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Faculty of Tropical AgriSciences

Decomposition of Cadavers in Freshwater Environment and Its Use in Forensic Practice

Master's thesis

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Declaration

I Bc. Monika Jirsíková hereby declare that this thesis entitled Decomposition of Cadavers in Freshwater Environment and Its Use in Forensic Practice is my own work under the direction of the supervisor of the thesis and by using literature and other information sources that are cited in the work and listed in the references at the end of work. As the author of the mentioned thesis I further declare that I am in connection with its creation did not violate copyright of third parties.

In Prague 20. 4. 2016

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Abstract

Diploma thesis deals with the issue of the determination of post mortem interval (PMSI) in dead bodies found in the aquatic environment. Literature research collects the available information about the bodies found in the water. The experimental part describes a trial whose purpose was to verify the data reported in the literature, also to map the actual process of decomposition as well as certain, which invertebrates were involved in degradation of bodies in the aquatic environment of the Czech Republic and finally, assess the data utilization for forensic entomology. For this purpose two freshwater localities were selected: the first with running water and the second with stagnant water. During the experiment purpose 14 rabbit carcasses were used each weighed about 2,500 g and weighted at the bottom of the water source. At the beginning of the experiment, all carcasses in both variants were simultaneously exposed. Subsequently, two cadavers were extracted at predetermined intervals, one from each variant, and evaluated. Sampling interval was given once a week. On all cadavers, the taphonomic changes and activity of the invertebrates were observed. Every organism found outside or inside the body was secured and subsequently determined. The water sample from the vicinity of the carcass was ensured at each sampling and analyzed. The water temperature at both sites was measured three times a day. In river locality: 110 *Gammarus fossarum* (Koch in Panzer, 1835), 220 *Lymnaea stagnalis* (Linnaeus, 1758), 276 *Planorbarius corneus* (Linnaeus, 1758), 127 *Erpobdella octoculata* (Linnaeus, 1758), 2 *Calopteryx splendens* (Harris, 1780), 3 *Asellus aquaticus* (Linnaeus, 1758) and 18 *Tubifex tubifex* (Müller, 1774) were secured. In pond locality: 10 *Gammarus fossarum*, 8 *Erpobdella octoculata* and 128 *Tubifex tubifex* were secured. In pond locality 75 tadpoles of *Bufo bufo* (Laurenti, 1768) and 41 tadpoles of *Rana temporaria* (Linnaeus, 1758) were secured. There were no serious chemical changes of the water mass in the vicinity of the cadavers.

Keywords: animals, forensics, decomposition, carcass, aquatic environment

Contents

1 Introduction

1.1 The Forensic Entomology

Forensic entomology is the continually evolving forensic discipline and it is recognized by the courts throughout the world. In its broadest sense, forensic entomology is *sensulato* the study of insects and other invertebrates involved in any legal action. Forensic entomology can be divided into three categories: urban, stored product and medicolegal (Catts and Goff 1992, Hall 1990). In the lastly named category the knowledge about invertebrates is utilized. Particularly the fact that insect species are attracted to decaying organisms and actively involved in decomposition process. The main task of forensic entomology is to use these findings in investigation of violent crimes such as murders, suicides, rapes, as well as physical abuse. The primary purpose of forensic entomology today is to determine the time of insect colonization of the corpses and consequently elapsed time since their death until body finding, however, the insect evidence can also reveal that the body has been moved to a another sites after death, or that the body has been disturbed at some time (Anderson, 1995). Most cases that involve a forensic entomologist are 72 h or more old, as up until this time, other forensic, mainly legal medicine methods are equally or more accurate than the insect evidence. Nevertheless, after three days, insect evidence is often the most accurate and sometimes the only method of determining elapsed time since death, capable to determine the time of death with the accuracy of the hours.

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1.2 Decomposition of the Body on the Land

The most important factor influencing the rate of decomposition processes is the temperature of the environment on which depends the speed of enzymatic processes associated with the decomposition of the dead body (Laupy, 1994). Among other variable factors belong the presence and amount of trauma on the body of the corpse, the activity of predators and rodents, moisture conditions, presence or absence of clothing and weight of corpse (Laupy, 1994, Šuláková, 2006). It is also very important to evaluate the extent to which is access of necrophagous insects limited (Smith, 1986).

According to Daněk (1990) in the Palearctic region, concretely in Central Europe, we can establish eight consecutive waves, during which necrophagous fauna colonize a freely exposed dead body. Number of phases varies geographically, for example in south Europe is counted with five phases.

In conditions of the Czech Republic, Šuláková (2006) states that the most optimal way of classification is into six consecutive succession waves and phases of decomposition:

1. The first wave - the state of the corpse: fresh

Typical for the first wave are two groups of invertebrates. To the first group wasps and ants of the order Hymenoptera belong and the second group is formed by higher Diptera, blow flies of the family Calliphoridae (Šuláková, 2014). Hymenopterans last on the corpse for only necessary required period when they receive food therefore they are not important from a forensic point of view. Flies usually appear immediately after death, attracted by the smell of fresh meat, blood and sweat. If the victim is helpless and bleeding, flies could lay their eggs on still living body. The speed of colonization of the dead body by the first wave of necrophagous fauna, depend on its availability (e.g. colonization by insects outside will be faster than in a closed flat). The most important representatives of the family Calliphoridae are mainly: *Calliphora vicina* (Robineau-Desvoidy, 1830), *Lucillia sericata* (Meigen, 1826), *Protophormia terraenovae* (Robineau-Desvoidy, 1830), and *Phormia regina* (Meigen, 1826) Also members of the family Muscidae can appear, from these especially synantropic species of housefly *Musca domestica* (Linnaeus, 1758) (Daněk, 1990), which, however, only occurs near a person e.g. households (Šuláková, 2014). Next on fresh corpses, we can discover representatives of the order Coleoptera (beetles), namely family Carabidae (ground beetles) and some species of wasps, earwigs and ants (Daněk, 1990, Šuláková, 2014).

2. The second wave - the state of corpse: bloated

This phase starts at the moment when gaseous substances of bacterial decomposition inside the body begin to form. They cause the bloating of the body and its strong odor. A crucial factor is the ambient temperature. At high temperatures in the summer can to bloating occur within hours (Šuláková, 2014). Attracted by the strong smell representatives of the families Muscidae and Sarcophagidae (flesh flies) appear. However in conditions of Central and Northern Europe their discovery on freely laying corpse is very rare phenomenon, but are typically present on corpses in flats. The most important representatives are *Sarcophaga carnaria* (Linnaeus, 1758) and *Sarcophaga argyrostoma* (Robineau-Desvoidy, 1830), which according to Šuláková (2014) forms 95 % of sampled larvae of Sarcophagidae family in conditions of the Czech Republic. At the same time, there is still ongoing destruction activity of larvae from the first wave. Smelly gases attract necrophagous beetles (Coleoptera). Primarily carrion beetles (Silphidae) of which *Necrodes littoralis* (Linnaeus, 1758) and genus *Thanathophilus* are the most considerable because usually reproduce on the body (Šuláková, 2014). Genus *Nicrophorus,* e.g. *Nicrophorus humator* (Gleditsch, 1767) or *Nicrophorus germanicus* (Linnaeus, 1758), and small round fungus beetles of the family Leiodidae can also appear (Šuláková, 2006) but they are less forensically significant (Šuláková, 2014). The first representatives of infraclass Acari (mites) are also emerging. Subsoil beneath the dead body is beginning to change, this fact is particularly apparent in plants which lose chlorophyll and its overall growth is slowing (Daněk, 1990).

3. The third wave - the state of corpse: biochemically active

The third phase is characterized by the liquefaction of the substrate (corpse) and includes two processes: saponification and fermentation. The process of saponification occurs in fat, with simultaneous development of volatile fatty acids, particularly butyric acid (Šuláková, 2006). In response of the butyric acid odor react in the Czech Republic frequently represented *Hydrotaea ignava* (Harris, 1780). Beetles attracted by smell of butyric acid are recorded representatives of the family Staphylinidae, especially *Creophilus maxillosus* (Linnaeus, 1758) and the first representatives of families Dermestidae (skin beetles) and Histeridae (clown beetles). On the corpse, the destructive activity of fly larvae from first and second wave continue, especially with undressed corpses, may larvae in 2-3 weeks digest major part of the tissue. Colonization by other invertebrate species depends on many factors, *e.g.* whether was a dead body dressed or whether perforation of the abdominal cavity occurred (Daněk, 1990).

Shortly after fermentation of fat, the protein fermentation occurs; which is also called "cheese fermentation" because caseous substances, which are produced, stink similar to a soft-ripened cheese (Daněk, 1990). "Cheese odor" attracts especially cheese skippers (Piophilidae), namely *Stearibia nigriceps* (Meigen, 1826) (Šuláková, 2014) and the fruit fly (*Drosophila funebris* (Fabricius, 1787)). At this time also peaks the incidence of beetles of the family Cleridae (checkered beetles) and Dermestidae. Adults of these beetles are attracted by the smell of the saponification processes in the third wave, and just in the fourth wave the incidence peaks, especially on the drier parts of the corpse. Typical representatives for the Czech Republic are: *Dermestes frischii* (Kugelann, 1792), *Dermestes undulatus* (Brahm, 1790), *Necrobia violacea* (Linnaeus, 1758) and *Necrobia rufipes* (De Geer, 1775). Proportionally to loss of muscle and other soft tissues of the dead body, decreases qualitative and quantitative number of typical necrophages, particularly from the family Silphidae. However, active biochemical decomposition can be considered as a single complex succession stage, because saponification of fats and cheese fermentation can occur simultaneously on different parts of the corpse and therefore species in this wave can colonize the corpse in different order (Šuláková, 2014).

4. The fourth wave - the state of corpse: advanced decomposition

The dead body of this phase reaches the stage of ammonia fermentation. This succession wave is characterized by the presence of representatives of the family Phoridae (scuttle flies), which are attracted by released ammoniac vapors and caseous substances. These small flies settle in substances of protein nature subject to putrefactive processes. Adults of typical necrophages can be found only in small numbers. Noticeably has diminished the number biophages as a direct result of depletion of food resources. In the subsoil of the corpse, the pupae of beetles could be found, newly hatched specimens and dead specimens (Daněk, 1990).

5. The fifth wave - state corpses: desiccation

This period usually occurs at the end of the first year and second year of corpse decomposition. Soft tissues of the corpse are drying up and the state leads to its gradual mummification. At this time, the genus *Trox* (Trogidae) and keratophagous beetles are found mostly under the drier parts of corpses, such as bone cavities of the large bones, dried skin, hair cover, *etc*. Typical necrophages occurs very sporadically because of the current state of the corpse, which does not produce enough food for them or their offspring. However, small rove beetles (Staphylinidae) still persist. Daněk (1990) state that subsoil of the corpse at this stage gets gradually to original form. As a consequence of these changes sequentially disappear the various saprophages of families Hydrophilidae (scavenger beetles), Scarabaeidae (scarab beetles) and in particular certain species of genus *Aphodius* and *Ontophagus*, which have filled by saprophytic lifestyle the additional place in the food chain. On the remains of corpse appear different kinds of mites that feed on proteins of animal origin, invade the bone marrow and accelerate the disintegration of the bones. These mites are mainly from family Uropodidae (Daněk, 1990).

6. The sixth wave - the state of corpse: dry remains

The corpse is already completely skeletonized, and on the skeleton rarely appear only dry residues of bowels, sometimes pieces of leather, dried muscle tissue and damaged cartilage (Šuláková, 2006). The insect that attacks the dry carcasses, bones, horns, leather, *etc*. occur. A typical representative is the larder beetle, *Dermestes lardarius* (Linnaeus, 1758).Larder beetles are mostly thermophilic and xerophilous species, which can be found on the corpse only if it was a long time in a confined space. An example might be attic, warehouse or abandoned apartment (Daněk, 1990).

1.3 Decomposition of the body in the water

With possibly some exceptions, there are no truly saprophagous aquatic insect species which are able to feed exclusively on carrions. It is in contrast with terrestrial indicators Diptera species (blow flies, flesh flies) that often provide a time frame of succession and assist in determining of post mortem interval. This fact is the main reason of search for other methods that will be able to refine the post mortem interval with the bodies found in the aquatic environment. Factors affecting postmortem condition in aquatic environments are numerous. Very important is temperature of the water source. Similar to terrestrial decomposition, high water temperature accelerates the decomposition and decomposition in cold water is slower. It implies that temperature of water source has huge effect on the PSMI sometimes even in order of tens of days. Oxygen level in the water also is an important factor to consider when determining the PSMI. Another important factor is as well as on the land the presence of clothes (Smith, 1986).

1.3.1 Fresh Water

In contrary to seven settled stages of decomposition on the land are in the freshwater environment recognized just five stages. According to Byrd and Castner (2009):

1. Submerged fresh stage

The period of time which starts with submersion of the carcass and end when carcass begins to bloat and rise to the surface. Timeline varies with geographic location, water sources microhabitat, size of the carcass, *etc*. (Byrd and Castner, 2009).

