of different ages. Fish are a strong biotic factor affecting the presence or absence of aquatic beetles (Fairchild *et al.*, 2000).

In addition to predation, another key factor affecting aquatic beetle communities is a competition. However, the evidence that the competition is really a strong factor varies especially concerning some groups of beetles (Dytiscidae). Species with similar morphology often coexist (Larson, 1985). It indicates the presence of coexistence mechanisms. The lack of interspecific competition among beetles is probably due to other factors - tolerance to environmental chemistry, interaction at higher trophic levels. Those factors apparently weakens the effect of competition (Vamosi and Vamosi, 2007).

Dispersal

Flight dispersal is the most important type of movement for many adult aquatic beetles. It may seem that some beetles, despite their size and weight, never fly (Hydroscaphidae, Lepiceridae). But the truth is that the vast majority of them are capable of multiple short flights. Some beetles can even fly kilometers away. Species that are unable to fly are spreading between habitats walking on the ground (Leiodidae). Beetles of smaller sizes are easier to spread further or more often as they have lower energy requirements (Verberk and Esselink, 2005).

Foraging behavior

A wide range of morphological features and habitats, among species of aquatic beetles, are associated with different ways of foraging. Aquatic beetles can be predators, herbivores or detritors. Predation is a common among aquatic beetles. A large number of them exclusively consume other animals. Predatory aquatic beetles catch and consume prey in different ways. Dytiscidae larvae have large mandibules that they use to puncture their prey and inject digestive enzymes. Other beetles absorb their prey or chew with mandibles. The types of prey vary, but usually it is other aquatic insects (Diptera larvae) or crustaceans. Water beetles can also catch small vertebrates. For example, the larvae of Dytiscidae easily attack tadpoles or small fish. Herbivorous beetles also differ in the type and method of foraging. They usually scrape algae and cyanobacteria off surfaces (Thorp and Rogers, 2015).

Biotic interaction

In addition to being predatory, water beetles are also predated by other invertebrates (odonate larvae) or vertebrates (fish) (Thorp and Rogers, 2015). The intensity of predation in aquatic ecosystems is great. It is not so surprising that water beetles have developed various defense mechanisms - chemicals, mechanical defense, crypsis. For many species of beetles and their larvae, the crypsis is a common defense. They can either hide in the gaps between the rocks, stay buried in the substrate, camouflage or they combine these strategies (White, 1989).

3.1.3 Order Hemiptera

The order Hemiptera was originally terrestrial and then secondarily associated to water habitats. Respiration, mating and general physiology are secondarily adapted to life in the water (Thorp and Rogers, 2015).

Habitat

Water Hemiptera are composed of semi-aquatic bugs (Gerromorpha), living primarily on the surface of water, and water bugs (Nepomorpha), which live submerged under water. Almost all aquatic Hemiptera species are dependent on atmospheric air for breathing and most of them can walk or fly. Water Hemiptera inhabit many types of water habitats (Thorp and Rogers, 2015).

Dispersal

Legs of water Gerromorpha are well adapted to skating on the water surface. The legs of Nepomorpha are adapted to swim and grab a prey. The hairs on the legs and body surface allow the gerromorphs to move without affecting the surface tension of the water.

Aquatic Hemiptera have different swimming skills and strategies. Naucoridae*,* Corixidae and Notonectidae are excellent swimmers, able to move fast. On the other hand, Gerriidae and Veliidae inhabit the water surface. Nepidae move slowly and are adapted to adhere to vegetation.

Most water Hemipter usually flies only when they want to expand or migrate from one habitat to another. Some Hemiptera fly only once or twice in their lifetime to migrate and during the rest of the life cycle the flight is not required (Thorp and Rogers, 2015).

Foraging behavior

Most aquatic Hemiptera are predatory and use rostrum to hunt. They feed on other invertebrates, but they can also catch small fish or frogs (Thorp and Rogers, 2015).

The great Belostomatidae are equipped with great predatory abilities. They can even feed on fish, frogs, snakes and birds. Aquatic Hemiptera puncture their prey with rostrum. The size of the predator's body is thus not limiting in relation to the size of the prey's body (Menke, 2011).

Gerridae and Veliidae are sensitive to water vibrations. And the fact allows them to find the prey that stucks on the water surface. Nepidae and Belostomatidae are often on the vegetation or in the rocky edges of the pond. Their legs are stretched out waiting for the prey. Notonectidae often float freely in pelagic water and wait for small prey, such as *Daphnia* and *Copepoda*. Although aquatic Hemiptera are usually predators and often are the top predators in their habitat. But sometimes they become a prey as well (Boersma *et al.*, 2014).

Biotic interactions

According to the Macan study (1966), fish predation can have a major impact on the use of habitats by Hemiptera (Macan, 1966). Cook and Streams (1984) found that if *Notonecta* coexist in the pond with fish, it will reflect its morphology and behavior. They try to reduce the risk of predation. Another important biotic interaction that affects their communities is cannibalism (Cook and Streams, 1984). Fish predation significantly affects the distribution and abundance of two large *Notonecta* species (via 2.5.4 The effect of fish predation on *Notonecta* spp.) (Bennett and Streams, 1986).

4 Methods

The research was focused on the ponds with intensive farming. The premise was that a high fish stock and a small cover of littoral vegetation had a negative impact on aquatic invertebrate populations. Twelve ponds with similar parameters were selected and experimentally tested to the occurrence of aquatic invertebrates depending on the presence or absence of fish in the pond.

4.1 Field experiment

In each pond we built two submerged fences (3x10m) to achieve fish exclusion inside of them. There was planted an artificial vegetation in the form of a floating island in half of them. The fences were always in close relation to the littoral of the pond. In addition, live catch traps were installed. Always one inside the fence and the other outside. Furthermore, we were catching a water invertebrate with a sweeping net again inside and outside the fence. The aim was to determine whether the density and biodiversity of water invertebrates will be higher inside the fence and lower outside. The question was whether the absence of fish and the presence of vegetation even in such a small scale has a positive effect on the occurrence of aquatic invertebrates (differences in numbers of captured invertebrates outside and inside the fence with vegetation). And how the vegetation influences the occurrence of aquatic invertebrates (fence with and without vegetation). Samples were further determined in the laboratory.

