CZECH UNIVERSITY OF LIVE SCIENCES
Faculty of Environmental Sciences
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Mortality of aquatic insects during overwintering

Bachelor Thesis

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BACHELOR THESIS ASSIGNMENT

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Thesis title

Mortality of aquatic insects during overwintering

Objectives of thesis

Majority of aquatic insects use aquatic environment for overwintering. This is mainly because aquatic environment mitigating the negative effect of extremes during the winter. However, there are also species that, for various reasons, overwintering beyond the aquatic environment. overwintering period is associated with a risk of high mortality. The aim of this work is to critically evaluate the advantages and disadvantages of different overwintering strategies with regard to mortality rates and energetic costs.

Methodology

Based on the available resources (WOS, monographs on the study topic), the student will prepare a well-arranged review on the topic. The work will also include a description of methods that can be used to evaluate mortality rates during overwintering in different insect groups.

Time schedule

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January to April 2019: Completion of Bachelor Thesis.

The proposed extent of the thesis

40 – 50 pages + attachments

Keywords

Insect, mortality, overwintering, climatic conditions, life history

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Recommended information sources

Denlinger D.L., Lee R E. (2010) Low Temperature Biology of Insects. Cambridge University Press.

Harabiš F, Dolný A, Šipoš J (2012) Enigmatic adult overwintering in damselflies: coexistence as weaker intraguild competitors due to niche separation in time. Population Ecology 54: 549-556.

Leather S. R., Walters K. F. A., Bale J. S. (1995) The Ecology of Insect Overwintering. Cambridge University Press

Manger R, Dingemanse NJ (2009) Adult survival of Sympecma paedisca (Brauer) during hibernation (Zygoptera: Lestidae). Odonatologica 38: 55-59

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Declaration

I declare that I worked on my thesis on my own and that I used only the sources listed in the Bibliography and Internet sources.

In Prague on ____________________

Frolova Mariia

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Abstract

Overwintering is a difficult period for most aquatic insects and therefore is often associated with a high mortality rate. In order to survive, insects use different overwintering strategies and adaptations.

The aim of this work is to critically evaluate the advantages and disadvantages of different strategies for overwintering, with the view of mortality and energy requirements as well as a description of the methods used to monitor mortality during the overwintering period in different groups of insects.

This Bachelor thesis has been divided into 4 theoretical parts. The first theoretical chapter discusses the main limiting factors on which insect mortality depends during overwintering. The second chapter describes the types of diapause. The third chapter deals with adaptations to the winter cold and ice. In the forth and final chapter, the basics of the methods used to identify insect mortality are discussed.

Key words: Insect, mortality, overwintering, climatic conditions, life history

Abstract

Přezimování je těžké období pro většinu vodního hmyzu, a proto je často spojeno s vysokou mírou úmrtnosti. Pro přežití hmyz používá různé strategie přezimování a adaptace.

Cílem této práce je kriticky zhodnotit výhody a nevýhody různých strategií přezimování s ohledem na mortalitu a energetickou náročnost a popis metod, používaných pro sledování mortality během přezimování u různých skupin hmyzu.

Bakalářská práce na téma Mortalita vodních skupin hmyzu během přezimování je rozdělena do 4 teoretických částí. První teoretická kapitola se zabývá hlavními limitujícími faktory, na kterých závisí úmrtnost hmyzu při zimování. Druhá kapitola popisuje typy diapauzy. Třetí kapitola se věnuje adaptaci na zimní chlad a led. V závěrečné čtvrté kapitole jsou diskutovány základy metod používaných k identifikaci hmyzí úmrtnosti.

Klíčová slova: Hmyz, mortalita, přezimování, klimatické podmínky,
životní historie

Content

1. Introduction

The topic of the Bachelor thesis is "Mortality of aquatic insects during overwintering". The cold period is a difficult period for most insects. In the cold period insects experience unfavorable conditions in different ways.

Many scientists have long wondered how they endure cold and how insects overwinter. The fact is that the habitat of the majority of aquatic inhabitants such as mosquito larvae, dragonfly larvae, bugs, and other bugs, are covered with a layer of ice during negative winter temperatures. (Oswood et al., 1991; Irons et al., 1993; Frisbie and Lee, 1997).

The general aim of this paper was to improve our insight in the key factors affecting the population dynamics of aquatic insects. Without a doubt, one of the key factors is ecological strategies for overwintering. That is why in this work various survival strategies will be considered.

Speaking about the relevance of my work, it is worth mentioning the role of aquatic insect species in the ecosystem. In fact, the role of insects in nature is primarily due to the diversity in communities and their large numbers. Insects - the basis of the food of many chordates. Predatory aquatic insects regulate the number of species they feed on. In addition, insects are consumers of plant and animal remains, which is also of great importance. Accordingly, insects are a necessary natural basis. Therefore, it is necessary to study insects in various aspects of their existence. Specifically, in my work we will discuss the mortality of aquatic insect species during overwintering.

2. Limiting factors affecting the survival of insects.

According to the law of minimum of Liebig (1840), some environmental factors limit the vital activity of the organism. This means that even if all factors are in the optimum zone, and one deviates from it, this factor is the most significant. In addition, based on the law of Sheffold (1913), it can be said that not only factors in minimum can be limiting, but also maximum factors alike.

Abiotic limiting factors of aquatic living organisms:

- oxygen
- temperature

- pH
- water flow

Biotic limiting factors of aquatic living organisms:

- predation

2.1 Oxygen

Oxygen has the property of accumulating in water, penetrating there through the surface layer. In all exchange reactions, there is a need for oxygen, therefore the concentration of oxygen in water is one of the main factors influencing the limiting properties of aquatic inhabitants (Kalutsky, 2005).

Let us consider how different types of aquatic insects are getting oxygen during the winter. In the presence of photosynthetic pigments (chrolophile), energy is converted to chemical energy. In other words, any green plants emit oxygen in the presence of light. Plants growing under water is no exception.

Depending on the structure of the tracheal system, the respiration of aquatic insects can be felt. There are only two ways insects can take oxygen living under water: with the gill and with spiracles or air bags (Bey-Bienko, 1980).

An insect that has a closed tracheal system without spiracles breathes with the help of special dilated trachea and outgrowths, called gills (Kalutsky, 2005). The gills are filled with hemolymph and have a large surface area (Chown, Nicolson, 2004). In this way, species such as, for example, the larvae of *Trichoptera*, *Plecoptera*, *Ephemeroptera* and *Odonata*, can be used for gas exchange (Bey-Bienko, 1980). When turning from a larva to an adult insect (imago), reduction occurs of the gills. The closed tracheal system is replaced by an open one (Shvanvich, 1949). If the insect has an open structure of the respiratory system, then it breathes, floating to the surface, using spiracles. While the air reserves are not consumed insects are under water. With the tracheal breathing mode, air penetrates all tissues and cells of the body. Carbon dioxide is released all over the surface of the body. Capillaries form knots from forked vessels. In dragonflies of the genus *Zygoptera*, the gills are located near the oral apparatus or on the sides of the body. The gills of these species are located on the inner side of the intestine (Lampert & Sommer, 1997).

If the insect has an open structure of the respiratory system, then it breathes, floating to the surface, using spiracles. While the air reserves are not consumed insects are under water (Kalutsky, 2005).

Let us consider the breathing of swimming beetles (*Dytiscidae*) in more detail. If the temperature of the water drops, all the life processes of this insect are inhibited, including the process of respiration. The beetle has developed elytra. If the elytra are folded, when the beetle emerges, it captures the air bubble into a special cavity (sub-neutral space) (Bey-Bienko, 1980). The beetle swims up to the air bubbles under water and keeps this bubble under the sheaths. Thus, this beetle, collecting bubbles of oxygen replenishes its reserves and can be under the surface of the water during the cold weather (Kalutsky, 2005).