2. Early floating stage

Due to the activity of anaerobic bacteria the large amount of putrid gases forms in the abdomen and pushed the abdomen (together with whole body) to the surface. In cases where the body is not burdened or anchored to the bottom, provides fertile feeding and breeding ground for terrestrial Diptera species. In this phase is also very significant algae growth (Byrd and Castner, 2009).

3. Floating decay stage

Cadavers in stage of floating decay (reaching water level) are extensively colonized by terrestrial fly species. Dead bodies have minor signs of decay. Flesh is sloughed, significant loss of muscle tissue is present, bones are separated from the joints, eyes and soft tissues lost their structure. Identity of a carcass is still evident (Byrd and Castner, 2009).

4. Advanced floating decay stage

Timeline of stage extremely varies with environmental influences. In carcasses is visible major deterioration, skull is revealed, bones of limbs are revealed and even missing. Eyes are usually missing, soft tissues are watery. Identification of a carcass becomes indistinguishable. Advanced floating decay stage ends with sinking of remains under water level (Byrd and Castner, 2009).

5. Sunken remains stage

Again, the duration of stage is variable. The main characteristics is that bones and bits of skin remaining in the bottom substrate (Byrd and Castner, 2009). Payne and King (1972) noted that decomposition is completed by bacteria and fungi during this stage.

1.3.2 **Salt Water**

It is extremely difficult to determine the elapsed time since submergence or death as so little is known about parameters of decomposition in the ocean (Anderson, 2008). The body is exposed to many changes in the surrounding temperature, pH and salt content. The longer the body was immersed, the more is difficult to determine the elapsed time since death. These factors make determination of the time of death a difficult process. Wentworth *et al.* (1993) state that so far the PSMI tends to be established by police inquiries into time last seen alive and not by examination of the body. The marine environment is the only habitat, which is not dominated by insects (Anderson and Hobischak, 2002). However, it is home to a broad range of invertebrates, therefore it is very likely that they will colonize in the same sequence as insects (Brusca and Brusca, 1990). Crustaceans, fish, gastropod mollusks, and echinoderms are the dominant scavengers in marine environments (Sorg *et al.*, 1997). In the ocean, carcasses may remain relatively intact for weeks (Anderson and Hobischak, 2004; Anderson and Hobischak, 2002), or can be scavenged very rapidly (Anderson, 2008)

1.4 The Water Decomposers

1.4.1 The Diatoms (Diatomeae syn Bacillariophyceae)

Diatoms are a large group of photosynthetic organisms. They can exist as unicellular organisms or form colonies with various shapes. Most distinct feature of diatoms is their shell, called frustule made of silicon dioxide hydrated with a small amount of water - ($Si0_2$ + H_20) (Round *et.al.*,1990). Diatom growth is completely determined by the content of silicon in the environment. Reproduction is asexual (significantly more frequent) or sexual. Diatoms are a very important part of the global primary production and one of the main aquatic photosynthetic groups (Round *et al*, 1990). Diatoms form a monophyletic evolutionary line. Subphylum Diatomeae counts more than 285 genera of recent diatoms, with approximately 100,000 extant species (Round *et al*, 1990). Diatoms inhabit various environments. Most species are pelagic or benthic in open water (oceans, rivers, lakes), but some species can live even under humid atmospheric conditions in soil. In conditions of the Czech Republic, they usually form noticeably developed biomass in banks of rivers, where produces visible reddish brown coatings on rocks or mud on the shore. As producers, diatoms play important role in food chain, they´re also excellent bio indicators especially in rivers. Decomposition and decay of diatoms creates siliceous ooze (siliceous pelagic sediment covering the deep ocean floor) (Burdige, 2006).

1.4.2 The Ramshorn Snails (Planorbidae)

Planorbidae is the family of aquatic snails, which use to breathe instead of gills perfused jacket wall and therefore breathe atmospheric oxygen (Burnie, 2002). In most cases, they inhabit fresh water with a few exceptions that are able to survive in water brackish. The shell of most species is levorotatory, smooth and relatively thin, and it is not provided with permanent lid (operculum) and reaches a height up to two centimeters. The body of ramshorn snail has characteristic reddish color, which is caused by the presence of hemoglobin in the hemolymph (Stankowitsch, 1992). The main component of the food, consist from algae, aquatic plants and organic residues. Planorbidae as well as Lymnaeidae are intermediate host of cercariae of various species of flukes (Trematoda). Best known and also the largest representative of the family in the Czech Republic is the great ramshorn (*Planorbarius corneus*, (Linnaeus, 1758)) (Dogel, 1961).

1.4.3 The Pond Snails (Lymnaeidae)

Representatives of this family are widely used in most of European rivers, lakes and smaller ponds. Shells, which reach a height of up to seven centimeters, are not provided with a permanent cap and are dextrorotary. Breathing is realized through perfused jacket wall as well as in the other gastropod from the order Bassomatophora. Significant is also the fact that these organisms can become intermediate hosting article in the life cycle of cercariae to 71 species of flukes (Trematoda) (Volf and Horák, 2007).

1.4.4 The Freshwater Mussels (Unionidae)

Members of the family Unionidae occur in North America, Europe, Asia, Africa, and the Indonesian Archipelago (Graf and Cummings, 2002). Freshwater mussels are bivalves belonging to the order Unionoida. Unionids are among the longest lived animals, with most species living for decades and some of them even over a century (Nowak and Kozlowski, 2013). The order Unionidae includes around 1,000 species worldwide (Bauer and Wachtler, 2001), but the exact number is currently disputed. Molusks of Unionidae family are acephalic, with two calcium carbonate shells. Instead of byss gland they have a foot and do not have true siphons. Individuals vary in shape, size and coloration, but are bilaterally symmetrical. Embryonic stages develop within the marsupia (brood pouch), or specialized portions of the gills, of the female. Once fully developed (glochidias), they are released from the female and must attach to the gills or fins of a fish host within a few days or they die. Many Unionids are species specific. When the glochidias are attached, the hosts dermal tissue, encapsulate each glochidium and forms a nodular cyst. Encapsulated glochidia pass through metamorphosis. Metamorphosis usually takes 10 – 30 days, when finished the glochidia breaks through the capsule. Last stage of development takes place in the sediment of water source and by the species lasts from one to eight years. Adult individuals can range from 30 to 250 mm. (Smith, 2001). Unionids inhabit permanent freshwater sources (rivers, lakes, ponds). The usual water depth at the site of occurrence is within two meters. Fresh water mussels are long-lived, living an average of 10 or more years (Cummings and Mayer 1992). Most of the mussels are sedentary, but they are capable of a restricted form of locomotion for which they are using a foot. Adult freshwater mussels are filter feeders; they continuously filter food particles out of the water (Watters 1998; Allen 1921).

1.4.5 **The Leeches (Hirudinea)**

This class is characterized by segmented, oval body and by the presence of two suction pads, the first on beginning and the second at the end of the body (the front is larger than the rear). The number of body segments of the leech is 33 or a multiple of that number, concerning the secondary articulation surface of the body. Leeches range in size from 1 to 30 cm, depending on the species competence (Volf and Horák, 2007). They inhabit both fresh and salt water and some of them even live on land. Most species feed on parasitic way - by sucking the blood of vertebrates. Smaller amounts of predatory species feed on small fish and invertebrates. In this class we distinguish five recent orders of leeches. Sychra and Schenková (2009) reported that in Europe today over 90 species of this class is known. The Czech Republic has been confirmed a total of 24 species of leeches, one of which is our most abundant leech *Erpobdella octoculata* (Linnaeus, 1758), belonging to family Erpobdellidae in which we classify three other species living in the Czech Republic, namely *Erpobdella nigricollis* (Brandes, 1900), *Erpobdella testacea* (Savigny, 1822), and *Erpobdella vilnensis* (Liskiewicz, 1925) (Sychra and Schenková, 2009).

1.4.6 **The Sludge Worms (Naididae)**

Naididae formerly Tubificidae are family of small 30-40 mm long annelids. A distinctive feature is the red color of the body, caused by the presence of hemoglobin and frequent asexual reproduction. Representatives of family Naididae have only one pair of testes and ovaries. Breathing ensures rear part of the body while the front part is usually drilled in the mud. Individual articles are covered with soft bristles which help to locomotion. The sludge worms inhabit slowly flowing and standing waters and are able to tolerate organically polluted water. They feed on organic debris. The most interesting representative living in waters of the Czech Republic is the sludge worm (*Tubifex tubifex* (Müller, 1774)) (Zahradník, 2007).

1.4.7 The Scuds (Gammaridae)

It is a large species numerous family of crustaceans from the order Amphipoda. These crustaceans exclusively inhabit aquatic (both freshwater and saltwater) environment. The front legs are modified for different activities, two pairs are used for capturing prey and digging, five other pairs of limbs are used for walking. They have a relatively long abdomen and lamellar telson, which is divided into two lobes. The development is direct. Amphipods are scavengers or predators. The order Amphipoda includes about 6,000 species, from very small benthic representatives up to 25 cm large pelagic forms (Brusca and Brusca, 1990). In the Czech Republic we can find 17 amphipod species from which are three representatives of family Gammaridae. *Gammarus fossarum* (Koch in Panzer, 1835), *Gammarus pulex* (Linnaeus, 1758), and *Gammarus roeseli* (Gervais, 1835) (Hammer *et al*., 1995).

1.4.8 The Caddisflies (Trichoptera)

Representatives of this order belong to the group of insects with a perfect metamorphosis (Holometabola). Their development takes place in the aquatic environment, so the adults reside near waters. The adults are similar to a moth. Their body and two pairs of wings are covered with short setae or scales. Orthognate head carries two pairs of filamentous antennae, which are as long as the body or longer. Well known fact about caddisfly larvae is, that their freshwater larvae inhabit either the tunnels that form in the mud or build a fixed or mobile boxes. The boxes are made of grains of sand, wooden chips and other material, which is glued together with silk. Caddisfly larvae inhabit slow flowing rivers, ponds and lakes. The larvae are herbivorous or catch plankton or other insect larvae. The order includes 47 families with 608 genera and 13,574 described species (Morse, 2011). In the Czech Republic, 262 species with documented occurrence (Chvojka and Komzák, 2008) can be found. Examples include *Philopotamus variegatus* (Scopoli, 1763) (Dogel, 1961).

1.4.9 **The Damselflies (Zygoptera)**

The damselflies from suborder Zygoptera are hemimetabolous insect found worldwide (Gordh and Headrick, 2011). Both nymphs and adults feed on other insects. Nymphs are aquatic living in a variety of freshwater habitats. During the development the nymph mould repeatedly and during the last mould they climb out of the water and finish the metamorphosis. Physical appearance is quite similar to dragonfly (Anisoptera). Compound eyes are large. The major part of the diet of the most species appears to be crustaceans such as water fleas (Heckman, 2008).

1.4.10 **The Stoneflies (Plecoptera)**

Most species of order Plecoptera can be found in rivers and streams of temperate and cold climatic belt of both hemispheres. The number of species in order exceeds 3,500 (Zwick, 2000). Stoneflies are very sensitive to water pollution. Head is orthognathic or prognathic. Mouthparts are chewing, in some species reduced. Nymphs are aquatic and inhabit the benthic zone of water sources with high oxygen level. Physiological appearance of nymphs is quite similar to adults, except of external gills. Stoneflies remain in nymphal stage from one to four years and undergo up to 33 molts before emerging and becoming adults. Larvae are usually carnivorous, less often herbivorous or detritivorous. The cannibalism occurs often. Lifespan of imagos is only couple of hours to couple of weeks. In the Czech Republic, 115 species can be found. The most common is *Perla burmeisteriana* (Classen, 1936) (Daněk, 1990).

1.4.11 **The Mayflies (Ephemeroptera)**

Worldwide are known more than 3,000 species are known (Brusca and Brusca, 1990). Together with dragonflies and damselflies belong to ancient group called Paleoptera. Mayflies are quite primitive, hemimetabolous insect. Nymphs occur exclusively in freshwater (except of few brackish species). Mayflies are morphologically and behaviorally diverse, and the larvae have been grouped into four life forms according to Ward (1992): (1) swimming, (2) creeping and climbing, (3) flattened and streamlined, and (4) burrowing. Each nymph comes through series of instars (stages) among the mites body size increases each time its. Final length ranges from 3 to 30 mm. The body is cylindrical and slightly dorsoventrally flattened. Head is covered with sclerotin and carries two large compound eyes. Mouth is designed for chewing. Thorax is segmented and each segment has a pair of legs. The abdomen is equipped with gills (up to seven pairs) and their location varies with species. The abdomen terminates with three slender thread-like spits (projections). In stage of subimago mayfly is not fully developed yet, but it physically resembles the adult. Main differences between subimago and imago (adult) are lack of coloration, cloudy wings and not fully developed genitalia. Stage of subimago lasts for 1 – 2 days. Imagos has two pairs of triangular wings, which cannot be fold over the abdomen, large compound eyes, flexible antennae, and nonfunctional mouthparts. The most important stage for forensic entomology is the nymph. Nymphs usually inhabit the rivers (streams) bottom sediments, layer of decaying vegetation or live under the rocks. Mainly they are detritivorous or herbivorous and feed on algae, detritus or fine particles in zones of decomposition. The best known representatives in the Czech Republic are: drake mackerel mayfly (*Ephemera vulgata* (Linnaeus, 1758)) and pond olive (*Cloeon dipterum* (Linnaeus, 1761)) (Zahradník and Severa, 2007).