4.1.1 Locations

We chose twelve ponds with similar parameters to make the conditions for aquatic invertebrates everywhere similar: intensive farming, adult carp, part of the pond system, at least one extensive pond nearby, similar littoral coverage (usually minimal).

Švábský pond

Švábský pond is a water area within an area of 15,5 ha. It is located near České Heřmanice and falls into its cadastral territory (ČÚZK). The altitude is 309 metres above sea level. The pond is separated from the surrounding fields by sparse vegetation of deciduous trees (Mapy.cz).

Poplužský pond

Poplužský pond is a water area with an area of 2.08 hectares. It is located near the village Žumberk and falls into its cadastral territory (ČÚZK). The altitude is 306 meters above sea level. The pond is mostly surrounded by forest and also by fields (Mapy.cz).

Mušlovský pond

Mušlovský pond is a water area with an area of 10,7 ha. It lies near the village Opatov, cadastral area of Opatov v Čechách (ČÚZK). The altitude is 418 meters above sea level. It is located in the middle of the forest. In the wider surroundings from the right, there is a large area of fields (Mapy.cz).

Pond Lačnov

Pond Lačnov is a water area with an area of 13 hectares. It is located in the cadastral area of the village Zálší in the district of Ústí nad Orlicí (ČÚZK). The altitude is 286 meters above sea level. The pond is surrounded by open meadows and only in the wider area is a forest and it is followed by fields (Mapy.cz).

	April	**May**	**June**	**July**	**August**	**September**	**October**
Temperature (°C)	9,5	10,9	20,7	18,6	19,1	13,4	9,6
Precipitation (mm)	23	109	51	44	81	63	46

tab. 1: information for the Pardubice region in 2019, ČHMÚ

Šejb pond

Šejb Pond is a water area with an area of 18.7 ha. It lies near the village Strmilov, cadastral area of Česká Olešná (ČÚZK). The altitude is 543 meters above sea level. The pond is surrounded in part by not very large forest and in most parts by fields (Mapy.cz).

Velký Obecní pond

Velký Obecní pond is a water area with an area of 3.1 ha and is located near the village of Nová Olešná and falls into its cadastral territory (ČÚZK). The altitude is 542 meters above sea level. Near the pond is a small forest, but is largely surrounded by fields (Mapy.cz)

Holub pond

Holub Pond is a water area with an area of 24.8 ha. It lies near the village of Nová Olešná and falls into its cadastral territory (ČÚZK). The altitude is 544 meters above sea level. The pond is largely surrounded by forest and to a lesser extent a meadow and a field (Mapy.cz).

Hraniční pond

Hraniční Pond is a water area within an area of 3.6 ha. It lies near the village of Nová Olešná and falls into its cadastral territory (ČÚZK). It is located at altitude 541 meters above sea level. The pond is largely surrounded by fields, partly also through the forest (Mapy.cz).

Svobodka

Svobodka pond is a water area within an area of 0,6 ha. It is located near the village Myštice, cadastral area Kožlí u Myštic (ČÚZK). The altitude is 456 meters above sea level. The pond lies very close to the village, surrounded mainly by fields (Mapy.cz).

Hájky

Hájky pond is a water area within an area of 1 ha. It is located near the village Rakovice and it falls into its cadastral territory (ČÚZK). The altitude is 440 meters above sea level.

Two thirds of the pond are surrounded by a vast forest and one third is followed by a meadow and then a field (Mapy.cz).

	April	May	June	July	August	September	October
Temperature (°C)	8,6	9,9	20,0	18,6	18,3	12,8	8,9
Precipitation (mm)	16	85	69	69	70	50	37

tab. 2: information for the South Bohemian region in 2019, ČHMÚ

Vočert pond

Vočert pond is a water area within an area of 5,2 ha. It is located near the village Hvožďany and it falls into its cadastral territory (ČÚZK). The altitude is 525 meters above sea level. The pond and its surroundings are located in a specially protected area and at the same time it is Special area conservation. The site is protected because of the occurrence of the fire-bellied toad. The aim is to maintain its stable and rich population (Mapy.cz, AOPK).

Nová Pozdyň

Nová pozdyň pond is a water area within an area of 4 ha. It is located near the village Hvožďany, cadastral area Pozdyně (ČÚZK). The altitude is 530 meters above sea level. The pond is partly surrounded by a meadow, but the vast majority of its surroundings are fields (Mapy.cz).

	April	May	June	July	August	September	October
Temperature (°C)	10,0	11,4	21,5	19,8	19,5	14,1	9,8
Precipitation (mm)	25	72	47	52	72	56	36

tab. 3: information for the Central Bohemian region in 2019, ČHMÚ

4.1.2 Method of sampling

We visited each of the sites once a month, from April to October 2019. Every month, the first day in the field we installed live catch traps, using the chicken liver as bait. The next day we took the traps and took samples of trapped aquatic invertebrates. We gained another sample using a sweeping net. We gained eight sample types from each locality: 1. fence with vegetation, live catch trap 2. fence with vegetation, sweeping net 3. fence without vegetation, live catch trap 4. fence without vegetation, sweeping net 5. outside of fence with vegetation, live catch trap 6. outside of fence with vegetation, sweeping net 7. outside of fence without vegetation, live catch trap 8. outside of fence without vegetation, sweeping net. First, we divided the aquatic invertebrates into groups (Odonata, Hemiptera, Coleoptera). Subsequently, we distributed the samples among experts who determined them into species.