The larvae of the genus *Eristalis* (a large genus of hoverflies, family *Syrphidae*) have another exceptional adaptation (Stubbs & Steven, 1983). Aquatic larvae of this insect are called "rat-tailed maggot" because of their peculiarity to breathe through a three-segment breathing tube located at the back end of the body. The larvae have an open tracheal system, which means they breathe atmospheric air. The larvae can be found in oxygen-free water, such as wastewater, thanks to a tube length of about 15-20 millimeters (Shvanvich, 1949).

Another example of adaptation is *Argyroneta aquatica*. Only this type of spider spends a lifetime under water (Gordeev & Perevozkin, 1997). Only occasionally does a water spider rise to the surface of the water to replenish atmospheric oxygen reserves (Lampert & Sommer, 1997). The main feature of the breath of this organism under water is that the spider breathes with the help of air bubbles that linger between the hydrophobic hairs on the legs. The bubble is held due to the forces of surface tension of water. Because of this, the spider appears to be silver. The bubble performs the function of the physical gill. Using the trachea, the spider receives oxygen from the bladder (Gordeev & Perevozkin, 1997). When the water is immersed, the nitrogen in the gaseous state is mixed by diffusion, and the size of the bubble decreases. (Lampert & Sommer, 1997).

Unlike the water spider *Argyroneta aquatica*, the *Anisops* water bug does not use an air bubble as the main way to get oxygen. *Anisops* have an air bubble under their belly. The experiment proves that when the bug is immersed in water without oxygen, the time it takes for the insect to stay under water will not change (Miller & Golladay

1964). This proves that the bubble does not play the role of a gill. However, when the hemoglobin changes, the time spent by the insect under water changes significantly. From this we can conclude that it is hemoglobin that is the main method of obtaining oxygen for this type of bugs. The fact is that for the predation and hunting *Anisops* it is necessary that the density of the insect's body and the density of the water of the reservoir be the same (neutral buoyancy) (Matthews and Seymour, 2006). This calls the bug to swim faster. Hemoglobin with elevated oxygen content helps to achieve neutral buoyancy. On account of this, the insect can spend under water for about 5-6 minutes (Miller & Golladay,1964).

Most often, aquatic organisms are faced with a lack of oxygen. Such conditions are found in deep waters, where there is a large amount of nutrients such as plankton and flowering plants. Less common is the reverse situation. Based on the law of tolerance of Sheffold (1913), oversaturation with oxygen is also a determining limiting factor. Such conditions may occur in the groundwater outflow areas (springs) or in the groundwater itself. An excess of oxygen can be caused by an active process of photosynthesis in these waters. Water plankton cannot submerge in water due to air bubbles attached to them (Dejours, 1975).

Due to the process of interpenetration of substance molecules (diffusion) through the water surface, the gradient distribution of oxygen molecules occurs, ensuring the gas exchange process, even for small organizations (Chown & Nicolson, 2004).

There are many reasons why different organisms receive oxygen in unequal amounts. First, some species adapt to anaerobic conditions with the help of anoxybiosis (the ability to exist in water with oxygen deficiency) in case the place is not provided with sufficient oxygen. (Dejours, 1975).

Difficult to obtain the process of obtaining oxygen can stop the water flow. Organisms whose habitat is a stream get oxygen faster. However, if the organism lives in a standing reservoir, for example, as *Chironomidae*, then the insect has to make movements with the body in order for the water with oxygen scattered in it to fall into the gills (Chown & Nicolson, 2004). Thus, we can conclude that the gas exchange process in the water flow is much faster than in a standing water body.

Collembola do not have a trachea and do not have a gill. Instead of this, they are stifled by the whole surface of the subject (Lampert & Sommer, 1997).

The rate of absorption of oxygen through the surface of the body depends on the surface area of the body. The rate of gas exchange in smaller organisms will be slower than that of larger insects (Lampert & Sommer, 1997). Diffusion allows assimilation and dissimilation to be carried out.

2.2 Temperature

Water temperature is one of the key factors that directly affect the survival of aquatic creatures. Many aquatic dwellers are bioindicators of their environment. Such a factor as temperature affects many key processes occurring in water, namely: the rate of photosynthesis of aquatic plants and, as a consequence, the amount of oxygen dissolved in water, the sensitivity of inhabitants to unfavorable conditions (diseases, parasitism, water pollution) and, lastly, the rate of exchange substances. According to the law of tolerance of Sheffold (1913), if the temperature can be the determining limiting factor, not only being at the minimum, but also at the maximum. It follows from this whether the temperature is high or it is low, in both cases this will adversely affect the metabolism of insects (Bey-Bienko, 1980). Metabolism can slow down or stop altogether, since metabolism is directly dependent on water temperature. Only at a certain temperature range can a high level of metabolism be achieved. If the temperature is withdrawn from temperatures tolerated by the body, the outcome may be fatal (Cossins & Bowler, 1987).

In addition, the temperature affects the rate of egg development and the time needed to change one generation (generation time). At an acceptable temperature, all these biological periods take less time. (Chown & Nicolson, 2004)

As already mentioned, the temperature directly affects the metabolism of aquatic inhabitants and insects in particular. However, the speed and other chemical processes are also dependent on the degree of heat of the environment. For example, the work of accelerators of all chemical reactions in the body (enzymes) is constant only at temperatures tolerated by the body (Chown & Nicolson, 2004).

Take for example the larvae of the family *Diptera*. They are able to postpone freezing. This is also the representative of the *Chironomidae* family. The inhabitants of the Arctic waters have similar features. These insects have freezing points of about -19 degrees Celsius (Scholander et al., 1953).

There are species that tolerate winter by dehydration. Such species include egg fly from the *Perlodidae* family- *Arcynopteryx dichroa*, even if the temperature is -30 degrees Celsius. But only if they are lined with ice. This was proven in their work by Gehrken and Sømme (1987). In addition to dehydration and endurance to cold, there are also mechanical means of protection (Lampert & Sommer, 1997).

Consider the situation with the larvae of the family *Diptera* - Empididae. The larvae of this family spend the winter in the stream with supercooling points of about -6 to -9 degrees Celsius (Oswood et al., 1991). In contrast, the habitat during the winter *Odonata* is a non-coarse lake. The supercooling points of these species are around -5 to -7 degrees Celsius, however, when they freeze, they die (Lee & Denlinger, 1991).

Similarly, the larva of the mosquito *Wyeomyia smithii*, living in *Sarracenia purpurea*, does not freeze during the winter (Evans & Brust, 1972). With supercooling points down to -6 degrees Celsius, it is snow cover that helps this species survive (Danks, 1971).

It is known that, depending on the temperature conditions in which organisms live, they are divided into two types. There are no temperature differences in the habitats of stenotherms. In these locations, the temperature is relatively stable and constant. In contrast, eurytherms prefer places where meaningful temperature fluctuations occur. This means that even with large and abrupt temperature changes such organisms will be able to move. Aquatic creatures such as insects are no exception.

In addition, there is a division of organisms into coldstenotherms and warmstenotherms, depending on their distribution.

The temperature of the air heats up and cools depending on the presence of sunlight. Together with the air temperature, the temperature change is also observed in the aquatic environment. However, unlike the terrestrial environment, the temperature of the water changes gradually and slowly, cooling and heating only with time. From this it follows that in aquatic environments sudden temperature changes are not possible, in contrast to terrestrial environments. Thus, temperature as a limiting factor in water is not as significant as on land (Cossins & Bowler, 1987). Indirectly, temperature can influence the competition between species. The absence of a certain species in a place in an elevated temperature does not give us the right to say that the reason for this is solely temperature, for it is possible that there are other factors contributing to this phenomenon (Lampert & Sommer, 1997).