1.4.12 **The Predacious Diving Beetles (Dytiscidae)**

Dytiscidae is a family of predatory beetles living in water for which they are perfectly suited. So far, 4,000 known species in 180 genera are recognized worldwide (Larson *et al.*, 2001).. The adults have dorsoventrally flattened oval body and strong fringed hind legs allowing skillful movement in the water. They grow up to seven centimeters. The darker colors dominate in representatives of the family. The significant sexual dimorphism is noticeable. In male beetles dominate smooth elythra while female elythra are edged. According to Zahradník and Severa (2007), in the Czech Republic, 130 species can be found, of which the most famous is the great diving beetle, *Dytiscus marginalis* (Linnaeus, 1758).

1.4.13 **The True Toads (Bufonidae)**

Family Bufonidae includes over 586 extant species in approximately 34 genera. Except of Antarctica and Australia they are native to every continent. Toads are mostly terrestrial, exceptionally live in stream and some species are arboreal. The adults have robust bodies and short limbs. Size ranges from 20 mm to 250 mm. Head is massive, jaws are strong and toothless (Zwach, 2008). Parotid glands contain a excrete peptide or alkaloid based toxins. Toxicity of poison varies with species. Skin is dry and covered with warts. Male toads possess a Bidder´s organ (ovary – like structure unique for Bufonids, which function remains unknown). Female toads lay eggs in short strands in stagnant water (typically smaller ponds or streams) (Zwach, 2008). In the Czech Republic, the common toad, *Bufo bufo* (Laurenti, 1768), is typical representative.

1.4.14 **The True Frogs (Ranidae)**

Ranidae family has the widest distribution from known frog families belonging to order Anura (Zwach, 2008). Number of species is over 600 in 23 genera. Representatives of this family occur in all continents, except for Antarctica. Features common for all species are smooth and moist skin, slim bodies and long legs with webbed feet. Ranidae vary with size, from really small representatives for example from *Pelophylax lessonae* (Camerano, 1882) found in our conditions to the largest frog in the world the goliath frog (*Conraua goliath* (Boulenger, 1906)). Significant number of species is aquatic or semi-aquatic. Adult females lay eggs in clumps, singly or in floating films (Zwach, 2008). After being laid the eggs undergo the metamorphosis. The length of this cycle is species specific, it can last from several days to over a month and even up to one year. Frog spawn hatches into larval stage, called tadpoles. Freshly hatched tadpoles have gills and the body is oval ended with tail. During the maturation hind legs appears first, together with increasing age the tadpole´s tail is reduced and finally completely absorbed. In early days tadpoles feed on algae or aquatic plants, later in development they become carnivorous and feed on plant-meat based diet. Representatives found in the Czech Republic are *Rana temporaria* (Linnaeus, 1758), *Rana arvalis* (Nilsson, 1842), *Rana dalmatina* (Fitzinger in Bonaparte, 1839) (Zwach, 2008).

1.4.15 **The Catfishes (Siluriformes)**

Order Siluriformes counts 36 extant catfish families, and about 3,093 extant species (Ferraris *et al*., 2007). Catfishes are very diverse group. Representatives of this family inhabit freshwater environment of all continents and prefer shallow running water. Catfish is typical bottom feeder, which is present at the bottom of the water source and seeks shelter in the pits, roots or even caves. Catfishes are very diverse group, but across families they share some basic features such as: barbells, absence of scales and mucous covered skin. The molecular evidence proved that the catfishes are a monophyletic group (Sullivan *et al.*, 2006). Sexual dimorphism is reported in about half of all families of catfish (Friel and Vigliotta, 2006). In the Czech Republic we can find two species a representative of the family Siluridae, the wels catfish (*Silurus glanis* (Linnaeus, 1758)) and the introduced brown bullhead, *Ameiurus nebulosus* (Lesueur, 1819) of the family Ictaluridae (Ferraris *et al.*, 2007).

1.4.16 **The Freshwater Eels (Anguillidae)**

This family includes 19 species of freshwater eels, characterized by a long, snakelike body, which is nearly circular in cross-section (Čihař *et al*. 1993). Dorsal, anal and tail fins create together the fin rim. The lateral line is very apparent. The body length reaches 50 cm in females and 150 centimeters in males. In relation to food intake the family Anguillidae, consist from two ecological forms: narrow-head eels feeding on small prey (insect larvae) and wide-head eels, which feed mainly on fish. Freshwater eels are known from specific way of reproduction, respectively catadromous migration, when they spend up to two years migrating through rivers to reach spawning grounds in the Sargasso Sea (Čihař *et.al.*, 1993). Hecker (2014) state that in the Czech rivers we can find only one representative – the European eel (*Anguilla anguilla* (Terofal and Militz, 1997)).

1.4.17 **The Pikes (Esocidae)**

Fish belonging to this family are found throughout all Europe. They are perfectly adapted to a predatory lifestyle. They are characterized by massive lower jaw, with long, sharp teeth pointing backwards (Čihař *et al*., 1993). The body of these animals is of slightly flattened on sides and almost equally wide from the rear end of the head to the base of the tail. Coloration is quite variable and depends almost exclusively on the environment. The total body length of an adult ranges from 140 cm to 150 cm. The weight may be up to 25 kg. The diet of pikes is composed almost exclusively of other fish species and intraspecific cannibalism is no exception (Giles *et al.*, 1986). The most abundant pike in the Czech Republic is the Northern Pike (*Esox lucius* Linaeus, 1758)) (Hecker, 2014).

1.5 Experiments Related to the Decomposition in the Aquatic Environment

Tomberlin and Adler (1998) conducted an experiment which dealt with decomposition and insect succession in cadaver of *Rattus rattus* (Linnaeus, 1758). During the summer and winter were selected two localities in northwestern South Carolina, the first in plowed field and the second was in container filled with 35 l of water. In winter, bodies exposed on land reached the phase of dry remains, but bodies exposed in water were only in phase of early floating. During the period of experiment (from 25th January 1991 to 1st September 1995) was recorded almost 30 invertebrate species was recorded, which was most likely caused by small size of experimental animal and small species diversity in chosen localities. During the winter period bodies on land was colonized with three species of flies *Cynomya cadaverina* (Robineau-Desvoidy, 1830), *Calliphora vicina*, and *Lucilia illustris* (Meigen, 1826). No invertebrates were found on bodies in freshwater environment. In summer period, bodies in both localities were colonized by *Cochliomyia macellaria* (Fabricius, 1775), *Lucilia sericata*, and flesh fly *Sarcophaga bullata* (Parker, 1916). This study is demonstrating seasonal decomposition and colonization patterns in contrasting environments and brought important findings for needs of forensic entomology.

Jirsíková *et al.* (2013) run an experiment which main purpose was to find significant organisms participating on freshwater decay, according to which will be able to settle the time elapsed from death (PSMI). Also the taphonomical changes on model organisms were mapped. The experiment started 18th May 2013.Seven rat bodies in locality with stagnant water and another seven rat bodies in location with flowing water were exposed. Sampling interval was settled on every third day due to size of the bodies of experimental animals and supposed speed of decay. During every sampling the temperature of the water source was measured. The invertebrates from the surface of the body were secured and each experimental subject was performed an autopsy. The experiment ended prematurely because of unexpected flooding in experimental area, but until this time was secured 274 representatives of family Gammaridae and 36 leeches from class Hirudinea were found in locality with stagnant water. Seven gastropods of family Lymnaeidae, 13 gastropods of family Planorbidae, two sludge worms of family Naididae and two leeches from class Hirudineawere found in locality with stagnant water. In animals exposed in stagnant water, the first visible signs of decay appeared ninth day of exposure. In animals exposed in flowing water, the first obvious signs of decay appeared after sixth day of exposition. Structure of internal organs remained intact and except of gastrointestinal tract, remarkable morphological changes did not appear in internal organs.

Main objective of Hobischak's and Anderson's (2002) experiment was to evaluate whether data on aquatic invertebrate development and succession on carrion has the potential to be used in determining time of death or submergence, as an aid in water death investigations. The research was conducted at Malcolm Knapp Research Forest in Maple Ridge in British Columbia. Experiments were conducted in four sites in still pond water and in four sites in flowing stream water. Eight pigs were killed on 31st August 1996 and prepared for the experiment (measured, weighed and partially dressed in clothes) and placed into the metal cages. Three of four carcasses per habitat were sampled for invertebrates and monitored for temperature, water chemistry, and benthic fauna. Fourth carcass in each area was used for control if weekly sampling disrupted the natural decomposition. Sampling interval was settled for every nine days for first nine weeks, than once a month for a period from December to April and after that for every two weeks until 8th September. Collected water samples were used to determine carbon dioxide content and pH. All collected invertebrates were preserved in 95 % ethanol for later identification. Insects in 10 orders, 34 families, and 46 genera were collected from the carcasses on both pond and stream control sites.

Anderson (2008) conducted an experiment focused on the decomposition of bodies in salt water, which was more focused on the decomposition activity of invertebrates and vertebrates. The experiment was conducted in the Saanich Inlet (glacially carved fjord), British Columbia, using a special underwater lab called The Victoria Experimental Network Under the Sea (VENUS) (www.venus.uvic.ca). On 5th August, a 26 kg heavy pig was killed by electricity, immediately placed in cold storage (4°C). On 7th August, carcass was removed from storage a weighted with four 2.268 kilogram (five pounds) heavy weight and dropped from the ship in the vicinity of underwater camera (Olympus C8080©). Experimental object was placed at a depth of 94 meters. Surveillance equipment monitored cadavers several times a day until the activities of aquatic organisms did not get beyond the range of the camera. For the duration of the experiment, indicators like salinity, oxygen level, clarity and temperature were monitored by VENUS equipment. The temperature of the water at the depth at which the pig laid remained very constant ranging from approximately 9.5 °C to 9.8 °C. The results showed that the body was rapidly colonized by the three spot shrimp (*Pandalus platyceros* (Brandt, 1851)), dungeness crabs (*Cancer magister* (Dana, 1852)), and squat lobster (*Munida quadrispina* (Benedict, 1902)).

1.6 The Aquatic Organisms in Forensic Practice

Byrd a Castner (2009) described the case from June 1989 when a pair of scuba divers discovered a car lying upside down on the bottom of the river, with the dead body of a woman inside in the Muskegon River in western Michigan. After the car was lifted out of the water the detectives noticed aquatic insects attached to the windshield, fenders, and door panels. Specimens were collected and sent to the Department of Entomology at Michigan State University for identification. Three different insect taxa were identified from the car: caddisfly cases, chironomid midges, and black fly pupal cocoons. The black fly cocoons provided evidence that was significant to establishing the PMSI. Cocoons of black flies are species specific and used in keys to identify specimens found in lotic habitats (Merritt *et al.*, 2008). Based on the identification of the cocoon and known life cycle of the black fly species present on the windshield of the car in late June, it was determined that the car had to have gone into the river long before June 1989 and most likely the previous fall (October/November 1988). Without this entomological evidence it would be not possible to convict a suspect from second-degree murder.

On 5th June 1997, the dead body of a partially submerged 19-year-old female was discovered in a small lake in western Michigan. She was last seen alive on the 5th or 6th June 1997. The woman's eyes and mouth were shut with duct tape, the body was weighted down with two cinder blocks and bound with chains before she was thrown into the lake alive. Due to putrid gasses body has raised to the surface. On July 7th from the head and face exposed to the air were collected insects larvae

(Calliphoridae). Samples were sent for identification at Michigan State University. Analysis showed the third larval instar the black blow fly *Phormia regina*. Based on the study of the number of days required to larvae development date of death was determined between 1st and 2nd July 1997. Another interesting aspect of this case was to recognize that the tadpoles and small spawn fed on the dead body (Byrd a Castner, 2009).

Another case described by Byrd and Castner (2009) was from Michigan. A plastic bag in the Red Cedar River north of Pardee, Michigan, was found on 13th June 2005. A bag contained rocks and a portion of the chest containing a bone, rib bones, hair, and other fragments of the human body. From the remains was collected a larvae of Muscidae, and several caddisfly larvae belonging to two families, Hydropsychidae (net spinners) and Limnephilidae (case makers). The fly larva, did not significantly assist in the investigation, but pointed to the fact that the remains were handled on land before they were sunken into the water. Due to the presence of species *Pycnopsyche guttifer* (Walker, 1852) and *Pycnopsyche lepida* (Hagen, 1861) it was possible to date the approximate timeframe for post-mortem interval from late April to late May 2005. In this particular case is very well illustrated the importance of caddis flies in certain forensic investigations.