5 Results

Generalized mixed (GLMM) models with negative binomial distribution in R package lme4 (Bates, 2017) were used to analyze the effect of fish elimination and supplementation of vegetation (independent variables) on the overall number of individuals of sampled taxa (dependent variable). Locality was always use as random effect. For the verification of cmodel selection standart diagnostic were used. Similarity of species composition on individual sites with / without elimination fish was analysed using Principal Component Analysis in R package Vegan (Oksanen, 2010). All analyses were performed in R 3.6.2 (R Development Core, 2018).

5.1 The effect of the fence on the family of organisms

We first investigated whether the fence had an effect on each of the families of the captured organisms. We processed the data for families where more than 10 individuals were captured. We also investigated the influence of vegetation, but none was found.

The effect of the fence was found in three groups: Gerridae, Notonectidae and Noteridae. There was significantly higher number of individuals than outside fences (*graph no. 1a, 1b, 1c*). On the other hand, the effect of the fence was not demonstrated in the rest of the groups.

We calculated the total number of individuals for each group, the total number of individuals caught and the total number of individuals caught per locality. Furthermore, we also found a difference in the total number of individuals trapped inside and outside the fence. More individuals were caught inside the fence, but the difference is negligible.

family	**ex**
Aeshnidae	24
Coenagrionidae	408
Corixidae	3144
Dytiscidae	96
Gerridae	115
Hydrophilidae	83
Libellulidae	113
Naucoridae	100
Nepidae	74
Noteridae	99
Notonectidae	226
total	4482

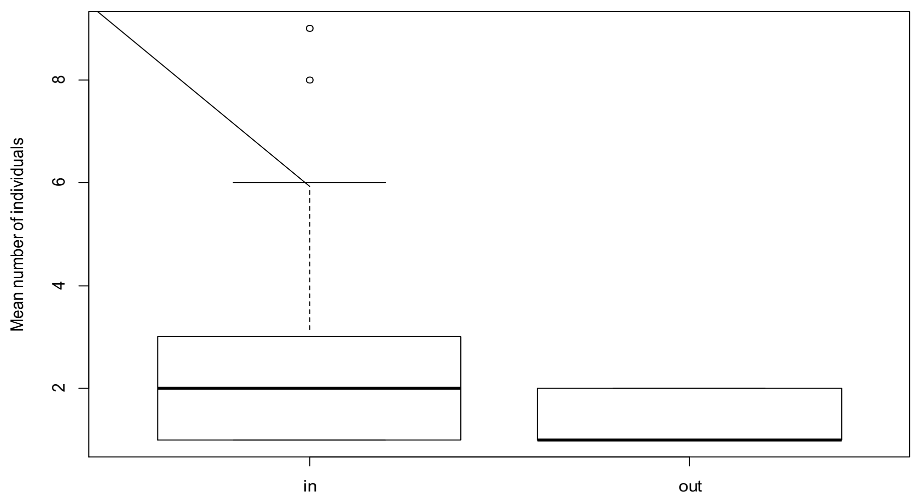
tab. 4: total number of individuals caught from each group

pond	**ex**
Holub	1092
Hraniční	197
Lačnov	389
Mušlový	168
Obecní	153
Pohoří	124
Poplužský	215
Pozdyň	691
Šejb	965
Šváb	380
Svobodka	56
Počert	89

tab. 5: total number of individuals caught on each locality

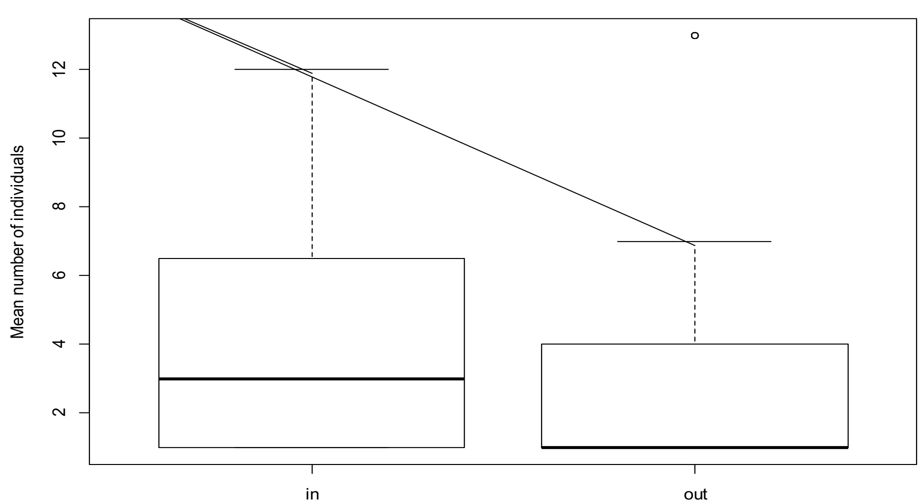
The graphs below show three groups of aquatic invertebrates for which the occurrence of a fence was crucial. Evidently more individuals were concentrated inside the fence, fewer individuals outside the fence. There are two groups of aquatic Hemiptera (Gerridae: *graph no. 1a*, Notonectidae*: graph no. 1b*) and one group of aquatic beetles (Noteridae*: graph no. 1c*).

Gerridae



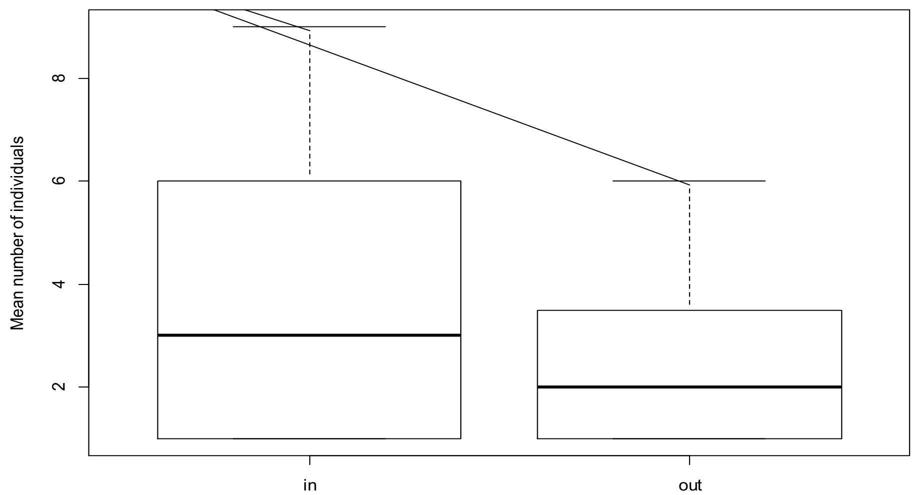
graph no. 1a: the number of individuals in the family Gerridae was higher inside the fence