Summing up the effect of temperature on the survival of aquatic insects during overwintering, we can say that the relationship between mortality and temperature is the place to be. In his work, Donath (1981) suggested that lower temperatures during the winter have a direct effect on the mortality of insects during the winter. However, Vinogradova (2007) states in her work that the direct link between temperature and survival is often exaggerated.

2.3 pH

The influence of the degree of acidity (pH) on the mortality of aquatic creatures is not always obvious. It remains clear that, as in the case of temperature and amount of oxygen, each individual species tolerates a certain value of acidity, and therefore pH is a limiting factor. As, for example, the larvae of *Tendipedidae* live in the waters of peat bogs, where the pH is slightly acidic (about 4). But this is rather an exception, in comparison with other water assailants (Yakhontov, 1964).

Nevertheless, the problem of acid rain is the most significant, since precipitation with a low pH value changes the chemical processes in the water, and therefore directly affects the survival of aquatic organisms (Chown & Nicolson, 2004). Sulfur oxides or oxides of nitrogen are mixed with droplets of water and fall in the form of precipitation and fog on the ground, causing harm to all living things. Acid dissolves mercury, aluminum, and lead, which bark in the soil and water. Of course, the harm caused by acid rain leaves a mark on the survival rate of insects, especially mosquitoes (Yakhontov, 1964).

2.4 Water flow

There are two reasons for flowing of organisms. The first, "active drift", is intentional, for example, for laying eggs in a certain place in the stream. And the second, "passive drift", when the body accidentally breaks away from the substrate and is carried by the stream from its habitat (Bogatov, 1988). This may be dangerous especially for those insects that attached to the substrate, which protects them from low temperatures during the winter (Hauer & Stanford, 1982).

Passive drift can affect the survival of aquatic species, since the likelihood of being carried away by the stream always exists (Lancaster & Hildrew, 1993). Being carried away from the habitat, where all conditions are suitable for the organism, there is a risk not to survive in another part of the stream. River flows cannot freeze during winter, so they are a permanent and suitable habitat. Most of the organisms in the water stream are forced to move upstream in order to lay the eggs there. The task of hatched organisms is to move to the lower part of the stream and occupy the most suitable habitat for all factors, where the probability of their death will be minimal (Anholt, 1995).

2.5 Predation

Predatory nature is one of the limiting factors and often has a great influence on the change in population size. Speaking about the influence of predation on the mortality of assailants, it is impossible to mention that a high level of mortality can be associated with high energy costs and a high probability of becoming a victimized predator. For example, such predators are insectivorous birds, insectivorous mammals (*Eulipotyphla*) such as hedgehogs (*Erinaceus europaeus*), moles (*Talpidae*), reptiles and amphibians.

During the winter, the female chooses the egg-laying site based on the environmental conditions. Consider the various existing adaptations of aquatic insect species for protection from predators.

Insects of the order *Odonata* of the genus *Lestidae* hide their eggs in dense stems of coastal plants and shrubs such as *Typha*, *Scirpus* and *Carex*, which is a mechanical defense against predators (Sawchyn & Gillott, 1974).

The dragonfly squad has another feature that distinguishes them from other insects. Dragonflies are predators and can resort to cannibalism. *Odonata* larvae and adults feed on mosquitoes and midges (Belyshev, 2016). According to research, the cannibalism of the *Odonata* larvae has an effect on the change in population size. However, it must be borne in mind that cannibalism is expressed in varying degrees in different species of dragonfly. In his work, Van Buskirk (1992) noted that the frequency of cannibalism in the natural environment is changing.

Aquatic insects choose habitats on the basis of two principles: all abiotic ecological factors (light, heat, and others) are in the optimum zone, and the risk of such a biotic factor as predator is minimal in this place (Danks, 2002). So, for example, an example of choosing a habitat from predators is burrows of mucus in a substrate in which representatives of *Chironomidae* hide from attacks (Van de Bund & Groenenkijk, 1994).

In addition to this, indirect influence is exerted by people (anthropogenic factor) and large animals, which are like farm animals (biotic factor), which can trample near-growing plants in which insects can hide during winter, such as, for example, Lestidae (Vinogradova, 2007).

The larvae of *Chironomidae*, *Simuliidae* and *Culicidae* are excellent objects for studying the effects of predation in freshwater bodies (Vinogradova, 2007)

3. The types of diapause

Diapause is a period of rest, which appeared due to the adaptation of an insect to adverse conditions. It consists mainly in the inhibition of all physiological processes, including development, growth and metabolism. Depending on the length of the day and the ambient temperature, the insect may enter or leave the state of diapause. Different types of insects can move to the state of the ranges at different stages of development, whether in the state of imago, eggs or larvae. It is in the state of diapause that most insects spend the winter. Some species may fall into anabiosis at the egg stage in the hot season (Yakovlev, 1974). In contrast to the hibernation of rodents, some birds and other animals, the anabiotic status of insects is much deeper and cannot be interrupted for a short time (Kalutsky, 2005).

3.1 Depending on life stage

- Egg diapause *(Ephemeroptera, Heteroptera, Odonata, Culicidae)*
- Larval diapause *(Odonata, Ceratopogonidae, Ephemeroptera, Culicidae)*
- Imaginal diapause*(Heteroptera, Culicidae)*

3.1.1 Egg diapause

Egg diapause is one of the adaptations of insects such as some species of mosquitoes and dragonflies and some others. Typically, diapausing insects at this stage debug eggs during autumn. Having survived winter at rest in spring, eggs hatch. Diapause at this very stage of life has several advantages. They are less susceptible to drought or cold and are more easily tolerated by these unfavorable conditions than, for example, insects diapausing at the stage of imago. Organisms that winter at the egg stage are more likely to survive during the winter (Yakovlev, 1974).

Let us consider a couple of representatives hibernating in a state of eggs.

Depending on the species, *Odonata* can tolerate winter either at the larval stage or at embryonic suffering (Corbet, 1980).

Mosquitoes (Culicidae) are typical representatives of insects, most of which are winter at the embryonic stage (Kalutsky, 2005). Examples of such species are *Psorophora*, *Ochlerotatus* and *Aedes*. In addition to fetal diapase, mosquitoes often have another adaptation called aseasonal quiescence. It will be described in detail in the next chapter. It is often difficult to distinguish aseasonal quiescence from egg diapause. To identify conduct experiments for determination (Vinogradova, 2007).

3.1.2 Larval diapause

Larval diapause is another way for insects to adapt to adverse conditions, characterized by slowing the process of transition from the larva to the pupa (Vinogradova, 2007).

In the larva stage, some species of mosquitoes and dragonflies winter.

Larval diapause is characteristic of such *Culicidae* as *Orthopodomyia*, *Toxorhynchites*, *Ochlerotatus*, *Wyeomyia* (Van Damme & Dumont, 1999).

Larval diapause is characteristic of dragonflies in the Palaearctic biogeographic region. Sometimes the larvae dehydrate for a time range (Van Damme & Dumont, 1999). The development and growth is accelerated or slowed down with a change in temperature and other environmental factors (Corbet, 1980).

3.1.3 Imaginal diapause

Some insects overwinter at the stage of imago. This diapause is called imaginal. Before winter, the breeding of insects occurs, after which some females may die (Gilyarov, 1986). Before going into diapause, the adult insect begins to prepare for the cold. Their body begins to increase body fat, metabolism slows down. These adaptations increase the likelihood of their survival during the winter (Yakovlev, 1974). In addition, the activity of individuals is sharply reduced, the sex glands no longer produce the germ cells, which is why the development of the sex products stops (Gilyarov, 1986). Thus, insects hibernating in the imago stage need time to receive signals of changes that interfere with metabolism in order to accumulate energy. In the event of a sharp change in temperature, there is a high risk of death.