Byrd and Castner (2009) described even a case from 27th April 1993, when the partially clothed body of the missing 16-year-old female was discovered in a large pond. Her body was floating face down in less than two meters of water. The autopsy revealed that the cause of death was strangulation. The body was already in an advanced stage of decomposition and the time spent in the water was estimated to 30 days. Large amount of the invertebrates was secured on the body and clothes of the victim. Concrete species reported by Byrd and Castner (2009) were amphipods (*Hyalella*), snails (*Physella*, *Planorbula*, *Promenetus*), clams (*Sphaerium*), water mites (*Lebertia*), dragonflies (*Gomphus*), damselflies (Coenagrionidae; *Ischnura*), chironomids (*Phaenopsectra*), pigmy backswimmers (*Pleidae*), crane flies (Tipulidae; *Erioptera*), and biting midges (Ceratopogonidae; *Bezzia*). Although secured invertebrates could not be used to accurate determination of the time of death and thus murder, based on their presence several conclusions was made. The victim was found in a pond littoral and the most of discovered taxa feed on algae, cyanobacteria, heterotrophic organisms and detritus. This implies that the body was in the water for long enough that it was settled by the above-mentioned organisms and subsequently provided nutritive substrate for colonization invertebrates. Due to found invertebrates, it was also possible to precisely determine how a dead body was moving by the wind and the waves on a pond (from deep water into the littoral).

2 Aims and hypothesis of the thesis

2.1 Aims of thesis

The aim of the study was to verify the speed of cadaver decomposition and its parameters in the aquatic environment, depending on various parameters of the environment (comparison of running versus standing water) and simultaneously establish aquatic organisms involved in the decomposition of cadavers and eventually their relation to each stages of the decay.

2.2 Hypothesis

Parameters of decomposition and composition of aquatic organisms on cadavers varies in distinct freshwater models.

3 Methods and Material

3.1 The Experiment Lužnice II

Diploma thesis was solved through a literature research and a field experiment. The experiment focused on the degradation of vertebrates, *Oryctolagus cuniculus* f. domesticus (Linnaeus, 1758) in freshwater conditions and took place in two variants – in flowing and stagnant water.

3.2 The Description of the Location

The field experiment was conducted on the Lužnice River and the former breeding pond.

Lužnice riveroriginates on the Austrian side of Novohradské Hory namely on the western slope of the mountain Aichelberg, at an altitude of 970 ma. s. l. Total length of Lužnice River is 208 kilometers and the average flow is 24.3 m^3 / s. The river flows into the river Vltava at Neznašov village (three kilometers north-west of Týn nad Vltavou). The experiment was conducted in Tábor section (App. 1.1. and 1.3.), where otherwise lowland river is gaining gradient on the order of 0.8 percent.

The breeding pond has an area of 2,000 m^2 (App. 1.2.and 1.4.). Its sides measure 40 m and 50 m. At the lowest point has a depth of 1.55 m, but the average depth is about 1 m. The only inflow is a small underground stream. The pond is no longer used for its original purpose therefore the experiment wasn´t disrupted by external factors.

For the experimental purpose14 rabbits carcasses were used each weighed about 2,500 g further a mesh netting with a diameter of 30 mm x 30 mm, weights (stones of an appropriate size) and a fishing line (App. 1.5.). The mesh enabled to fix experimental bodies to the desired station and depth but at the same time not prevent aquatic organisms from accessing them. Individual carcasses were marked with numbers 1 to 14.

In the river locality, the experimental subjects (No. 1-7) were placed in 1.5 m depth with one meter distance and tied to the boat.

In the pond locality, the experimental subjects (No. 8-14) were placed in 1.0 m depth, placed with one meter distance and secured by a wooden pin.

The experiment was launched on May 1st 2015 at 8 a.m.by exposing seven subjects into stagnant water (the pond) and then exposing another seven subjects into the flowing water (the river). The time interval between locations didn't exceed 90 minutes. May 1st 2015 represented the first day of the exposition.

3.3 The Data Collection

At the beginning of the experiment, all carcasses in both variants were simultaneously exposed. Subsequently, two cadavers were extracted at predetermined intervals, one from each variant, and evaluated.

Sampling interval was given for every seventh day due to the size of experimental animals and anticipated decomposition rate set, on 9th May, 16th May, 23rd May, 30th May, 6th June, 13th June and 20th June respectively.

After removal from water, both cadavers were placed into separate plastic containers filled with approximately 4% formalin solution and thus stored until laboratory analyses. In the laboratory, each subject was visually examined for the overall state of decomposition at first and following by its autopsy.

On all cadavers, taphonomic changes and activity of invertebrates were observed. Every organism found outside or inside the body was secured and subsequently determined. The water which remained in the sampling boxes was sieved and the invertebrates found were secured. Evaluation of decomposition such as the condition and appearance of hair, skin, internal organs, soft tissues, *etc*. was as a part of the taphonomic changes. For all organisms, their localization (on the surface of the cadaver or inside the body), species, composition, and abundance were assessed.

The water sample from the vicinity of the carcass was ensured at each sampling and analyzed in chemistry lab of Faculty of Agrobiology, Food and Natural Resources.

The water temperature at both sites was measured three times a day. Temperature data were recorded using a needling thermometer DET3R (Voltcraft®) every day during the experimental period. Times of measurement were at 7 a.m., 1 p.m., and 7 p.m.

The entire experiment including laboratory evaluation was photographically documented.

The experimental animals were collected during the winter of 2014/2015 from natural deaths from breeders within a radius of 10 kilometers from the test site. Each animal was placed in the freezer within 2 hours from death. On 31st May were all cadavers removed from freezer, placed into testing sites after free thawing.

The copies of consents from the owner of the pond and the administrator of river Vltava basin are found in the appendices (App. 5.1 and 5.2).

4 The Results

The field experiment was dealing with the decomposition of vertebrates in the aquatic environment in conditions of the Czech Republic provided data showing decomposition in two different habitats, running water vs. stagnant water. Experiment results follow decomposition of carcasses in both locations in seven-day intervals. The total duration of the experiment was 51 days.

4.1 Description of examination of the body and the results of the autopsies

1. Sampling May 9th 2015 – Day 9 of the exposition

The River (cadaver No. 1)

In comparison with the body from the pond this carcass visually showed more advanced decomposition (App. 2.1). On the stones used as weights were found representatives of families Lymnaeidae and Planorbidae, and on the surface of the body were found representatives of the family Gammaridae, the class Hirudinea, and suborder Zygoptera. The fur was contaminated during the extraction from the water by pollen, which was currently present on the river surface. A weak odor of decay was present.

The Autopsy:

Eyes were cloudy (App. 3.1.), the fur firmly held in the skin and internal organs kept their shape and texture, except of gastro-internal organs, which were massively enlarged due to presence of decomposition gases (App. 2.2.).

The Pond (cadaver No. 8)

No obvious signs of decay of the cadaver were recorded (App. 2.3.). The invertebrates were absent on the surface of the carcass or body cavities. The cadaver itself was rigid after removing from the water. A weak odor of decay was present.

The Autopsy:

The invertebrates were not found inside the body. Eyes were slightly cloudy. Skin showed no signs of decay and did not carry signs of any mechanical damage. Lungs, heart, liver, stomach, and reproductive system were in normal condition. The digestive system mainly small and large intestine were significantly enlarged by the presence of decomposition gases (App. 2.4.).

2. Sampling May 16th 2015 – Day 16 of exposition

The River (cadaver No. 2)

After removing from the river cadaver was covered with mud (App. 2.5.). On the surface of the body and stones was present a large amount of representatives of families Planorbidae, Lymnaeidae, and subclass Hirudinea. Decomposition odor was more intensive than in previous cadavers.

The Autopsy:

The body was gently rinsed before the autopsy and all water ran through the strainer to prevent loses of invertebrates. The fur could be pulled out of skin quite easily and the top layers of the skin started to tear. Eyes were blurry. Internal organs still held their physiological shape. Only changes were in gastrointestinal tract, which was extremely enlarged because of decomposition gases (App. 2.6.).

The Pond (cadaver No. 9)

Compared to the first body retrieved from the pond this cadaver was affected by the decomposition quite noticeably (App. 2.7.). The body was heavily colonized with tadpoles of families Ranidae, Bufonidae and heddles from family Tubificidae. Tadpoles fed in the eye sockets, mouth cavity and in the ears. Disruptive odor intensified.

The Autopsy:

In the coat, bald spots appeared and by using little force it can be pulled out from the skin. Skin lost its firmness and the top layers started to tear. The left eye was practically missing there was only a small part of ocular tissue left, apparently due to the activity of tadpoles (App. 3.2.). The right eye was wholly blurred the body laid at the bottom substrate of a pond on the right size, which explains the fact that this eye remained intact by the tadpoles. Gastrointestinal tract was enlarged by putrid gases and on the surface of the intestines were present a little capsules of granulated fat (App. 2.8.).

3. Sampling May 23th 2015 – Day 23 of exposition

The River (cadaver No. 3)

The body was covered with a thick layer of mud. The disruptive odor was intensive. On the surface of the body representatives of the families Gammaridae, Lymnaeidae, and Planorbidae, the class Hirudinea, suborder Zygoptera, and two waterlouses (*Asellus aquaticus* (Linnaeus, 1758)), were present (App. 2.9.). Disruptive odor was very intensive.

The Autopsy:

The skin was torn and in some places muscles were revealed (App. 3.3.). The fur released spontaneously. Tissue around the nose and lips lost its shape. Ear cartilage lost shape and was partially missing. Eyes were missing. Internal organs were soft. Digestive tract was enlarged with mass of decomposition gases (App. 2.10.).

The Pond (cadaver No. 10)

In the coat the pattern of mesh was visibly embossed (App. 2.11). The body was in an advanced stage of decomposition. The disruptive odor was intensive. The amount of tadpoles, which colonized the corpse, was much smaller.

The Autopsy:

The most significant changes of degradation were noticeable in the cranial part of the animal. Both eyes were missing, lip tissue was gone, the teeth and nasal turbinates were exposed. Ear cartilage began to meld. In the coat were bald spots, the fur itself could be easily removed from skin. In the tail a light pressure was used to remove skin with surrounding tissue to reveal the tail vertebrae. The internal organs were soft and lost contours, but they were still recognizable. The contents of decomposition gas reached its maximum (App. 2.12.). The feces in unchanged condition was present in rectum. The cecum still contained the remains of food.

4. Sampling May 30th 2015 – Day 30 of exposition

The River (cadaver No. 4)

A considerable part of the skin with the hair remained attached to the mesh (App. 2. 13.). The body was covered with mud and colonized by large amount of aquatic gastropods. Decomposition odor was intense.

The Autopsy:

The skin was torn, muscles were visible on limbs. Muscles were disappearing and tendons and bones were exposed. Head lost its shape but the large amount of its tissue was still present. The internal organs were soft, without contours, and on the digestive tract was evident, that it started to deflate (App. 2.14.).

The Pond (cadaver No. 11)

Cadaver number 10 was in advanced stage of decomposition (App. 2. 15.). Decomposition odor was extremely intense. On the body a large number of tadpoles were still present.

The Autopsy:

Animal's head completely lost its contours and the majority of soft tissues. The remaining tissues were very soft and watery. Just one third of the ear tissue remained. The tongue was extensively broken down. The mandible held only on the remains of tendons. Brain matter was mushy. Limbs were strongly affected by destructive changes. The skin and muscle tissue were missing on the soles, and phalanges were revealed. The internal organs were soft and without contours. The intestines started to deflate (App. 2.16.).

5. Sampling June 6th 2015 – Day 37 of exposition

The River (cadaver No. 5)

Compared to the previous carcasses decay advanced significantly (App. 2.17.). On the body surface was found massive amount of freshwater gastropods.

The Autopsy:

The fur was missing on 50% of body surface. The process of saponification of fats began. Head lost its shape, but in comparison with cadaver No. 12 there was still a lot of its tissue left. Hind limbs were partially skeletonized. In the abdominal cavity a large amount of fluid was present (App. 2. 18.). Liver completely lost their shape and changed color.

The Pond (cadaver No. 12)

The cadaver was significantly degraded (App. 2.19.). Large amount of tadpoles was present on the surface of the body. The entire body lost its shape.

The Autopsy:

The fat reached a stage of partial saponification. Head, anterior part of the body and limbs were left without almost any tissue. Skin was partially missing and its remains were torn. Mandible, atlas, and axis were fully exposed. Cranium had leftover tissue. Soles were partly skeletonized with missing phalanges. The intestinal tract remains slightly enlarged by putrid gases (App. 2.20.). Lungs were fully decomposed, heart was soft.

6. Sampling June 13^{th} 2015 – Day 44 of exposition

The River (cadaver No. 6)

Body was in a very advanced stage of decomposition (App. 2. 21.). The cadaver was colonized with massive amount of freshwater gastropods and leeches, one waterlouse, and 12 scuds were present.

The Autopsy:

The cracks in the skin occur all over the body and muscle tissue gleams through them. The limbs were fully skeletonized. The soles were separated from shines. In the abdominal cavity a large amount of fluid was present. Intestines were still bloated (App. 2. 22.).

The Pond (cadaver No. 13)

It was very difficult to distinguish which species it is (App. 2. 23.). From the body surface large number of healds of the family Tubificidae was secured.