Notonectidae



graph no.1b: the number of individuals in the family Notonectidae was higher inside the fence

Noteridae



graph no. 1c: the number of individuals in the family Noteridae was higher inside the fence

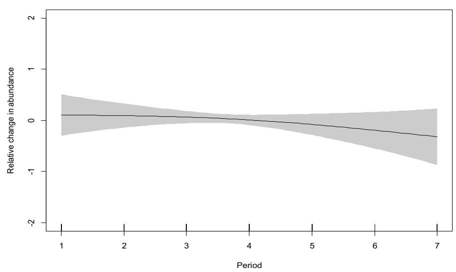
	Gerridae	Notonectidae	Noteridae
Model.0 (AIC)	193.77	323.86	145.47
Model.1 (AIC)	195.77	317.39	147.03
Model.2 (AIC)	189.55	322.45	142.78
P value (M2)	0.01262	0.06476	0.03044

tab. 6: Model.1 indicates whether or not vegetation was favored, Model.2 indicates whether or not a fence was preferred, p value for Model.2

5.2 Influence of seasonality

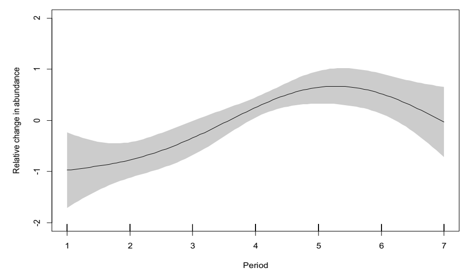
The following graphs show the changes in the relative abundance of individual groups during the season. Data collection began in April (1) and ended in October (7). We selected only the groups that include more than 100 individuals (*graph no. 2a – f*). Graphs for the rest of the groups are attached.

Notonectidae



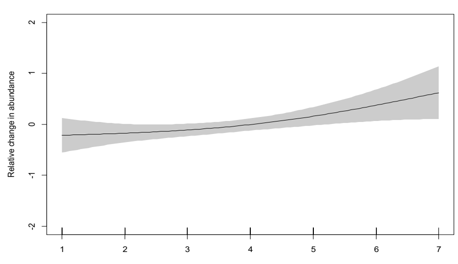
graph no. 2a: the relative abundance of individuals decreases slightly in the season

Coenagrionidae



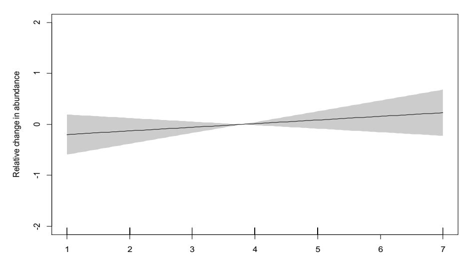
graph no. 2b: the relative abundance of individuals has noticible peak in August, then slowly decreases

Gerridae



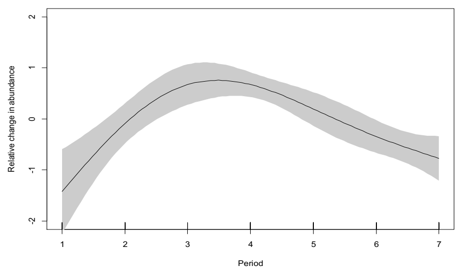
graph no. 2c: the relative abundance of individuals increases during the season

Libellulidae



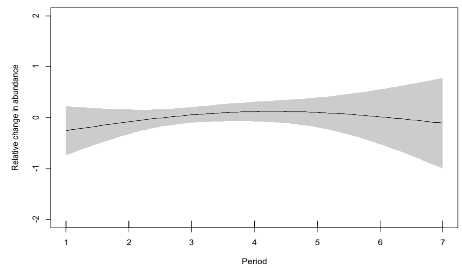
graph no. 2d: the relative abundance of individualsincreases slightly in the season

Naucoridae



graph no. 2e: the relative abundance of individuals has noticible peak in June, after which the abundance of individuals declines rapidly

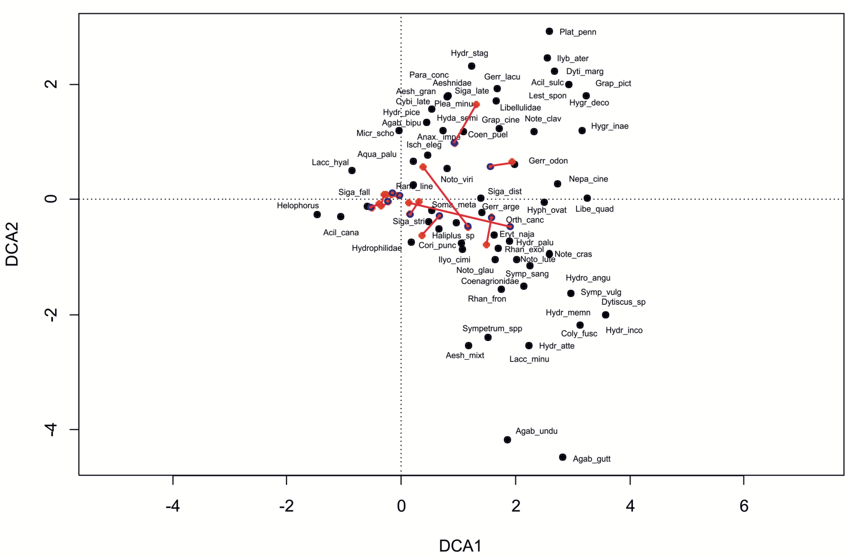
Corixidae



graph no. 2f: the relative abundance of individuals is relatively stable throughout the season

5.3 Number of species in localities

Finally, we investigated whether there was any relationship between the numbers of individual species across the sites. Consideration was also given to whether the species were present inside or outside the fence. The ordination chart shows that there are differences between sites. However, the differences are very small between the species caught inside and outside the fence.



graph no. 3: ordination chart shows differences between localities, but small differences between the species caught inside and outside the fence. Red lines connecting localities inside red dot and outside blue dot.