Mosquitoes of the genus *Culex, Anopheles* and *Culiseta* are most often diapaused in the adult stage. Also, the *Odonata* of the genus *Sympecma* overwinters at the stage of him on level areas of the earth's surface. This is exceptional because most *Odonata* has egg or embryonic diapause (Kalutsky, 2005).

3.2 Depending on the consistency

- Obligatory
- Facultative

3.2.1 Obligatory

Obligatory diapause is characteristic of insects living in places with inconstant conditions, which are often unfavorable conditions (lack of water, low temperature). Such diapause allows passage of one generation per year, that is, often with a one-year life cycle (Yakovlev, 1974).

3.2.2 Facultative

It is characteristic of insects that produce more than one generation per year. Occurs only when adverse conditions occur. If significant environmental changes have not occurred, then diapause does not occur (Yakovlev, 1974).

3.3 Depends on the choice of habitat

When the temperature drops, insects begin to look for a habitat for the time of winter. This place must match all the necessary conditions for survival, including protection against predators. The survival depends on the habitat choice of insects.

Different insects have different adaptations associated with the choice of a place for overwintering. Some people adapt to life in their summer habitats, others prefer to change places for the time of winter (Pajunen & Jansson, 1969). The former usually live in river streams or in underground waters, where the water does not freeze and does not become covered with ice, which makes these environments suitable for dwelling during the winter. However, most of the insects belong to the second group and change their summer habitat to the winter one. This is due to the fact that the places where they live in the summer freeze with the onset of low temperatures (Pajunen and Jansson, 1969). After warming, these species return to their original habitats.

An example of this strategy is some species of aquatic beetles that develop in “summer” ponds and move to “winter” ponds at the time of winter (Williams, 1997).

Species that remain in frozen habitats include insects of the family *Simuliidae*, living in reservoirs in areas with permafrost soil (Wiggins and Winchester, 1984), *Empididae* in a river flow in Alaska (Irons et al., 1993), *Odonata* in the waters of the North American steppe (Daborn, 1971), Culicidae living in trees (Copeland & Craig, 1990) and midges living in Nymphaea (Paterson, 1971).

Another example is the *Gerridae* family that winters under a layer of snow at the adult insect stage (Spence & Andersen, 1994).

One of the possible places of insect shelter during the winter is the substrate. An example would be the larvae of *Chironomidae*. When oxygen becomes insufficient, the larvae are immersed in the nutrient medium and become inactive. Substrate is a suitable place for summer growth, but can hardly be saved from exposure to low temperatures (Olsson, 1983).

Insects are divided into two types:

- Those that spend the winter on land
- Those that overwinter in the water

3.3.1 Owerwintering on land

Closer to the beginning of winter, insects begin to look for a suitable shelter. Those insects that winter on the ground can hide in man-made structures (artificial shelters) or in places conscious of nature (natural shelters). Most often in the first case it will be cellars, basements, tunnels, greenhouses, lofts of houses, bark of chests. In the second case, the insects find shelter under the bark of trees, animal burrows and dry grass (Yakovlev, 1974). Mostly during low temperatures, female mosquitoes do not feed on the blood of animals and people. However, an exception is possible if the bloodsucking winters in a heated room where cattle are kept. In this case, they can feed on blood, but without producing sweat. An example of overwintering this way can serve as an anopheles mosquito (Yakovlev, 1974).

3.3.2 Overwintering in the water.

Some aquatic insects overwinter in water. Many larvae of aquatic insects spend the winter there. The temperature of the aquatic environment changes much more slowly than the temperature on land. It is easier for individuals overwintering in the aquatic environment to recover from the cold, as they have time to adapt to new conditions (Colinet et al., 2006). A typical representative is the inhabitant of the slowly flowing water bodies of *Dytiscus marginalis*. These beetles can winter in the water or near the water on the shore in a wet land (Yakovlev, 1974). The larvae of *Dytiscus marginalis* most often crawl out onto the surface of the earth and tempered in the ground at the nearest bush.

4. Aquatic Insect Adaptations to Winter Cold and Ice

During low winter temperatures the duration of cold weather, and the ability of insects to adapt to new conditions of existence after a decrease in temperature are of great importance for insects survival (Lee & Denlinger, 1991). Insects have different adaptations for fast adaptation and survival in cold conditions. When temperatures drop, these organisms start preparing for the cold. Resistance in cold is increased due to the fact that all processes in the body slow down including metabolism, organisms lose their activity. So they are preparing for the transition to the stage of rest, called diapause. There are several ways that insects in temperate climates can survive adverse conditions and diapausee is the most common of them. Cold hardiness and insecticide resistance in general helps to survive in low temperatures and under adverse conditions (Danks, 2007).

Thus, this chapter will consider the properties of a population that are aimed at increasing the likelihood of survival, the so-called different strategies for overwintering in order to survive.

4.1 Physiological adaptation

4.1.1 Cold hardiness and habitat conditions

Habitat during the winter is of great importance. As well as on the temperature of the environment, such an important protective mechanism against cold as cold hardiness depends on the degree of freezing of the habitat. (Danks, 2005).

It is easier for individuals overwintering in the aquatic environment to recover from the cold, as they have time to adapt to new conditions (Colinet et al., 2006). The reason for this is the fact that the temperature of the aquatic environment changes much more slowly than the temperature on land. If in a humid climate the temperature drops below zero, then due to the increased specific heat capacity of water, temperature changes will be slow.

In their work, Baust and Morrissey (1976) showed, using the example of stumps, which are moisture environment, that the ability of water to react to changes in its chemical composition without jumps in pH could improve the effects of temperatures.

However, on the other hand, this ability of water and the slow melting of ice (due to buffering) provoke the appearance of recrystallizations, which appear to soften some proteins.

Consider how the choice of habitat at the time of winter affects the survival of insects. For example, insects increase the likelihood of their survival if they diapause deep in the substrate. During winter, this habitat retains heat (Danks, 1971). Choosing a non-freezing stream of water as a place for overwintering also lowers the risk of dying. Overwintering in the snow, insect have thermal insulation gets the necessary oxygen that flows through spaces between the snow (Danks, 2007).

At this point in time, an extremely small number of tests have been conducted that would show which environmental factors are decisive for the survival of insects. The available data are not sufficient for a detailed analysis of the survival of aquatic insects during overwintering. In spite of this, there is enough data on the recovery of the assailants who winter in a frozen state.

Studies related to the determination with temperature fixation showed that, for example, *Wyeomyia smithii* (called pitcher plant mosquito), inhabiting the plant *Sarracenia purpurea* does not tolerate freezing. It is the isolation from the snow in the flower that helps it survive in the winter (Farkas and Brust, 1986).

*Odonat*a living in the steppes of North America overwinter at the larva stage in ice-covered reservoirs (Daborn, 1971). In the same habitat, the frost resistance of different individuals may differ depending on conditions (Bennett et al., 2005). This can not help pushing the conclusion; adaptations of *Chironomidae* may vary depending on the conditions, which means that information about them is needed in the specification (Danks, 2007).

4.1.2 Mechanical protection against ice

In addition to chemical adaptations, such as antifreeze proteins and cryoprotectants, insects also have mechanical adaptations that protect the body from external ice. Such a mechanic adaptation may manifest itself in different ways in different insect species (Yakovlev, 1974).

Consider as an example of a mechanical adaptation the cocoons that representatives of the *Chironomidae* family make for the summer and for the winter. Summer cocoons are more freely attached, while winter ones are very close to the insect's body. If the ambient temperature drops sharply, *Chironomidae* change their summer cocoon to winter (Danks, 1971).

In addition to cocoons, mechanical holes can serve as holes in the substrate and layers of ice. For example, insects that are in the lower layers of the substrate under the layer of ice during the winter can dig deeper if the substrate freezes. This way they are protected from ice damage. This mechanical protection is most developed in insects who prefer to build their shelters at the base (Yakovlev, 1974).