The Autopsy:

Fur with skin was missing on 70 % of the body surface. Whole body flattened and deflated. It reached the stage, where it would sink if it was not loaded. Intestines flared outwards (App. 2.24.). Heart, lungs, liver, and stomach were no longer possible to recognize from each other. Front limbs and hind limbs were fully skeletonized and most of the phalanges were missing. Visceral fat was changing its structure.

7. Sampling June 20^{th} 2015 – Day 51 of exposition

The River (cadaver No. 7)

Cadaver number seven did not reach such a degree of decomposition as carcass number fourteen (App. 2. 25.). Number of secured freshwater gastropods decreased.

The Autopsy:

Skin with coat was still present on 60% of the body surface. Large mass of decomposed tissues was still present on the head. Eyes and ear were missing. Hind limbs were partially skeletonized. Phalanges on the left sole were missing. The tail was missing. Internal organs lot their original texture (App. 2. 26.).

The Pond (cadaver No.14)

Only a several representatives of families Gammaridae and Tubificidae were present. The bones of the limbs were secured only because they were captured in the mesh (App. 2. 27.).

The Autopsy:

The fur remained only on 10% of the body surface. Joint and tendon connections of limb bones were destroyed. A significant amount of bones were missing. The mandible was separated from the skull. The intestines lost their shape (App. 2. 28.). Internal organs were no longer distinguishable. Ribs punctured through the decomposed rib muscles.

4.2 Comparison of temperatures measured at both locations

Temperature was measured three times every day (at 7 a.m., 1 p.m., 7 p.m.) always on the same spot in the vicinity of cadavers.

Table 1 and Figure 1 represent the summary of temperatures measured in the river locality. Table 2 and Figure 2 represent the summary of temperatures measured in the pond locality.

No severe temperature drops or rises were measured during the experimental period in both localities. The two colder periods from 6th May to 11th May and from 26th May to 7th June occurred after rainfall.

The average temperature in the river locality was 16.4 °C.

The average temperature in the pond locality was 18.0 °C.

Table 1: Summary of temperatures measured at the river locality [°C]

The Date	7:00	13:00	19:00
1.5.	12.6	12.8	13.0
2.5.	13.0	13.3	13.5
3.5.	13.4	13.9	14.3
4.5.	14.0	14.8	14.8
5.5.	14.6	15.2	15.0
6.5.	13.0	13.1	13.0
7.5.	12.6	13.0	13.2
8.5.	13.2	13.2	13.5
9.5.	13.4	14.0	14.3
10.5.	14.3	14.8	15.2
11.5.	15.1	15.6	16.0
12.5.	15.9	16.5	16.7
13.5.	16.5	16.8	17.0
14.5.	16.9	17.0	17.1
15.5.	17.0	17.4	17.5
16.5.	17.0	17.2	17.5
17.5.	17.4	17.7	18.0
18.5.	17.9	18.2	18.5
19.5.	18.3	18.1	17.9
20.5.	17.9	17.9	17.8
21.5.	17.6	17.5	17.6
22.5.	17.5	17.4	17.3
23.5.	17.2	17.5	17.4
24.5.	17.4	17.6	17.7
25.5.	17.6	17.4	17.0
26.5.	15.0	15.1	14.9
27.5.	14.9	14.9	14.8
28.5.	14.8	14.8	14.6
29.5.	14.3	14.0	14.1
30.5.	14.0	14.3	14.3
31.5.	14.2	14.5	14.5
1.6.	14.6	14.9	15.0
2.6.	15.1	15.3	15.6
3.6.	15.5	15.6	15.8
4.6.	15.7	16.0	16.2
5.6.	16.0	16.4	16.8
6.6.	16.8	17.2	17.6
7.6.	17.4	17.6	17.8
8.6.	17.7	17.7	17.8
9.6.	17.6	17.8	18.0
10.6.	17.8	17.7	17.8
11.6.	17.3	17.4	17.5
12.6.	17.2	17.5	17.6
13.6.	17.5	17.5	17.6
14.6.	17.5	17.7	17.8
15.6.	17.9	18.2	18.4
16.6.	18.5	18.7	18.8
17.6.	18.8	19.0	19.2
18.6.	19.0	19.1	19.1
19.6.	19.1	19.5	19.7
20.6.	19.7	19.8	19.8

Table 2: Summary of temperatures measured in pond locality [°C]

The Date	7:00	13:00	19:00
1.5.	12.8	13.0	13.2
2.5.	13.3	13.5	13.9
3.5.	13.9	14.5	15.0
4.5.	14.8	15.0	15.3
5.5.	15.2	14.4	14.2
6.5.	13.1	13.3	13.7
7.5.	13.6	13.6	14.0
8.5.	14.2	14.3	14.9
9.5.	14.6	14.8	15.3
10.5.	15.1	15.6	15.8
11.5.	15.6	16.0	16.1
12.5.	16.1	16.6	16.8
13.5.	16.5	16.7	16.6
14.5.	16.5	16.9	17.2
15.5.	17.0	17.3	17.2
16.5.	17.2	17.3	17.6
17.5.	17.6	17.6	18.0
18.5.	17.8	18.0	18.2
19.5.	18.1	18.5	18.7
20.5.	18.4	18.6	18.9
21.5.	18.7	19.0	19.3
22.5.	19.1	19.5	19.6
23.5.	19.4	19.6	19.7
24.5.	19.6	19.9	20.0
25.5.	19.8	20.0	20.2
26.5.	20.0	20.4	20.5
27.5.	20.2	20.5	20.7
28.5.	20.5	20.8	21.1
29.5.	20.9	21.3	21.5
30.5.	21.2	21.0	19.9
31.5.	19.8	19.8	19.7
1.6.	19.6	19.5	19.6
2.6.	19.4	19.1	18.9
3.6.	18.2	18.0	17.9
4.6.	17.5	17.6	17.5
5.6.	17.3	17.4	17.2
6.6.	16.8	17.0	17.2
7.6.	17.1	17.5	17.6
8.6.	17.5	17.6	18.0
9.6.	17.9	18.3	18.5
10.6.	18.4	18.7	19.0
11.6.	18.9	19.2	19.5
12.6.	19.4	19.6	19.6
13.6.	19.5	19.5	19.4
14.6.	19.4	19.5	19.7
15.6.	19.6	19.7	20.0
16.6.	20.0	20.1	20.2
17.6.	20.1	20.4	20.7
18.6.	20.8	21.2	21.3
19.6.	21.5	21.7	21.9
20.6.	21.9	22.1	22.4

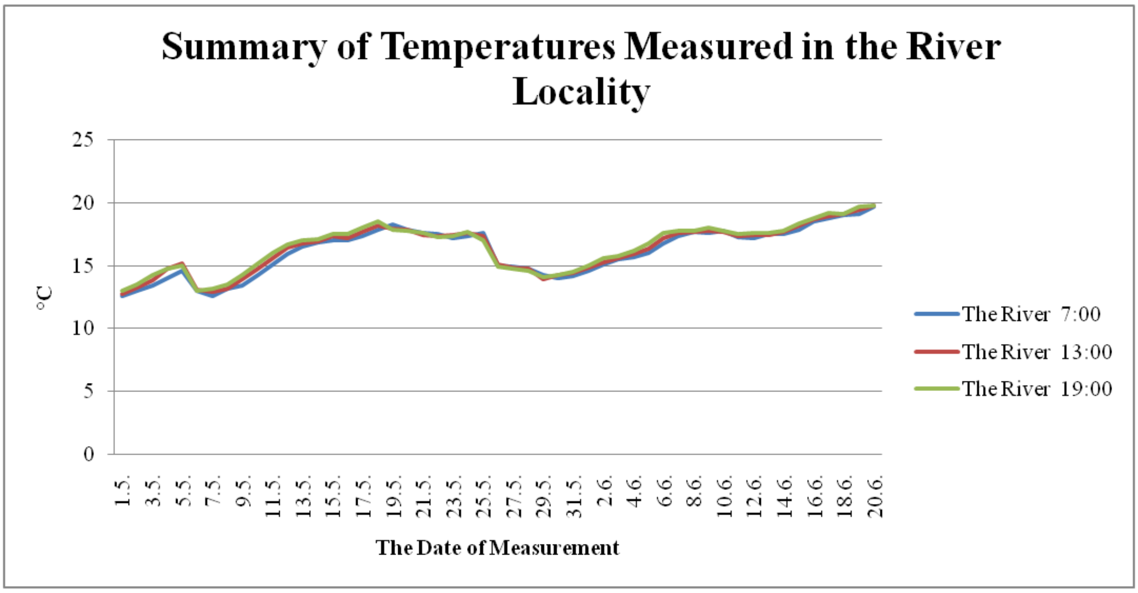


Figure 1: Summary of temperatures measured in the river locality [°C]

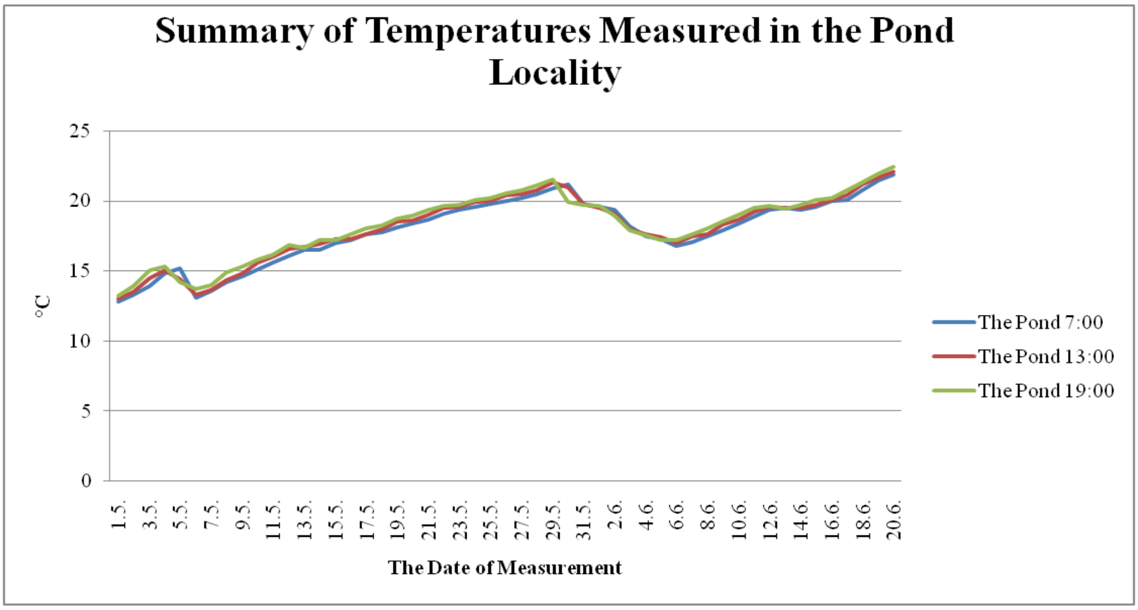


Figure 2: Summary of temperatures measured in pond locality [°C]

4.3 The results of the analysis of the water samples

The analysis of the water samples was realized in chemical laboratory of Faculty of Agrobiology, Food and Natural resources. In each water sample values of chlorides, phosphates, ammonia, nitrites, nitrates and pH were determined.

Cross-section among the water samples from the first, the third, the fourth and the seventh sampling were analyzed.

Table 3 and Figure 3 represent values of measured chemical compounds in the river locality.

Table 4 and Figure 4 represent values of measured chemical compounds in the pond locality.

Table 5 represents values of pH in both localities.

During analysis was found that the water mass in vicinity of cadavers practically did not change and the remaining samples were stored for future analysis of diatoms. The most important indicators are nitrates, nitrites, and ammonia. The decomposition processes are closely associated with the release of various forms of nitrogen. The value of pH was not significantly changed.