6 Discussion

6.1 Field experiment

The field research and subsequently processed data have several levels, which allows us to explore the issue from several angles, as it was intended during the construction of the experiment in advance. All groups caught in the field experiment were pioneering species that quickly colonize new environments and they were not very selective in their habitat prefferences. They usually have a short life cycle and occur in large numbers. However, some families of aquatic Hemiptera favored microhabitats with submerged vegetation and therefore they can be considered as habitat selective. Despite a few exceptions, there were mainly small species from different families in the ponds. It can be assumed that they spread easily among different habitats.

6.1.1 Effect of the fence and the vegetation

We first evaluated whether the presence of a fence has an effect on individual families of aquatic invertebrates. We found that the presence or absence of the fence had no effect on the majority groups of aquatic invertebrates. In other words, the presence or absence of fish did not affect the occurrence of these groups. In the contrary, families with an increased presence within the fences were proved. These are the groups Gerridae, Notonectidae and Noteridae.

Gerridae and Notonectidae are aquatic Hemiptera. As mentioned above, these groups have very well-developed defense against the fish predation. According to the Schilling (2009) study, Notonectidae can even be considered as the indicators of fish absence. It is a group that is threatened by fish predation. They breathe air oxygen and need to swim to the surface from time to time. During this journey they become an easy prey for fish. They are also relatively large species that are easily detectable for fish (Schilling *et al.*, 2009). Surprisingly, Notonectidae occurred more frequently in fences with no vegetation artificially added. This may seem strange in particular because the association of Notonectidae to vegetation is widely reported. The presence of vegetation as a potential shelter reduces the risk of predation of *Notonecta* species. The smallest species usually try not to move when fish is present. Some species can even climb out of the water (Cook and

Streams, 1984). Similar preferences apply to the family Gerridae. They are non-specialized predators, as well as Notonectidae, and they more frequently occurred inside the fence. Gerridae, however, again preferred to choose the fence, where vegetation was not present. Aquatic Hemiptera are directly associated to the vegetation. However, they need submerged vegetation to provide a shelter from the predators. The floating vegetated islands in our experiment were apparently poorly designed, because they did not contain any submerged vegetation. Gerridae are very sensitive to water membrane vibrations. Thanks to their sensitivity to the vibrations, they look for a prey, which can be stuck on the water surface. Further, they are able to detect the predators thanks to the vibrations as well (Boersma *et al.*, 2014). It is therefore possible that the presence of the fence dampened these vibrations and the bugs felt safe inside the fence. And what is more no fish could get into the fence and this is an other reason the bugs could feel safer.

Noteridae is a sister group of Dytiscidae. They are therefore closely related and even have the same way of movement in the water. They often swim below the surface of the water and move the limbs simultaneously. Noteridae larvae can use oxygen from the aerenchyme of the aquatic plants. That also allows them to make an underwater cocoon full of air (Thorp and Rogers, 2015). Noteridae are water beetles that actively dig in mud, they are called burrowing beetles. Thus, they are exposed to intense predation pressure as carp also dig in mud. Unlike Dytiscidae, that usually catch a prey in the water column (Thorp and Rogers, 2015). That may be why we found most of the Noteridae living inside the fence.

Family Corixidae was clearly the most abundant group caught. Although the Schilling's (2009) study points to the sensitivity of this group to the presence of fish, it was shown in our experiment that the occurrence of Corixidae is not affected by the presence of fish. The most common species of the Corixidae family was clearly *Sigara falleni*. It is a small, phytophagous species that feeds mainly on detritus and plants. Although it is a good swimmer and abundant in pelagial, it is probably too small for fish and is not so affected by their occurrence. Both Corixidae and Notonectidae are excellent swimmers. However, Notonectidae are predatory and also larger in size. Their active hunting and body size are probably the ideal combination to be exposed to the risk of the predation. That could be the reason we found more Notonectidae inside the fences, while Corixidae were not so abundant.

6.1.2 Effect of seasonality

Although the examination of the seasonality of individual groups of aquatic invertebrates was not the subject of the experiment, there appeared some differences in species composition of samples across the season. Some groups peaked in the first half of the season (Naucoridae), while others in the late summer (Coenagrionidae). The abundance of Notonectidae was higher at the beginning of the season and then gradually decreased. The trend was opposite in two groups (Gerridae, Libellulidae). The frequency of Corixidae was relatively stable and very high during the season. However, the numbers of individuals of Corixidae varied widely between the localities.

6.1.3 Differences in species composition between the localities

Finally, we investigated whether there were differences in the species composition between the localities. And whether there were differences in the species composition inside and outside the fences at the individual locality. There are some differences in species composition across localities, but not very significant. This is probably due to the fact that the aim was to select sites that would be very similar to provide similar conditions for the occurrence of aquatic invertebrates. The effort was to increase the attractiveness of a small area of otherwise breeding, unattractive pond. The species composition inside and outside the fence did not differ much. There appeared just three groups of the aquatic invertebrates that preferred safe microhabitats inside the fence.