Some aquatic insects dig minks in the substrate. In winter, minks serve as mechanic protection from ice damage and in summer for summer activity. Numerous studies have confirmed the fact that such adaptations are very effective and without them insects can die from various mechanical effects (Scholander et al., 1953; Danks, 1971).

All of these types of mechanical defenses, such as creating holes in the substrate and tight cocoons, are associated with insect status such as diapause, and are definitely necessary in order to reduce the mortality rate of insects during the winter (Danks, 2007). From this it follows that obtaining more information about this phenomenon is necessary to study the survival patterns of aquatic insect species during the winter. Of course, we now have very little information, due to the fact that research has been conducted on this topic and we cannot know all the possibilities for the manifestation of this adaptation. Therefore, experiments related to the study of the dependence of survival and mechanical protection are extremely needed (Danks, 2007).

4.1.3 Cold hardiness and dehydration

After conducting numerous studies on aquatic insects at the larval stage, it was concluded that the larvae that survived the adverse conditions in the sizeable bodies of water survived through three adaptations: freezing, inoculation and dehydration. The last adaptation will be discussed in this chapter.

All processes in the body, chemical or physical, take place in the aquatic environment of the body. For most living organisms, drying or dehydrating will lead to death. However, there are types of living things that can tolerate dehydration. Such organisms include bacteria, some plants and insects. For some aquatic insects hibernating on land, dehydration may mean a high risk of not surviving; for others, drying out is an essential adaptation to the cold. Due to this adaptation, insects such as *Chironomidae*, for example, are frost-proof and resistant to cold. In addition, the concentration of substances in the body remains high (Holmstrup & Zachariassen, 1996). This only means that at the same temperature level, the pressure exerted by water is lower than that of ice (Holmstrup & Westh, 1994).

It is known that in species that are not exposed to freezing in winter, dehydration occurs only if they are surrounded by ice. In other words, this step is used only when there is ice.

If ice is around, then with the help of dehydration some insects can resist the process of inoculation freezing. That is, if the process of dehydration occurs in the insect's body, inoculated freezing does not occur (Gehrken & Sømme, 1987). By inoculated metering is another adaptation, which is also important for the survival of some species of aquatic insects. Some frost-resistant insects have the ability not to freeze at zero temperatures with the help of the fats in their body. However, if such fats are not produced, the body (into the holes and walls) of the insect, surrounded by ice, is penetrated by water, which later turns into ice. This process is called inoculated freezing (Gehrken & Sømme, 1987).

In order to prevent freezing, it is necessary that environmental changes occur as gradually as possible. As already mentioned, in the aquatic environment, rapid changes in temperature are not characteristic, while the temperature actually changes much faster. Permeable cuticles that have some aquatic insects help prevent water from getting inside. Cuticles allow you to realize the balance outside and inside. For example, some large insects diapause at an adult insect stage lose moisture faster than female *Culex pipiens* during diapause (Benoit & Denlinger, 2007).

4.1.4 Cold hardiness and time

An important factor affecting the survival of insects is time. It is known that the temperature of the aquatic environment changes more slowly than the temperature on land. However, despite this, the tolerance of cold by insects is very dependent on how quickly the temperature changed in the aquatic environment, since time is needed to prepare and create suitable conditions for overwintering. The processes for which time is needed are changes in development, receiving signals about changes to inhibit metabolism, time for accumulating energy and also other processes taking place in insects as preparation for diapause (Danks, 1987). In addition, before going into a state of diapause, insects stop eating in order to clean the intestines. For this process, it is also necessary to spend a certain amount of time.

For example, representatives of *Odonata* have a high risk of death if the temperature drops sharply (Gehrken, 1989).

Another example is the *Chironomidae* family, who uses a tight-fitting cocoon as mechanical protection during the winter. In order to change the summer cocoon to a winter organism, it is necessary to spend a certain amount of time (Danks, 1991).

Thus, for any adaptation, be it mechanical or other types of preparation of insects for diapause, such as, for example, a slowdown in metabolism, are not static. Adaptation to new environmental conditions takes time (Danks, 2007). Based on this, it can be concluded that research to understand the emergence of cold hardiness and adaptations based on survival estimates is not enough. Experimenters should consider the life cycle of the body and the rate of temperature change (Danks, 1991).

4.1.5 Occurrence of low molecular weight cryoprotectants

Cryoprotectants are substances that protect living objects from the damaging effects of freezing. Low-molecular cryoprotectants penetrate into the cell. In addition to gritsirina contain sugars, proline, alanine, polyhydric alcohols, trehalose and sorbitol. At the moment we know very little about these substances. For example, *Chironomidae* freeze during winter and for these species new information on low molecular weight cryoprotectants would be extremely helpful. The survival of insects during the winter depends on these substances (Storey & Storey, 1992). After a strong hypothermia, species resistant to prevent freezing of the cells of the organism and the formation of ice in the body produce nucleic proteins. With the help of cryoprotectors, insects can survive temperatures from -25 degrees to -70 degrees Celsius (Durman, 2001).

4.1.6 Occurrence of antifreeze proteins

Antifreeze proteins (ATF) is a type of cryoprotectant. These substances are a type of proteins that are produced in the cells of the body during low temperatures. These proteins are adaptations and have an evolutionary origin. The growth of cryoprotectants occurs when the temperature drops below 0 degrees Celsius (freezing point). ATF has many important features. First, antifreeze proteins tend to prevent the growth of ice crystals, soaking into the surface of the ice crystal and disrupting their structure. In this way, they lower the freezing point. Absorption of AFP on the ice surface leads to ice recrystallization inhibition (IRI) (Knight et al., 1984). Some aquatic insects freeze even at high freezing temperatures (Frisbie & Lee, 1997).

Secondly, these proteins allow the insect freezing of arid climate to be inoculated. For example, the work written by Duman (2001), we can understand that the effectiveness of antifreeze proteins depends on the surface of the insect's cuticle. If the pores in the cuticle are small, then ATF better prevents the appearance of ice (Zachariassen & Husby, 1982).

There are two types of antifreeze proteins: antifreeze proteins (AFP) and antifreeze glycoproteins (AFGP). Both substances are important elements of low-temperature adaptation (Trunova, 2007). There is a lot of information about both of these species, which suggests that the substances are well studied.

Consider insects in which ATFs was found. These proteins can be found in both aquatic and terrestrial insect species. For some species of *Chironomidae* living in cold conditions and frozen in streams or pond substrates, smaller recrystallization proteins play a role, as they require more days to warm up and several degrees below zero to thaw (Wharton, 2003).

Antifreeze proteins have been found in insects of the *Gerridae* family at the embryo stage and in the adult stage (Gehrken & Sømme, 1987). ATF can be found in other insect species. In land insects, these protein compounds have the function of protecting the membrane and preventing nucleation.

The great advantage of frost resistance is that antifreeze proteins are able to suppress secondary recrystallization, therefore the lethal temperature is usually below -40 degrees Celsius, therefore mortality in insects with such a strategy is low (Durman, 2001).

4.1.7 Postdiapause quiescence

In some insects, diapause ends in the first months of winter, or even at the end of autumn, environmental conditions, such as temperature and the presence of ice, prevent insects from continuing their development. In such cases, there is a state of post-diapause quiescence. This condition is characterized by elevated fat content in the body, slowing down of all processes, complete inactivity and resistance to cold, and even the characteristic color of diapausing insects. development and other processes. Both of these states make insects survive these unforeseen situations, lack of food, drought, and others. In general, this condition has similar external and internal signs of diapause (Danks, 2007). Thus, post-diapause quiescence is a transitional period between diapause and development, depending on environmental conditions and arising only if these conditions are unfavorable. Post-diapause quiescence comes immediately, if the conditions are unfavorable and ceases immediately, as soon as the conditions have changed for the better and the development of the body has become possible (Danks, 2007).