Table 3: Analysis of water samples from river locality [mg/l]

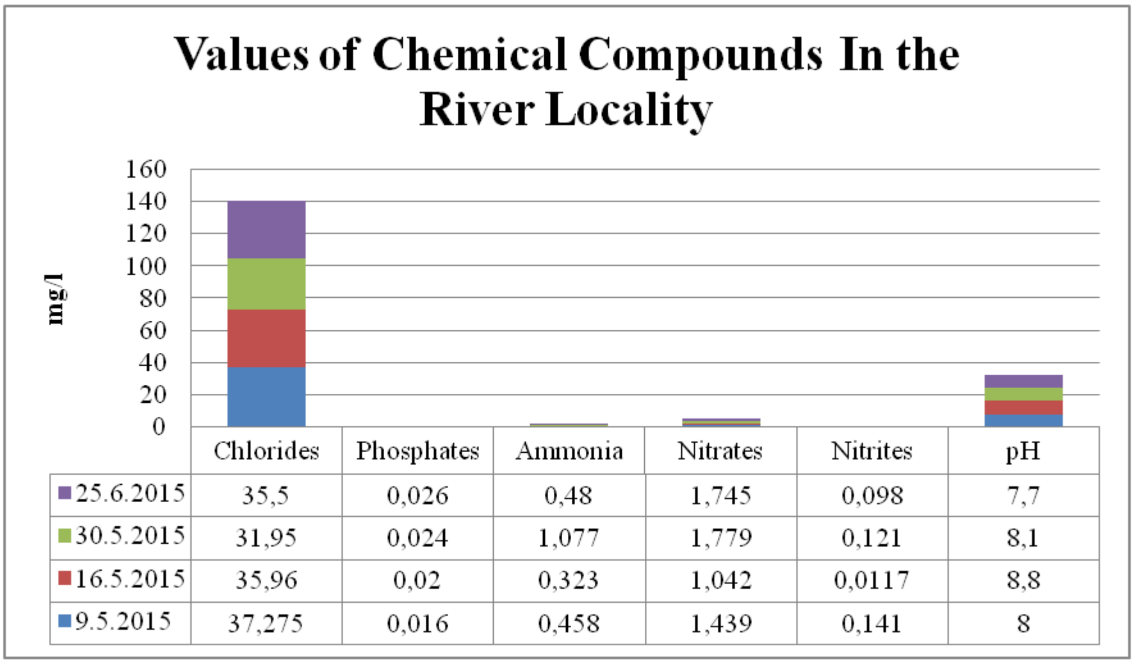
The River	**9.5.2015**	**16.5.2015**	**30.5.2015**	**23.6.2015**
Chlorides (Cl^-)	37.275	35.960	31.95	35.500
Phosphates($PO4^{3-}$)	0.016	0.020	0.024	0.026
Ammonia (NH_3)	0.458	0.323	1.077	0.480
Nitrates (NO_3^-)	1.439	1.042	1.779	1.745
Nitrites NO_2^-	0.141	0.117	0.121	0.098

Table 4: Analysis of water samples from pond locality [mg/l]

The Pond	**9. 5. 2015**	**16. 5. 2015**	**30.5.2015**	**23.6.2015**
Chlorides (Cl^-)	86.975	80.147	82.775	81.325
Phosphates($PO4^{3-}$)	0.027	0.021	0.018	0.019
Ammonia (NH_3)	0.651	0.859	0.830	0.808
Nitrates (NO_3^-)	2.700	1.8	1.2	0.535
Nitrites (NO_2^-)	0.089	0.085	0.087	0.071

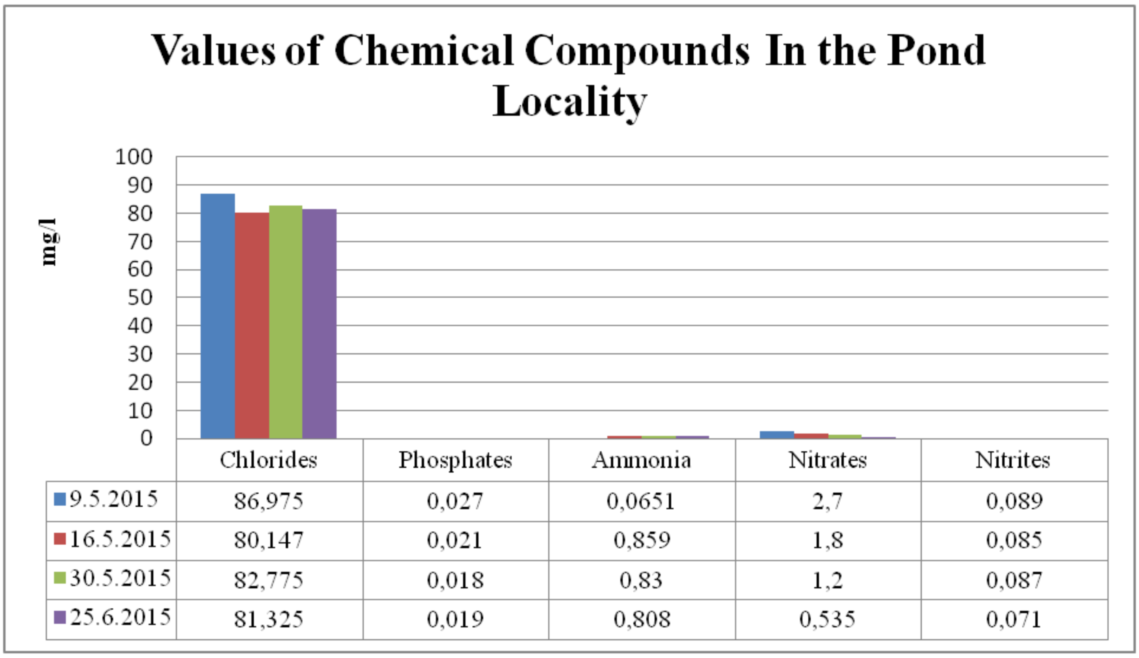
Table 5: The analysis of water samples: pH

pH – The Pond	7.1	7.1	7.7	8.0
pH – The River	8.0	8.8	8.1	7.7



	Chlorides	Phosphates	Ammonia	Nitrates	Nitrites	pH
25.6.2015	35,5	0,026	0,48	1,745	0,098	7,7
30.5.2015	31,95	0,024	1,077	1,779	0,121	8,1
16.5.2015	35,96	0,02	0,323	1,042	0,0117	8,8
9.5.2015	37,275	0,016	0,458	1,439	0,141	8

Figure 3: Measured quantities of chemical compounds in the river locality [mg/l]



	Chlorides	Phosphates	Ammonia	Nitrates	Nitrites
9.5.2015	86,975	0,027	0,0651	2,7	0,089
16.5.2015	80,147	0,021	0,859	1,8	0,085
30.5.2015	82,775	0,018	0,83	1,2	0,087
25.6.2015	81,325	0,019	0,808	0,535	0,071

Figure 4: Measured quantities of chemical compounds in the pond locality [mg/l]

4.4 **Determination of secured organisms**

The results show that the species of freshwater invertebrates vary significantly with environment.

In the river gastropods from families Lymnaeidae (App. 4.4.) and Planorbidae (App. 4.5.) were the most abundant. Significant amount of leeches (Hirudinea), (App. 4.3.), and scuds (Gammaridae) was also secured.

In the pond locality sludge worms (Tubificidae) were the most abundant. The smaller amount of scuds (Gammaridae) was also secured. Cadavers were also subjected to strong activity of tadpoles.

Table 6 and Figure 5 represent the number of invertebrates secured at each sampling in the river locality.

Table 7 and Figure 6 represent the number of invertebrates secured at each sampling in the pond locality. Figure 7 represent the number of tadpoles found on the cadavers from the pond locality.

In river locality,there were ensured: 110 *Gammarus fossarum*, 220 *Lymnaea stagnalis*, 276 *Planorbarius corneus*, 127 *Erpobdella octoculata*, 2 *Calopteryx splendens* (Harris, 1780), 3 *Asellus aquaticus* (Linnaeus, 1758) and 18 *Tubifex tubifex*.

In pond locality, there were ensured 10 *Gammarus fossarum*, 8 *Erpobdella octoculata* and 128 *Tubifex tubifex*.

In pond locality, 75 tadpoles of *Bufo bufo* and 41 tadpoles of *Rana temporaria* were secured. All tadpoles were gently removed from cadavers and immediately returned to the pond. The tadpoles were first spotted on 13^{th} May 2015 and metamorphosed into juvenile frogs from 1^{st} to 5^{th} June.

Images of all found taxons can be found in Appendices.

Table 6: The River: Invertebrates secured at each sampling [spec.]

The River	Gammaridae	Asellidae	Hirudinea	Zygoptera	Tubificidae	Lymnaeidae	Planorbidae
1. Sampling 9.5.2015	5	2	4	1	-	10	15
2. Sampling 16.5.2015	25	-	26	-	-	34	50
3. Sampling 23.5.2015	32	-	19	1	4	25	43
4. Sampling 30.5.2015	17	20	-	.	44	44	51
5. Sampling 6.6.2015	16	-	16	-	6	46	34
6. Sampling 13.6.2015	13	3	30	-	8	38	56
7. Sampling 20.6.2015	2	-	12	-	-	25	27
Total	110	3	127	2	18	220	276

Table 7: The Pond: Invertebrates secured at each sampling [spec.]

The Pond	Gammaridae	Hirudinea	Zygoptera	Tubificidae	Lymnaeidae	Planorbidae
1. Sampling 9.5.2015	1	-	-	-	-	-
2. Sampling 16.5.2015	-	3	-	19	-	-
3. Sampling 23.5.2015	3	-	-	-	-	-
4. Sampling 30.5.2015	-	-	-	-	-	-
5. Sampling 6.6.2015	1	3	-	14	-	-
6. Sampling 13.6.2015	-	-	-	36	-	-
7. Sampling 20.6.2015	5	2	-	59	-	-
Total	10	8	-	128	-	-

Table 8: The Pond: Number of tadpoles secured at each sampling [spec.]

The Pond	Number of secured tadpoles	
	Bufonidae	Ranidae
1. Sampling 9.5.2015	-	-
2. Sampling 16.5.2015	21	10
3. Sampling 23.5.2015	10	9
4. Sampling 30.5.2015	18	7
5. Sampling 6.6.2015	26	15
6. Sampling 13.6.2015	-	-
7. Sampling 20.6.2015	-	-
Total	75	41

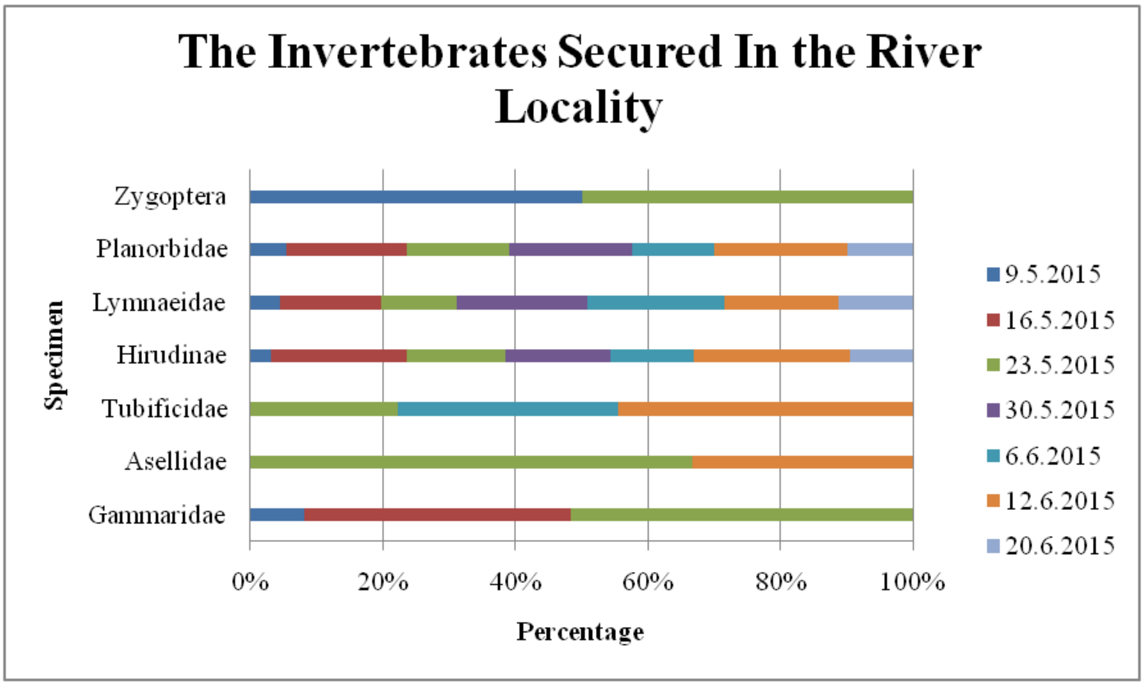


Figure 5: The River: Invertebrates secured at each sampling [%]

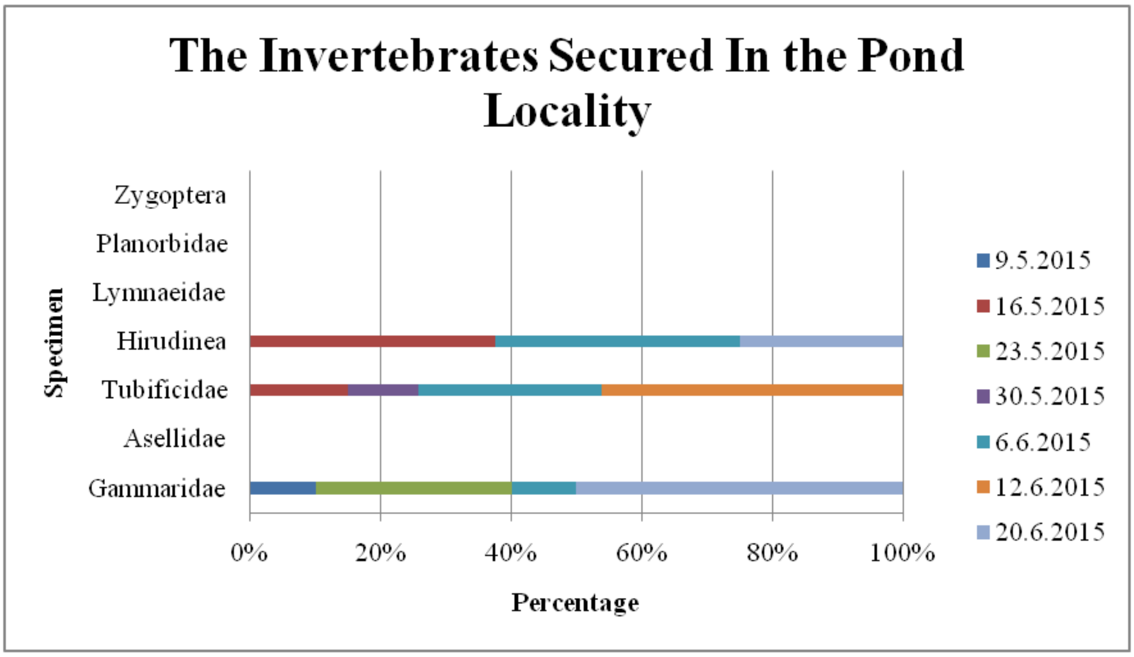


Figure 6: The Pond: Invertebrates secured at each sampling [%]