6.2 The changing world

In the context of the changing world (climate, landscape, water use and environmental policy) we can now perceive ponds as a biodiversity hotspots (Céréghino *et al.*, 2014). However, the current intensive management of the ponds causes the degradation of these valuable habitats, resulting in a loss of aquatic invertebrate species. It is proven that the breeding ponds under the extensive management can offer more ideal environment than the ponds that are under special protection. In any case, heterogeneity is important at various scales: heterogeneity of fish stock, heterogeneity and sufficient cover of littoral vegetation, heterogeneity of vegetation around the pond (shrubs, trees, meadows, other water surface) and last but not least also heterogeneity of the whole pond systems. In her study Buczynska et al. (2007) describes the occurrence of a large number of aquatic invertebrate species on a system of breeding ponds that are surrounded by wetlands, streams, canals, forests and meadows. They are the subjects of the extensive management. There are even more rare species of the aquatic invertebrates on this pond system than in the nearby protected reserve where fish farming is prohibited (Buczynska *et al.*, 2007). The aim of our experiment was to create heterogeneous islands on an otherwise homogeneous pond with an extreme amount of fish and to find out if such a small area can positively influence the occurrence of the aquatic invertebrates on the intensive ponds. It turned out to have a positive effect on some species. They were mainly the most mobile groups. It can be assumed that the colonization of these islands will continue in the following years. The conservation goal should not be to put everything under the protection, but to try to understand the interactions in the pond communities, to use the knowledge to protect them by increasing the heterogeneity of their environment on many levels and to move from intensive management to extensive.

7 Conclusion

Twelve ponds under the intensive management in three regions of the Czech Republic were selected for the experiment. By installing fences and floating islands of vegetation, we tried to simulate more ideal environment for the aquatic invertebrates, even on a small area. The aim was to determine whether even such a small ideal area will have a positive effect on the occurrence of the aquatic invertebrates. The ponds were chosen to have the closest parameters possible and thus offer a similar environment for the aquatic invertebrates. The most important characteristics of the ponds were: intensive pond management, intensive fish stock and minimal littoral coverage.

It turned out that most species did not respond to the presence or absence of fish. However, we found that aquatic Hemiptera were highly selective concerning the habitat. They preferred fences and inhabited micro habitats in the form of submerged vegetation, which was natural. The vegetation planted by us did not attract them because it did not have the suitable parameters. The species composition varied only slightly across the ponds. That indicates the localities were very similar. The species we picked were mostly pioneer species and fast settlers, that disperse quickly.

It is obvious that the experiment could be extended in many ways, for example by involving several groups of aquatic invertebrates or amphibians. However, the aquatic invertebrate communities are very complex and we know very little about their interactions. There is certainly a need of a further exploration in the field of intraspecific and interspecific interactions across the aquatic invertebrate communities in order to understand them and to improve their protection.

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ČÚZK: Nahlížení do katastru nemovitostí, <https://nahlizenidokn.cuzk.cz>.

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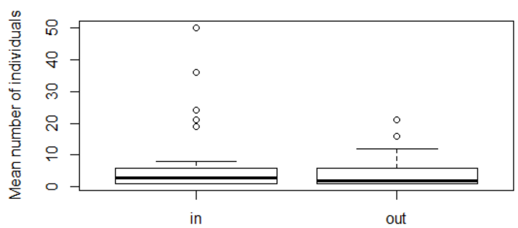
Mapy.cz: Zobrazení jednotlivých rybníků v kontextu krajiny.

9 Attachment

Attachement no. 1:

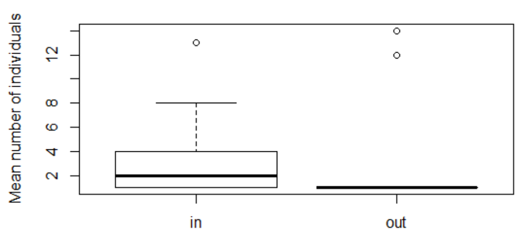
The following graphs show the other groups of aquatic invertebrates, where the effect of the fence was not demonstrated (*graph no. 4a – 4h*).

Coenagrionidae



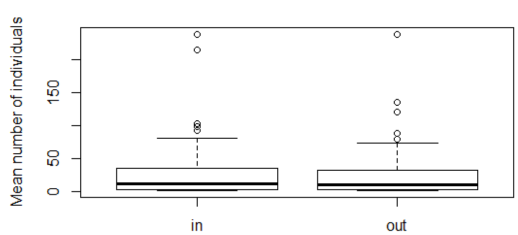
graph no. 4a: for the family Coenagrionidae the effect of the fence was not demonstrated

Hydrophylidae



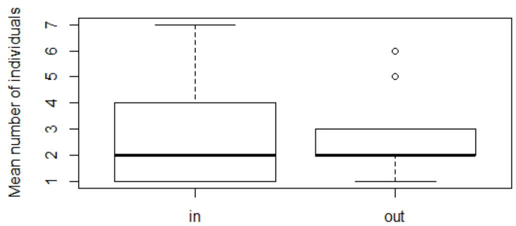
graph no. 4b: for the family Hydrophylidae the effect of the fence was not demonstrated

Corixidae



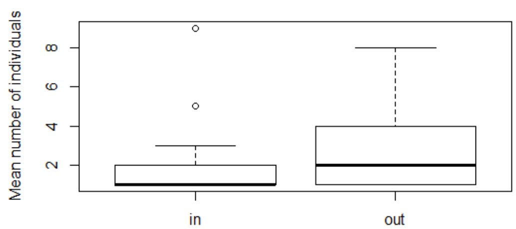
graph no. 4c: for the family Corixidae the effect of the fence was not demonstrated

Libellulidae



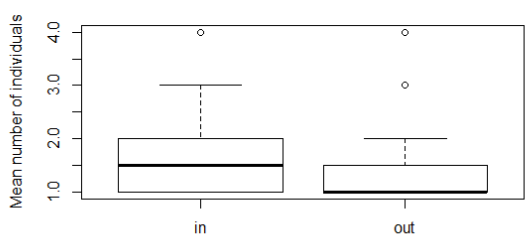
graph no. 4d: for the family Libellulidae the effect of the fence was not demonstrated