For example, the reproductive diapause of the species *Chrysopa perla*, called green lace (*Chrysopidae* family) ends in the first month of winter. At about the same time, the development of the insect stops. It rises as soon as the temperature rises to the desired level (Tauber & Tauber, 1986).

Tauber and Tauber in 1976 in their work cite many examples of quiescence during post-diapause. For example, in bedbugs and insects feeding on grains, such adaptation is manifested everywhere when adverse conditions exist. After the occurrence of favorable conditions, all life processes such as development, metabolism are restored (Tauber & Tauber, 1976).

However, organisms that do not go into a state of diapause under the regular influence of such imminent factors as, for example, lowered temperatures begin to produce cryoprotectants that protect them from freezing (Asahina, 1969).

4.2 Seasonal movements

Not many examples of insect seasonal migrations are known. Most often, insect migrations are caused by wind and rarely bear the character of purposeful migrations. The insect must be large to resist the wind. In search of a favorable climate and food, some insect species can migrate. More and more studies are being conducted to track the seasonal movements of insects, which is not an easy task. However, studies show that some insects survive this winter in winter.

The complexity of such an experiment lies in the large amount of time spent on the selection of a site and the sampling of samples from sites of the same type (Danks, 2005). Also, to efficiently obtain information, it is necessary to control the movement of the insect with the help of tiny radio transmitters (Hayashi, 1994).

For example, some species of dragonflies migrate in search of moisture and never return to the places from which they flew (Yakovlev, 1974).

4.2.1 Features of aquatic species overwintering on land

Contrary to the fact that the aquatic environment is safer for overwintering insects, some insects give preference to overwintering on land. Such insects have their own characteristics that distinguish them from insects overwintering in water (Danks, 2006).

Despite the fact that in the winter, insects do not need a large amount of energy resources, in the period of autumn and spring, in the winter they should rationally use their energy. Energy consumption by insects during winter decreases, which is a criterion (along with a decrease in metabolism) for the start of the development of substances that protect insects from the damaging effects of freezing (cryoprotectors) (Danks, 2006).

Currently, information on energy consumption under different climatic conditions of aquatic species at the imago stage and the larva stage is highly demanded (Irwin & Lee, 2003).

We have enough information about winter sustainability of *Culicidae* at the adult stage (Kim et. al., 2006).

4.3 Adaptations to Aseasonal Exigencies

4.3.1 Aseasonal quiescence

When adverse conditions occur, organisms cease to reproduce and develop. This state is called aseasonal quiescence. It can even begin in the summer and occurs when insects must endure unfavorable conditions and helps conserve energy during food shortages.

In contrast to diapause, seasonal dormancy can occur at any stage of development and occurs only in the event of adverse conditions. Diapause is not always an adaptation to seasonality and can occur due to an increased population density (Chernyshev, 1996). Aseasonal quiescence, unlike quiescence, which is thermally induced and caused by drought conditions, can occur at any stage of development (imago, larval stage, egg stage) and therefore has received more attention. Aseasonal quiescence can be of two types: exogenous and endogenous. Exogenous seasonal dormancy occurs because of the influence of droughts or high or low temperatures. Seasonal repression occurs when the insect hasin adverse environmental changes and starts a physiological restructuring of the body (Chernyshev, 1996).

Aseasonal quiescence is a fairly common strategy that helps insects survive periods of stress such as drought, heat, lack of food and others. A distinctive feature of seasonal dormancy is the immediate start work of all body functions as soon as the environmental conditions are normalized and become favorable for development, reproduction and nutrition (Tauber & Tauber, 1986).

Most often, seasonal dormancy arises when the conditions of the surrounding environment are unstable. In places such as rocks, trees and groundwater, sharp fluctuations in conditions are often encountered and are often unstable habitats for insects. Due to seasonal dormancy, individuals may experience unpredictable conditions (Chernyshev, 1996).

Another useful adaptation is the uneven hatching of eggs, which can often be caused by changing environmental conditions. For example, eggs that are immersed in water may spit out more than others. Sometimes individuals of the same population can react differently to environmental signals (Yakovlev, 1974).

There are many examples of aseasonal quiescence in nature. Consider some of them. For example, such a type of *Culicidae* as *Culex nigripalpus* in normal conditions develops within six to seven days. With unforeseen changes in environmental conditions, the period of its development can be delayed for more than twenty days (Tauber & Tauber, 1986) .

Another example is the representatives of the *Culicidae* species *Anopheles* messeae, whose females leave eggs in southern Russia. However, sometimes reservoirs that are habitat for mosquitoes dry up. Mosquito females stop breeding and wait for environmental conditions to return to normal (Eichler, 1951).

As another example, we will discuss another species of the family *Culicidae*. Eggs of the species *Aedes aegypti* require contact with water for hatching and a high level of humidity. If moisture is not sufficient, then the hatching is not allowed for an unlimited amount of time until the moisture level is optimal for egg hatching (Clements, 1963).

Also, with a lack of moisture, *Hippelates* eggs cannot develop. They can fall into a state of aseasonal quiescence for up to 39 days (Spielman, 1962).

The last example I would like to mention is that of the *Chironomidae* family at the larva stage. If the reservoir dries out, the development of organisms stops and dehydration occurs. In a state of seasonal dormancy, the larvae can withstand a strong increase in temperature (they can withstand temperatures of one hundred degrees Celsius for 60 minutes), drying (they can be in this state for about nine years). A few parts after the resumption of the moisture level, the larvae continue their development (Hinton, 1960).

4.4 Evolution of overwintering strategies

Currently, there is little information about cryoprotectants, cold exertion and diorpause in general (Danks, 1971). With regard to the development of strategies for the survival of aquatic insects, at the present time is not known as cold tolerance, degradation, antifreeze proteins and other adaptations depend on various environmental factors. Such factors include the remoteness of the organism from the producers in the food chain (trophic level), winter temperature, a set of specific abiotic and biotic conditions (habitat), and the historical development of organisms (phylogenesis). However, as an example of evolution, the cold endurance of the Chironomidae midges, which developed over time, can serve as an example (Danks, 1971).

4.5 Climate change

Climate change is currently in the nature of gradual global warming. The reason for this is the influence of the human factor, and subsequently there is a change in the temperature of the Earth's climate system, reflected in an increase in the average sea level, a decrease in the amount of snow and ice. This can not fail to affect the life of all living beings. In particular, due to the increase in winter temperatures, the frost resistance of insects will be less common. Moreover, there will be changes to the supercooling points of the insects, which will become higher. Cryptoprotector profiles will become more variable (Danks, 1991). However, a change in temperature is most likely for species living on land, which will be affected by higher temperatures in the summer (Danks, 2006).

In addition, global warming affects the habitat of insects as it affects the decrease in the amount of ice, so the study of aquatic insect species and their habitat in different climatic conditions is very useful and relevant (Danks, 2007).

There are two important research approaches to conducting environmental experiments that can be used to obtain information on the survival of aquatic insect species during low temperatures.

The first approach is research carried out in order to obtain more information about the insect species. This is necessary to establish the links between the ecology of a species and its physiology (Danks, 2007). For some types of analyzes (for example biomass), the data is summarized.

The second approach is a study based on obtaining new information on various adaptations to cold, such as, for example, resistance to cold. However, one should not neglect other characteristics such as the variety of food and the life cycles of an insect (Danks, 2007).

5. Discussion.

When considering the key limiting factors of the aquatic environment affecting the survival of aquatic insect species, it was found that not only the lack of oxygen is a limiting factor, but also its excess. Techniques have been described by which insects can breathe under water, namely, with the help of a trachea, a gill, or even with the help of a tracheal tube or air bubbles (Gordeev & Perevozkin, 1997). I discussed the fact that the degree of acidity directly affects the survival of aquatic organisms (Chown & Nicolson, 2004). Cannibalism and predation also plays a big role as the mortality rate can be associated with the risk of becoming a victim of a predator and with high energy costs (Belyshev, 2016).