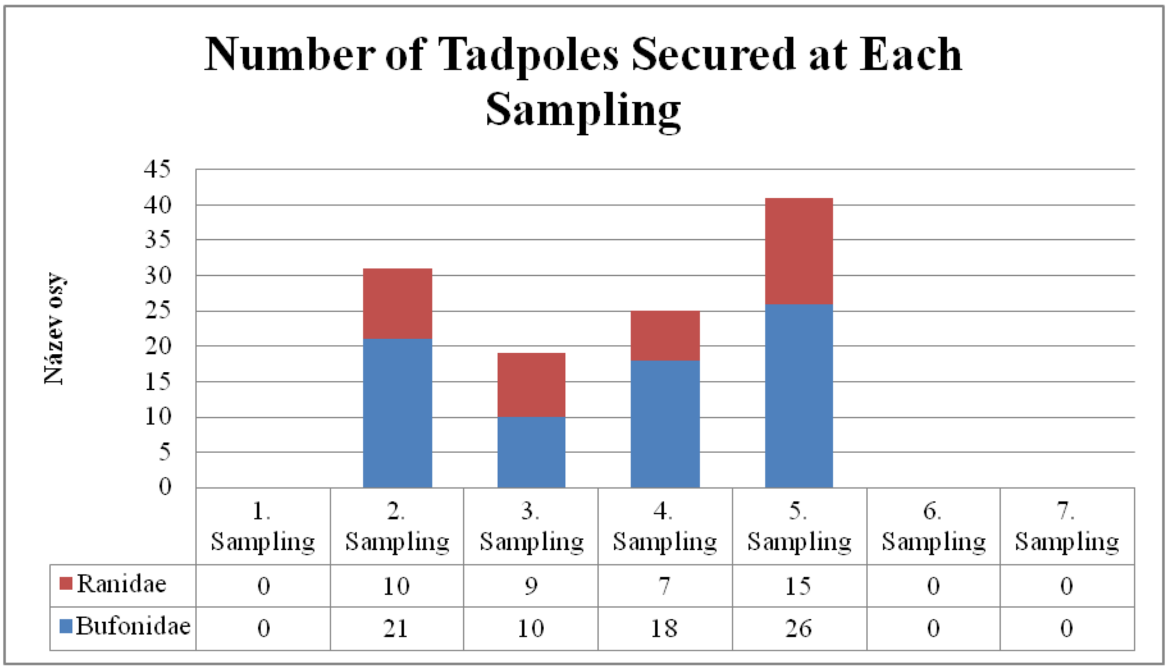


Figure 7: The Pond: Number of tadpoles secured at each sampling [spec.]

4.5 Statistical Evaluation

For the statistical evaluation, datasets describing numbers of secured invertebrates were selected. The species and their distribution in each sampling compared were by using statistical program Statistica 12 (Dell Inc., Tulsa USA). First of all a test of data normality run to see if data were normally distributed. If the results were significantly different, the value of p had to be lower than 0.05. The result was $p = 0.001377$ in dataset from river locality (Tab. 9) and $p = 0.017132$ in dataset from pond locality (Tab. 12). These results allowed to use analysis of variance ANOVA, concretely one way ANOVA to test differences in numbers of individuals among each sampling. Once the difference was significantly proved post- hoc tests were done to see particular differences in amounts. In pond locality Scheffé's test, and Fishers Least Significant Difference (LSD) test were used, in pond locality, only Scheffé's test was used.

4.5.1 The River Locality

For the statistical evaluation datasets describing numbers of secured invertebrates in the river locality were selected. The test of data normality proved that data were equally distributed and showed significant statistical difference ($p = 0.001377$) (Tab 9). Scheffé's test (Tab. 10) did not show the exact differences between the numbers of invertebrates and individual sampling. LSD test (Tab. 11) proved, there was a statistically significant difference between groups as determined by one-way ANOVA. The amount of Gammaridae was significantly different in sampling No. 5 and No. 6, the amount of secured Asellidae was significantly different in sampling No. 5 and No. 6. The amount of collected Hirudinae was significantly different only in sampling No. 6, as well as Tubificidae. The amount of Lymnaeidae differed in sampling No. 1, No. 2, and No. 7. The amount of Planorbidae was significantly different in sampling No. 1, No. 2, No. 3, No. 4 and No. 7, but at the same time the amount in samplings No. 1 and No. 2 did not differ much. Figure 8 is graphical representation of measured data.

Table 9: The Value of p

Efekt	SČ	Stupně volnosti	PČ	F	p
	Jednorozměrné testy významnosti pro Abundance (statistica) Sigma-omezená parametrizace Dekompozice efektivní hypotézy				
Abs. člen	9994,461	1	9994,461	61,94378	0,000000
Druh	4774,929	6	795,821	4,93235	0,001377
Chyba	4679,071	29	161,347		

Table 10: Scheffé's test

Scheffeho test; proměnná Abundance (statistica)
Pravděpodobnosti pro post-hoc testy
Chyba: meziskup. PČ = 161,35, sv = 29,000

Č. buňky	Druh	{1} 15,714	{2} 8,3333	{3} 15,500	{4} 17,833	{5} 31,714	{6} 39,429	{7} 1,0000
1	Gammaridae		0,993403	1,000000	0,999982	0,491336	0,093154	0,905311
2	Asellidae	0,993403		0,996771	0,978269	0,341245	0,084250	0,998644
3	Tubificidae	1,000000	0,996771		0,999987	0,658386	0,211426	0,937086
4	Hirudinea	0,999982	0,978269	0,999987		0,695065	0,195565	0,846585
5	Lymnaeidae	0,491336	0,341245	0,658386	0,695065		0,968950	0,208111
6	Planorbidae	0,093154	0,084250	0,211426	0,195565	0,968950		0,054839
7	Zygoptera	0,905311	0,998644	0,937086	0,846585	0,208111	0,054839	

Table 11: Fishers Least Significant Difference (LSD) test

LSD test; proměnná Abundance (statistica)
Pravděpodobnosti pro post-hoc testy
Chyba: meziskup. PČ = 161,35, sv = 29,000

Č. buňky	Druh	{1} 15,714	{2} 8,3333	{3} 15,500	{4} 17,833	{5} 31,714	{6} 39,429	{7} 1,0000
1	Gammaridae		0,406647	0,978712	0,766426	0,025415	0,001554	0,159239
2	Asellidae	0,406647		0,466016	0,298929	0,012375	0,001345	0,532062
3	Tubificidae	0,978712	0,466016		0,777990	0,050915	0,005424	0,197779
4	Hirudinea	0,766426	0,298929	0,777990		0,059152	0,004782	0,115399
5	Lymnaeidae	0,025415	0,012375	0,050915	0,059152		0,265182	0,005286
6	Planorbidae	0,001554	0,001345	0,005424	0,004782	0,265182		0,000738
7	Zygoptera	0,159239	0,532062	0,197779	0,115399	0,005286	0,000738	

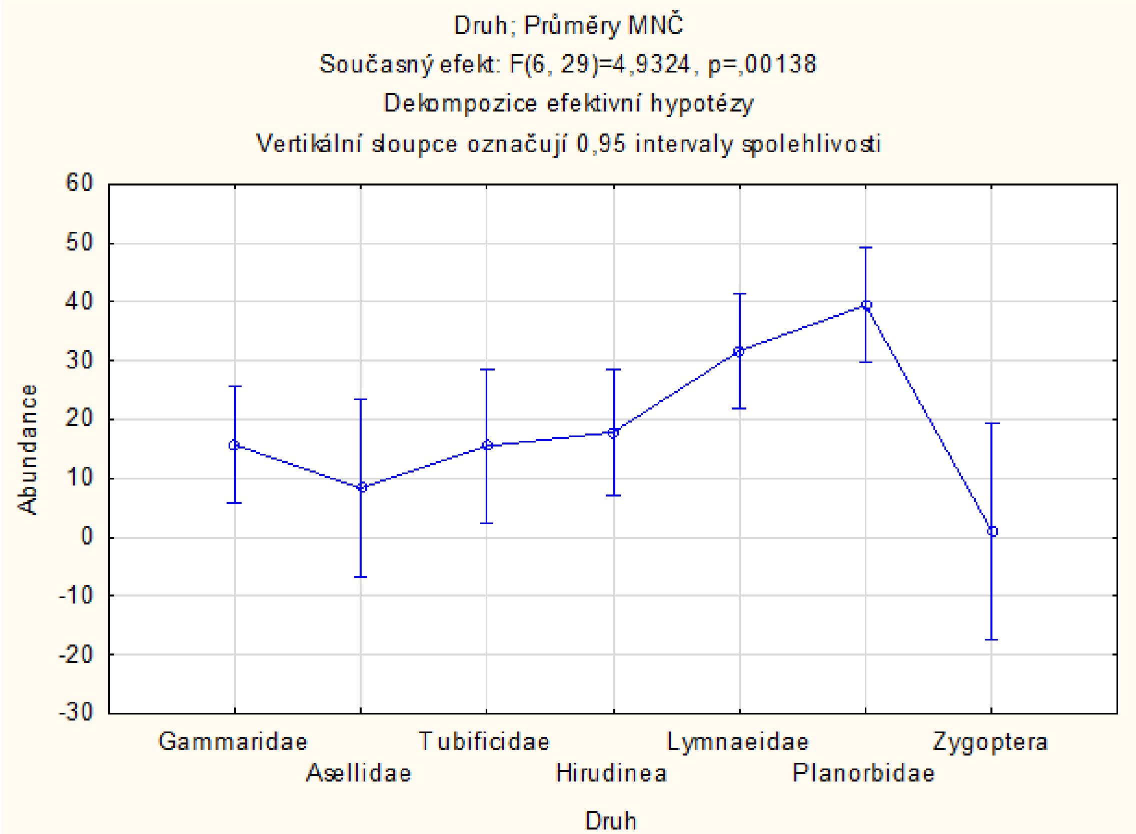


Figure 8: Graphical representation of measured data in the river locality

4.5.2 The Pond Locality

For the statistical evaluation describing numbers of secured invertebrates in the pond locality were selected datasets. The test of data normality proved that data were equally distributed and showed significant statistical difference (p = 0.017123) (Tab 12). Scheffé's test (Tab. 13) proved, there was a statistically significant difference between groups as determined by one-way ANOVA. The amount of Gammaridae was significantly different in sampling No. 2, the amount of secured Tubificidae differed in sampling No. 1 and No. 3 and the amount of Hirudinea was significantly different in sampling No. 2. Figure 9 is graphical representation of measured data.