Naucoridae



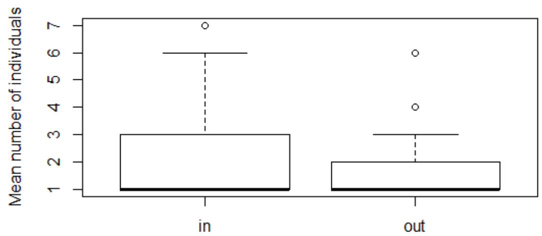
graph no. 4e: for the family Naucoridae the effect of the fence was not demonstrated

Nepidae



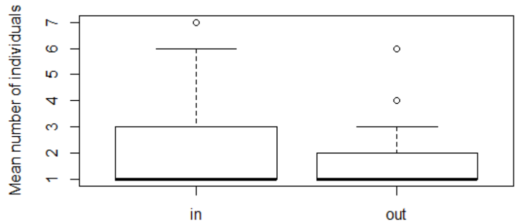
graph no. 4f: for the family Nepidae the effect of the fence was not demonstrated

Dytiscidae



graph no. 4g: for the family Dytiscidae the effect of the fence was not demonstrated

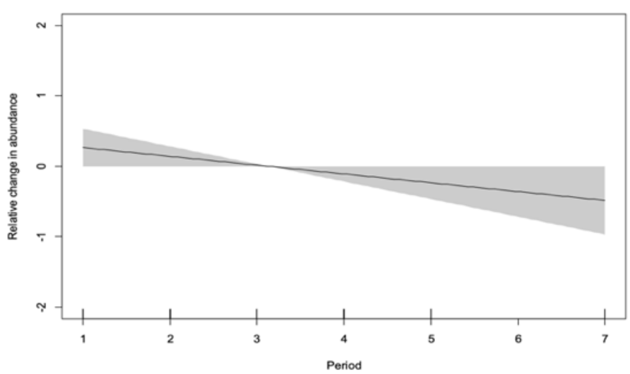
Aeshnidae



graph no. 4h: for the family Aeshnidae the effect of the fence was not demonstrated

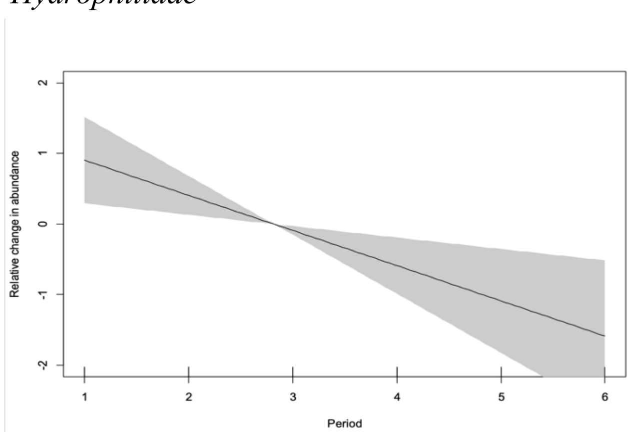
Attachement no. 2:

Dytiscidae



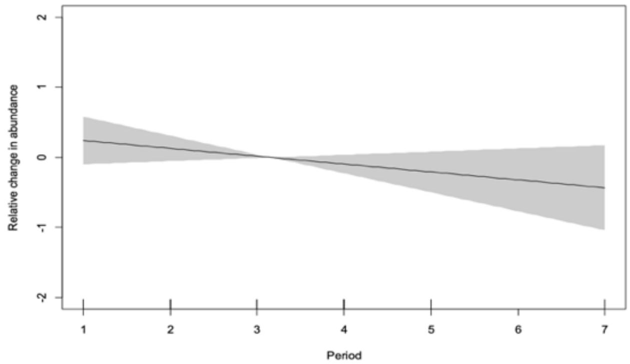
graph no. 5a: the relative abundance of individuals decreases in the season

Hydrophilidae



graph no. 5b: the relative abundance of individuals decreases in the season

Noteridae



graph no. 5c: the relative abundance of individuals decreases in the season

Attachement no. 3:

picture no. 1: the fence withou vegetation

picture no. 2: the fence with artificial vegetation

picture no. 3: live catch trap with chicken liver inside

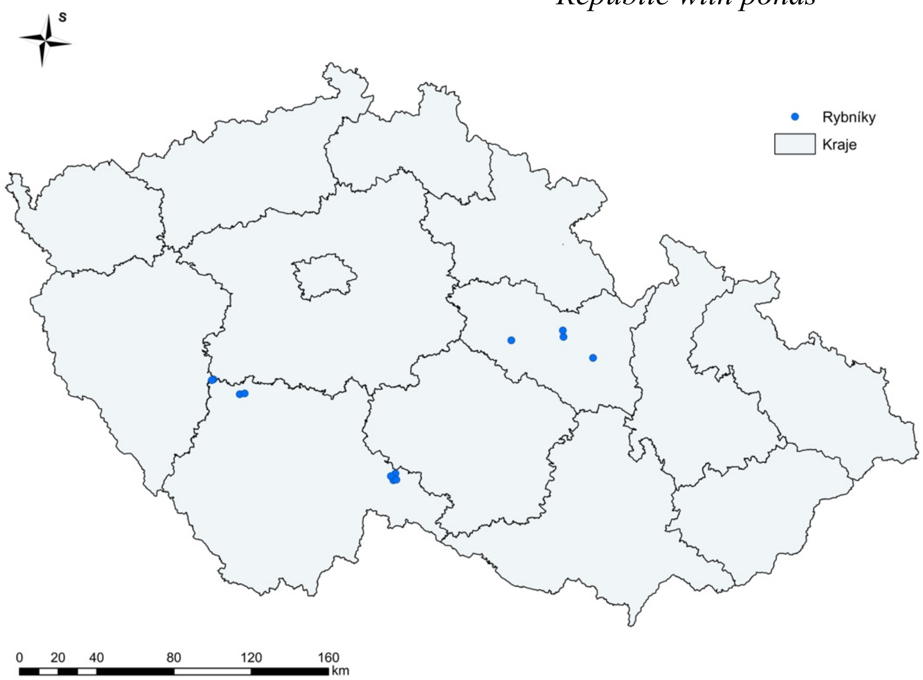
picture no. 4: field workspace and sweeping net