Different types of insects can winter at different stages of development, whether in the state of imago, eggs or larvae. Insects hibernating in the imago stage do not tolerate unfavorable conditions worse than insects in an egg state. Organisms that winter at the egg stage have a lower mortality than adult insects (Yakovlev, 1974).

Survival of insects depends on the choice of habitat at the time of winter. As well as on the ambient temperature, such an important protective mechanism against cold, as frost resistance, depends on the degree of freezing of the habitat (Danks, 2005). Winter habitat must meet the necessary criteria, be safe from the attack of predators and not covered with ice. Insects can winter in river flow, ponds, in man-made structures and in the substrate and in natural shelters. Insects hibernating in the aquatic environment, it is easier to recover from the cold, as they have time to adapt to new conditions (Colinet et al., 2006).

Overwintering in the substrate increases the likelihood of survival since this habitat retains heat well and provides mechanical protection (Danks, 1971). Cocoons also protect against mechanical damage and thereby increase the likelihood of survival (Yakovlev, 1974).

Having considered the physiological adaptations of insects we can conclude that due to such adaptation as dehydration some species of insects are resistant to cold. In addition, some frost-resistant insects do not freeze at zero temperatures with the help of fats in their bodies (Gehrken & Sømme, 1987). Such type of cryoprotectants as antifreeze proteins (ATF) protect the body from freezing, preventing the growth of ice crystals (Knight et al., 1984). A condition called aseasonal quiescence helps endure adverse conditions and help save energy when there is a shortage of food (Chernyshev, 1996).

6. Methods that can be used to study the mortality of insects.

In such a science as ecology, such a method of scientific knowledge as observation is one of the first and most important. Using this method, the observer minimally intervenes in the process or does not intervene at all. The study takes place in open nature under natural conditions. Another method of scientific knowledge is experiment. Unlike observation, in the experiment, the researcher can control the process and reproduce the conditions and even artificially change to find possible correlations between the influencing factor and the response of the organisms. Most often, the experimenter uses different devices to control conditions. This is a great advantage of the experiment (Radkevich, 1998). Especially important is the experiment in the natural sciences. In ecology, with the help of experiment, the behavior and reactions to different changes in environmental conditions of large groups of living beings such as communities, populations and communities are studied. Individuals in ecological experiments are not investigated. In addition, in ecological studies, heterogeneity of populations and seasonal changes are taken into account (Radkevich, 1998). Ecological studies can use general and private research sites. The general methods include methods by which a logical conclusion was drawn based on the transition from a particular position to a general (induction) and vice versa, a conclusion made on the basis of a general conclusion from which a private conclusion (deduction) was obtained. The method of trial plots, transect method and others are ascribed to private ones (Akhutin, 1976). Also ecology has always been under the use of the methods of other natural sciences such as biology, geography, chemistry, physics, and others. Experiments conducted in a natural environment, that is, in the open nature, the so-called field experiments are of the greatest importance for the ecologist. In addition to the natural sciences, this type of experiment is also used in other sciences. Often, natural experimenters are related to physiology. However, in such aspects as physiology consider the influence of the factor of the environmental medium on specific physiological processes and functions, while, as the ecology, the influence of surrounding factors in general on the population (Akhutin, 1976).

Ecological experiments have the main task - the study of the functioning of all ecological processes and due to what it takes place. In other words, the explanation of all the relationships of living beings in their natural habitat. For a better understanding of the ecological relations between populations, it is necessary to re-monitor the conclusions made in laboratory conditions. After conducting an environmental study, the results are analyzed and, if necessary, conduct additional research in the laboratory or in wildlife (Akhutin, 1976).

With the help of the experiment, it is possible to establish the influence on the dynamics of a population of such factors as humidity, light, temperature, predation and others (Korsunsky, 1983).

Regular experimentation allows science to evolve. In particular, environmental experiments give science a presentation about the dynamics of populations, about the influence of various factors on this dynamics, how the development of the organism changes depending on the season. The problem of active reproduction of invasive species outside their natural habitat threatens biological diversity. With the help of ecological research, we can determine how invasive species adapt to new environmental conditions. In addition, research may help predict the further development of species (Kiselev, 1998).

Experiments differ in the degree of influence of the experimenter on the process. From this degree of control experiment depends greatly on the result of the study. The studies are divided into field and laboratories. Some factors, such as the weather, cannot be controlled by the experimenter in any way, from which it can be summed up that experiments conducted in nature cannot be controlled by man. It is very important that the data obtained in laboratory and the data obtained in natural conditions do not contradict, but on the contrary complement each other. Let us consider in more detail the main characteristics of both types of research and find out the positive and negative sides (Kiselev, 1998).

6.1 Field experiment

A field experiment is a study conducted in natural conditions. Conducting such an experiment, the experimenter cannot control the process. For field experiments, it is important to simulate as accurately as possible the conditions in which the research process takes place (Akhutin, 1976).

Consider the pros and cons of field experiments. The advantage of natural experiments is that the results obtained during the experiment are extremely close to real life, since the connection between experiment and real life is extremely close. Thus, the results can be easily transferred to real life and applicable to real conditions (Radkevich, 1998).

In addition, the second significant advantage is that the behavior of the test living beings has a greater similarity with real life than when conducting research in the laboratory (Radkevich, 1998).

Moreover, this method is suitable for identifying the result of the influence of factors in the aggregate on the body (Kiselev, 1998). On the other hand, this method has certain disadvantages. First, the inability to accurately determine which of their factors most influenced the result. Under natural conditions, there are a large number of external factors that can significantly affect the result; therefore, sometimes the results of the experiment can be incorrect and sometimes even contradictory (Korsunsky, 1983). Also, the data may not be accurate due to the lack of control over the experiment, which is a big disadvantage.

Definitely, the downside is the difficulty of reproducing the experiment without detailed instructions. In addition, sometimes one or another experiment is simply impossible due to long distances or unsuitable weather conditions. In addition, large samples are necessary for natural experiments (Radkevich, 1998).

Thus, field studies are less controlled than laboratory studies, which leads to reduced accuracy and significant standard deviation. Many environmentalists give preference to field conditions. They state that laboratories are far from real natural conditions (Kiselev, 1998).

Advantages

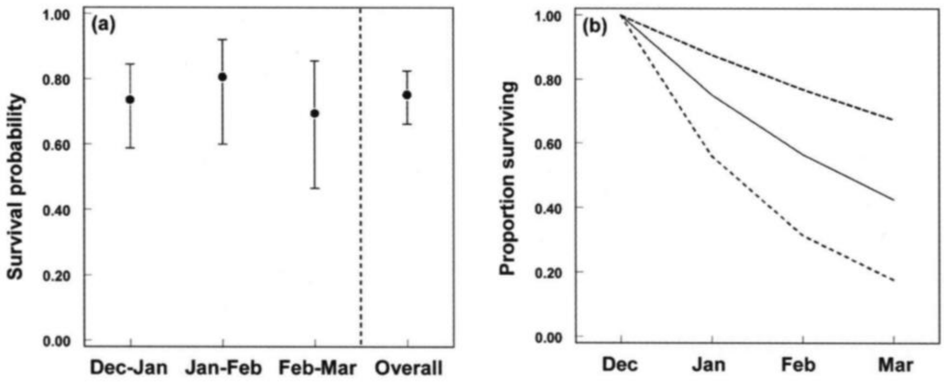
- research object implemented in everyday life, so the results can be easily transferred to real life.
- behavior in a field experiment is more likely to reflect real life because of its natural setting, i.e. higher ecological validity than a lab experiment.