Table 12: The value of p

Efekt	Jednorozměrné testy významnosti pro Abundance (statistica) Sigma-omezená parametrizace Dekompozice efektivní hypotézy SČ	Stupně volnosti	PČ	F	p
Abs. člen	1657,633	1	1657,633	10,61168	0,011569
Druh	2204,515	2	1102,258	7,05633	0,017132
Chyba	1249,667	8	156,208		

Table 13: Scheffé's test

Č. buňky	Scheffeho test; proměnná Abundance (statistica) Pravděpodobnosti pro post-hoc testy Chyba: meziskup. PČ = 156,21, sv = 8,0000 Druh	{1} 2,5000	{2} 32,000	{3} 2,6667
1	Gammaridae		0,030507	0,999848
2	Tubificidae	0,030507		0,044248
3	Hirudinea	0,999848	0,044248	

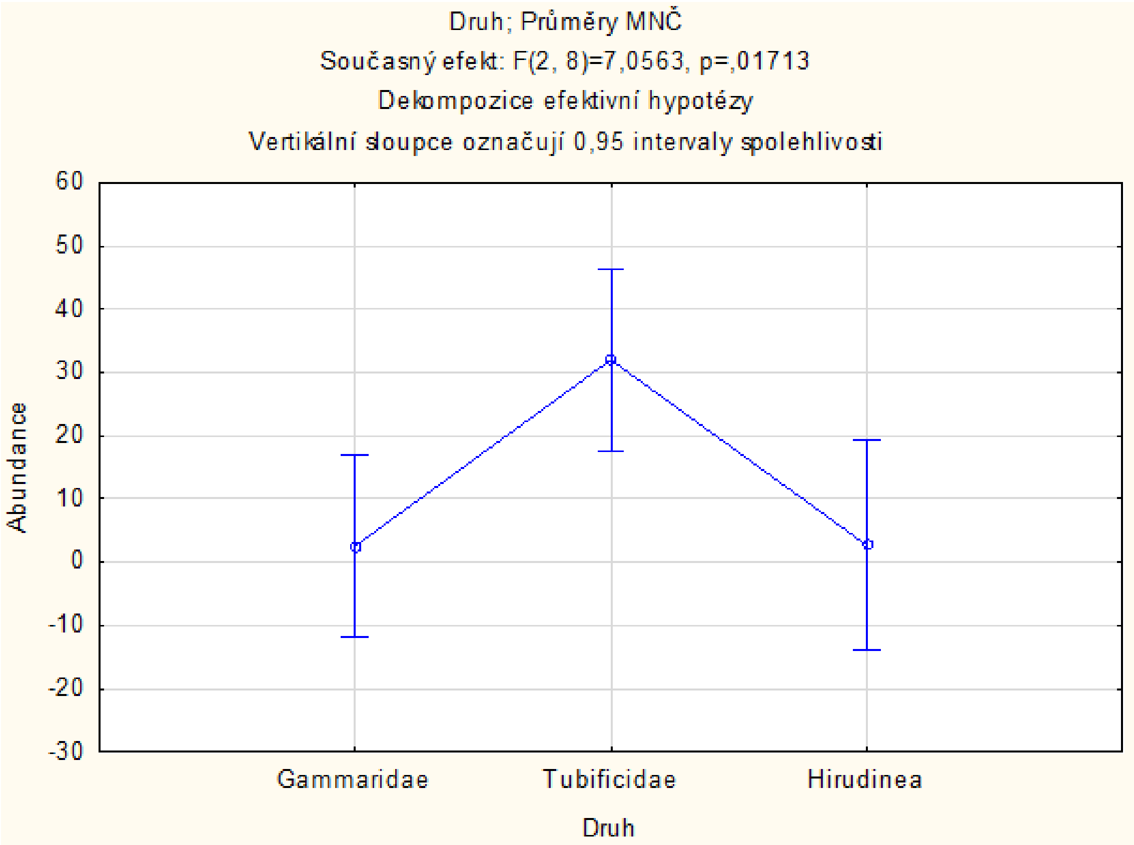


Figure 9: Graphical representation of measured data.

5 Discussion

The literature research of the diploma thesis showed that the existing knowledge about decomposition of the bodies of vertebrates in water environment are fragmented and sporadic or even absent.

The field experiment was implemented to bring new facts to problematics of freshwater decomposition of the vertebrates. The field experiment was designed for conditions of the Czech Republic and directly related to the previous similar experiment (Jirsíková *et al.*, 2013).

The presence of invertebrates was recorded on cadavers from both localities in the first sampling. However, sites significantly differed in species composition and number of individuals. Finding of abundant number of *Gammarus fossarum* in cadavers from river site agree with the statement of Anderson (2008) that from day one the dead body is colonized by various species of crustaceans (Crustacea) specific to a given aquatic habitat. Significantly smaller number of *Gammarus fossarum* in pond locality can be explained by a barren environment of the pond. Overall species composition was more diverse in flowing water. The cause of massive amount freshwater gastropods appears to be their food preference. Brusca and Brusca (1990) explain that freshwater gastropods prefer to feed on algae, which were widely present on the stones. Due to the fact, that they consume also detritus, high rates of detritus releasing from decomposing cadavers attracted them and increase amounts. Two banded demoiselles (*Calopteryx splendens*) were attracted by numerous communities of freshwater invertebrates (Heckman, 2008). Species composition in pond was compared to river significantly smaller. Crucial factor in pond locality was presence of tadpoles of families Bufonidae and Ranidae. The response of tadpoles to the presence of large amounts of organic matter was already documented by Byrd and Castner (2009). Presence of tadpoles significantly increased the speed of decomposition. Strong activity of leeches (Hirudinae) was recorded by Hobischak (1997) in her experiment with pig cadavers. The rare occurrence of *Asellus aquaticus* together with massive occurrence of *Gammarus fossarum* confirm the allegation of Anderson (2008) that in the aquatic environment the corpse is initially colonized by crustaceans.

In monitoring of speed I cannot lean towards any of the existing studies. The field experiment is the first one mapping the taphonomic changes in rabbit cadavers. Monitoring of taphonomic changes showed that decomposition was faster in the pond locality, despite the fact that first seven days, the decomposition in the pond proceeded more slowly. From the first sampling, the weak odor of decay was present in cadavers from both localities and during each sampling was more intensive. After 16 days of exposition, cadavers from both variants reached the early bloated stage. On the third sampling, the differences on cadavers became evident. The common factor for all examined cadavers was the side of the animal that lay on the bottom of water source which was significantly less affected by destructive activity of freshwater invertebrates and tadpoles. The colonization by freshwater invertebrates was not affected by a large amount of mud regularly found on the cadavers from the river locality. Cadavers in pond locality reached the stage of advanced decomposition after 23 days. The same noticeable changes occurred after 30 days of exposition in cadavers from the river locality. The process of saponification of the fat begun after 30 days in cadavers from pond locality and 37 days in the cadavers from the river locality. After 44 days of exposition, the cadavers from pond locality already became indistinguishable, front limbs and hind limbs were fully skeletonized. The most noticeable changes were evident on the head and limbs. After 51 days of decomposition, the cadaver in pond locality was affected by decomposition changes very severely. Fur was practically missing and internal organs lost its structure. Cranium, front limbs, and hind limbs were fully skeletonized. Tendons were loosened thereby the small bones of phalanges were lost. The cadaver from the river locality was less affected by decomposition changes, but even it was at a very advanced stage of decomposition. A lot of fur was still left, limbs were only partially skeletonized and cranium was still covered by large mass of decomposed tissue.

The presence of tadpoles in the pond locality played a significant role in decomposition of cadavers in the pond. From the moment when the tadpoles hatched from eggs, the decomposition was greatly accelerated. During their presence from 13th to 5th June, tadpoles consumed about 20% of body tissue, which together with the higher water temperature caused faster decomposition.

The field experiment did not prove satisfactorily the activity of fish as it were captured in results of authors Anderson (2008), Anderson and Hobischak (2004), Bozzano and Sarda‘ (2002). In river locality, the reason could be frequent presence of fishermen, which disturbed the fish. Absence of fish activity in the pond could be explained by insufficient number of carnivorous species.

The analysis of the water samples did not prove any major changes in water chemistry. In the river locality may be the reason moderate stream, which did not allow chemical compounds to accumulate. In pond locality, there were observed values slightly higher than in river, but clearly for significant influence of water chemistry was necessary larger amount of biomass. Small changes in water chemistry were caused more likely by rainfall than by decomposition of cadavers.

The temperature of both water sources was relatively stable. In the river locality, two colder periods from 6th May to 11th May and from 26th May to 7th June were recorded. The reasons were rainfall in both the experiment locality and inflows of the river. In this period, the pond locality was also influenced, but due to the smaller surface, flow rate, and the lack of inflows, the water was warmed up faster which accelerated decomposition of cadavers. From the results is obvious, that only 1.6 °C can make large difference in speed of decomposition.

Statistical evaluation shoved that most significant differences in distribution were in families Lymnaeidae and Planorbidae in the river locality (Fig. 11) and in family Tubificidae in the pond locality (Fig. 12).

Despite the amount of secured invertebrates no forensically significant organism was discovered. For further experiments I recommend use of funnel traps to catch all invertebrates attracted by cadavers and run a trial in each season.

6 Conclusion

The MSc. thesis obtained the data related to the decomposition of dead bodies by literary research and verify obtained data by field experiment. The experiment proved that the locality, species diversity and especially water temperature are absolutely crucial factors for the rate of decomposition. The field experiment provides new insights to the issue of the decomposition in freshwater environment.

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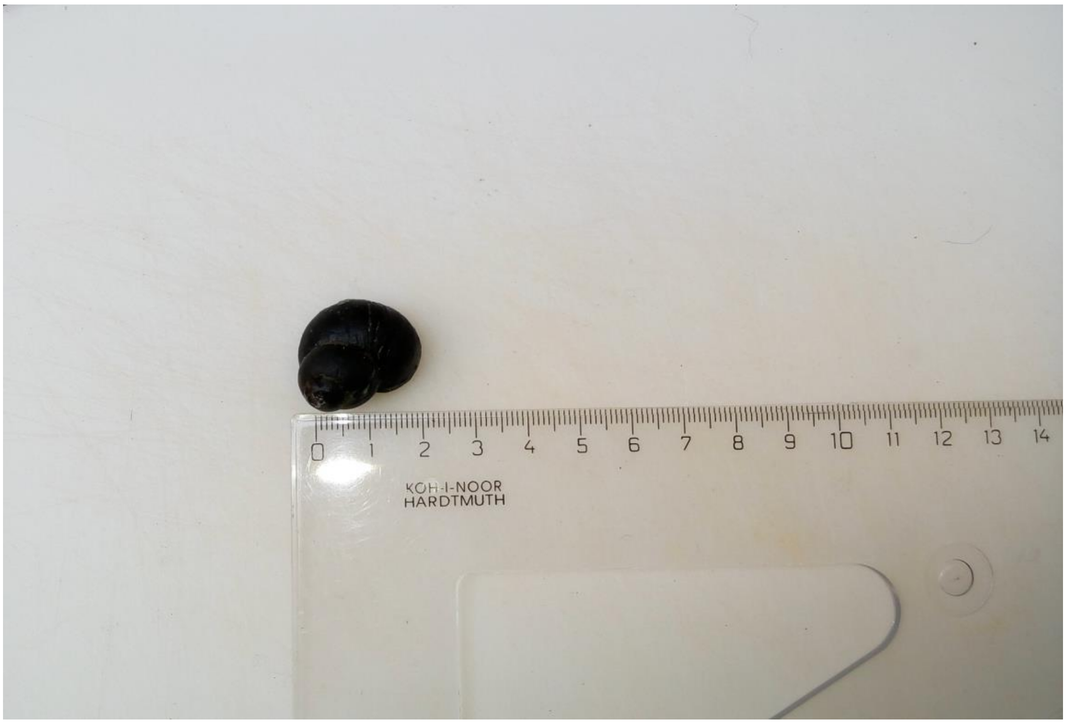

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Informovaný souhlas

Pro: Vaněk

Období realizace: 1. 5. 2015 – 20. 6. 2015

Řešitel projektu: Bc. Monika Jirsíková

Vážený pane Vaňku,

obracím se na Vás se žádostí o povolení provedení vědeckého experimentu, jehož cílem je pomocí mapování rozkladu obratlovců ve vodním prostředí stanovit signifikantní organismy podílející se na sladkovodním rozkladu obratlovců a jejich následné využití v kriminalistické praxi.

Pokud s účastí na experimentu souhlasíte, připojte podpis, kterým vyslovujete souhlas s níže uvedeným prohlášením.

Prohlášení

Prohlašuji, že povoluji konání výše uvedeného vědeckého experimentu. Řešitelka experimentu mne informovala o podstatě výzkumu a seznámila mne s cíli a metodami a postupy, které budou při výzkumu používány, podobně jako s výhodami a riziky, které z účasti na projektu vyplývají. Souhlasím s tím, že všechny získané údaje budou zpracovány, použity jen pro účely výzkumu a že výsledky výzkumu mohou publikovány.

V TÁBOŘE dne 15. 04. 2015

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Appendix 5.2. *The informed consent from the owner of the breeding pond (The copy taken on 10. 4. 2015)*

Informovaný souhlas

Pro: Povodí Vltavy, státní podnik

Holečkova 8, 150 24 Praha 5

Období realizace: 1. 5. 2015 – 20. 6. 2015

Řešitel projektu: Bc. Monika Jirsíková

Vážená paní, vážený pane,

obracím se na Vás se žádostí o povolení provedení vědeckého experimentu, jehož cílem je pomocí mapování rozkladu obratlovců ve vodním prostředí stanovit signifikantní organismy podílející se na sladkovodním rozkladu obratlovců a jejich následné využití v kriminalistické praxi.

Pokud s účastí na experimentu souhlasíte, připojte podpis, kterým vyslovujete souhlas s níže uvedeným prohlášením.

Prohlášení

Prohlašuji, že povoluji konání výše uvedeného vědeckého experimentu. Řešitelka experimentu mne informovala o podstatě výzkumu a seznámila mne s cíli a metodami a postupy, které budou při výzkumu používány, podobně jako s výhodami a riziky, které z účasti na projektu vyplývají. Souhlasím s tím, že všechny získané údaje budou zpracovány, použity jen pro účely výzkumu a že výsledky výzkumu mohou publikovány.

Ve Veselí nad Lužnicí dne 10.4.2015

Povodí Vltavy,
státní podnik
Holečkova 8
150 24 Praha 5

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