picture no. 5: marked sample

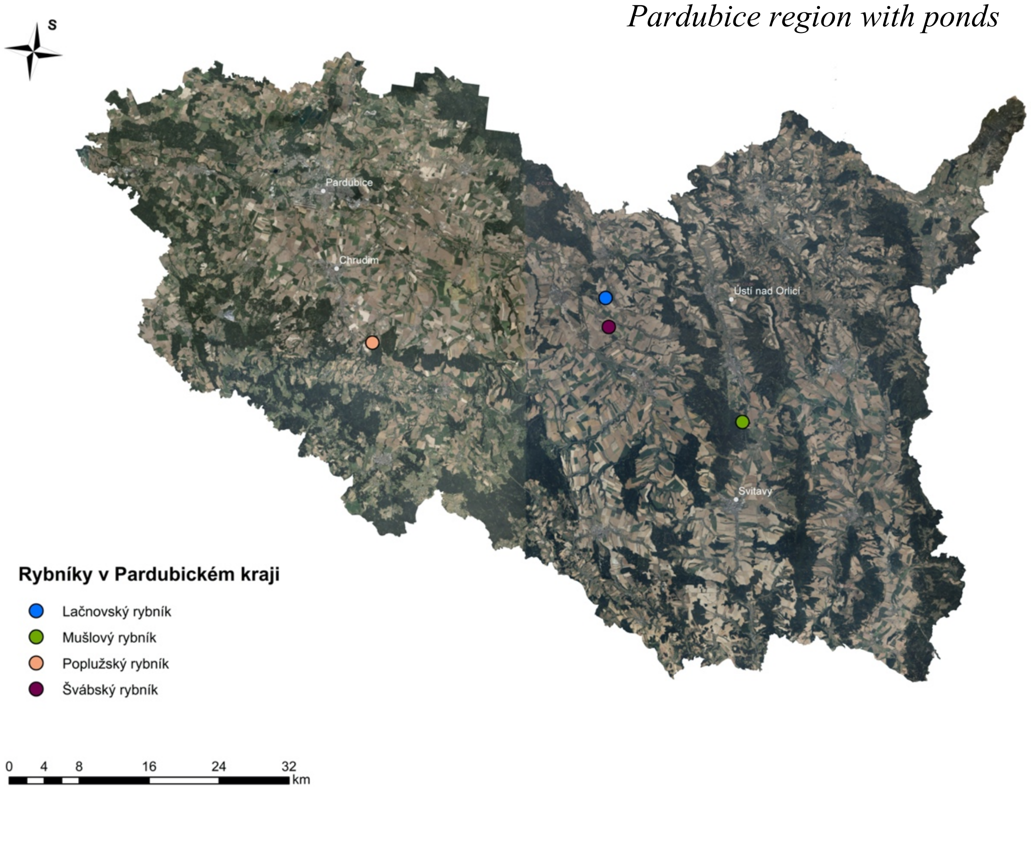
picture no. 6: Nepa cinerea under binocular magnifier

Attachement no. 4:

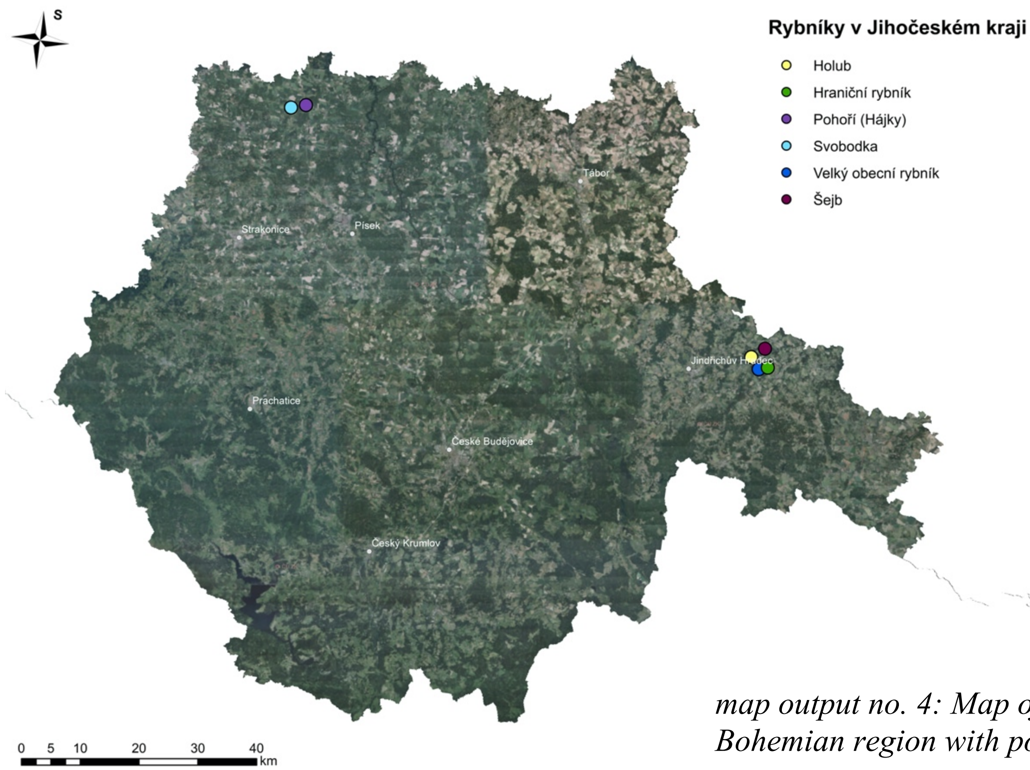
map output no. 1: Map of the Czech Republic with ponds



map output no. 2: Map of the Pardubice region with ponds



map output no. 3: Map of the South Bohemian region with ponds



map output no. 4: Map of the Central Bohemian region with ponds