Disadvantages

- inability to carry out because of the greater distances
- the complexity of the performance due to weather and seasonal factors
- unreality of control over all indicators, as well as the presence of extraneous influence from the outside world.
- difficulty of replicability (field experiments often require special access or permission, or technical detail—e.g., the instructions for precisely how to replicate a field experiment
- larger sample sizes for field testing

A field experiment is a supportive method for studying aquatic insects. Consider specific case studies that were carried out according to the method of a field experiment. For example, in their work Manger and Dingemanse (2008) "Adult survival of *Sympecma paedisca* during hibernation", during the winter they caught the dragonflies of the species *Sympecma paedisca* and marked them with tags on one of the wings. Later they visited this place to determine the survival of dragonflies of this species. The objective of the study was to determine the survival rate of the *Sympecma paedisca* in the adult stage and to determine the factors that influence the mortality rate of insects of this species during the winter. It was assumed that the survival rate will be different in different months of winter. The results of each month were compared with the model based on previous studies (White & Burnham, 1999). It turned out that the differences between were insignificant.

The first graph shows that probability of survival in December-January (0.73 ± 0.13) were higher than in February-March (d 0.69 ± 0.22), but lower than in January-February (0.80 ± 0.20) The second graph illustrates that most of the insects did not survive the winter period. The probability of survival was 0.42. Thus, most of the studied insects did not survive. Since the winter temperature was not too low, it was probably predators that was the cause of the high mortality, since the high risk of predation leads to high energy costs (Manger & Dingemanse, 2008).



Picture 1: (a) Graph showing how *Sympecma paedisca*'s survival rate has changed from December to March (± SE) (Manger & Dingemanse, 2008); (b) A graph demonstrating the survival of the Sympecma paedisca specimens noted at the beginning of the experiment during the winter (± 95% confidence intervals) (Manger & Dingemanse, 2008).

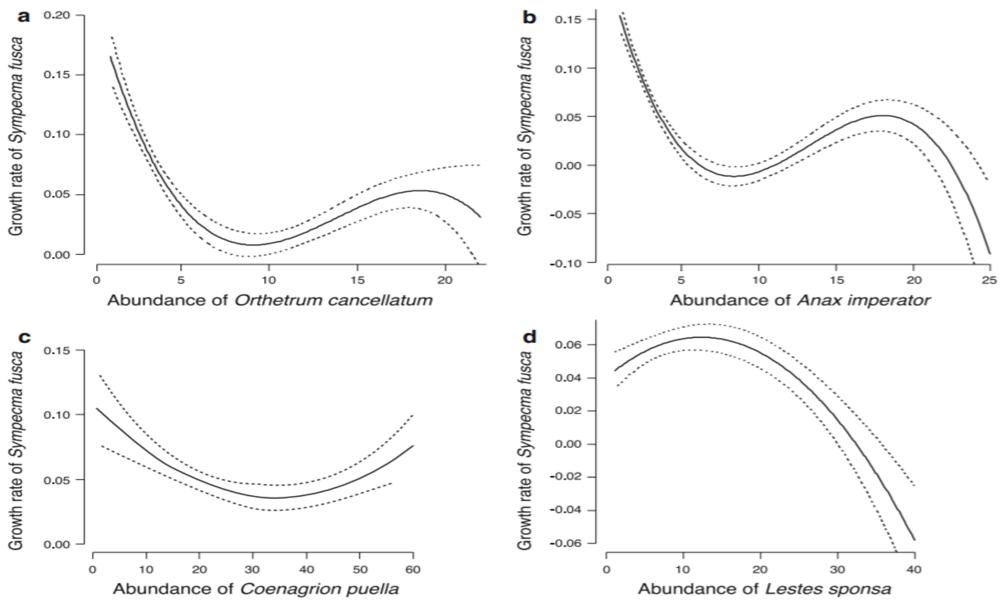
Another example of a field experiment is the work of "Enigmatic adult overwintering in damselflies: coexistenceas weaker intraguild competitors due to niche separation in time" written by Harabiš, Dolný and Šipoš (2012). With the help of a field experiment was tested hypothesis regarding the population dynamics of *Odonata* in adult life stage. Samples were captured for the purpose of comparing the population dynamics of damselflies during the winter in four common species of *Odonata*. When choosing species, an important condition was that the species had different overwintering strategies.

It turned out that there were significant differences in populations of different species between periods. When comparing the results with the model (LME), which is based on a 5-year sample, it turned out that there is a relationship between the growth of the *Sympecma fusca* dragonfly population and the estimated number of individual species. The table below shows the effect of the estimated number of four *Odonata* adults on the relative growth of the *Sympecma fusca* population using linear mixed models (LME). It is also shown in the table that during the seasonal maximums the estimated number of damselfly larvae during the season is the highest (Harabiš et al., 2012).

	df	*F*	*P*
Intercept	1	3.97	0.048
Orthetrum	1	23.60	<0.001
Intercept	1	1.30	0.257
bs (*Lestes*)	3	7.59	<0.001
Intercept	1	9.44	0.003
bs (*Coenagrion*)	3	6.72	<0.001
Intercept	1	3.81	0.053
bs (*Anax*)	3	13.57	<0.001

Picture 2: The table shows the effect of estimated abundance of four species of adult *Odonata* on relative population growth of *Sympecma fusca*. Linear mixed models (LME) were used.(Harabiš et al., 2012).

In conclusion, based on the models, it can be said that there is a negative non-linear relationship between the estimated number of individuals and the relative growth of the *Sympecma fusca* population, which is evident in the graphs below (Harabiš et al., 2012).



Picture 3. Relationship between the relative population growth of *Sympecma fusca* and the abundance (estimated number of individuals) of individual species: (a) *Orthetrum cancellatum*; (b) *Anax imperator*; (c) *Coenagrion puella*; (d) *Lestes sponsa*. Dashed lines indicate SE. The scales of x- axes and y-axes differ among panels (Harabiš et al., 2012).

6.2 Laboratory experiment

A type of experiment in which conditions for research are created artificially, within the framework of a scientific laboratory. The factors that ensure the interaction between the subjects under study are selected solely from the interests of the experimenter. The factors of interest are called incentives, and the subjects in question are a group of subjects. For the study of living objects used extreme exposure to environmental factors. The experimenter can change the concentration and composition of substances, the degree of toxicity of substances (chemical factors), the amount of light, humidity, pressure, radioactivity (physical factor), interaction with other organisms, manipulation with genes, cells, organs (biological factor) (Kiselev V.N., 1998). To change the state of the object of research, the researcher purposely acts on the object of research.

Like the field experiments, the laboratory has its pros and cons. The big advantage is that a high degree of control is provided over the experiment in the laboratory environment. The artificially created environment is carefully monitored and isolated from any unnecessary impacts. On this basis, the impact of environmental factors may be minimal. Such experiments also have a high degree of internal reliability, due to the fact that studies with the same objects often show the same result. It requires fewer samples, does not require the participation of a large number of studied organisms. Experimenters are not limited to weather conditions, time, geographical location and season. On this basis, in vitro experiments are less expensive, in contrast to in vivo experiments (Kiselev, 1998).

The main objective of a specially organized experiment in vitro is to ensure high internal reliability. However, the external accuracy of the field experiment is higher than that of the laboratory. Usually, several dependent variables and one independent variable are selected and external variables are controlled by the elimination method, in cases where it is necessary to ensure maximum control over dependent and independent variables. Additional random stimuli are called irrelevant, which under natural conditions are much more difficult to control (Kiselev, 1998).

Experimental observation is characterized by the fact that the conditions in which organisms are placed can accurately dose and evaluate its influence of any single factor, unlike observation in nature (Radkevich, 1998).

In a laboratory experiment, it is possible to control the entire process to any degree. Full control over all environmental factors is possible only with complex special installations (Radkevich, 1998).

Sometimes subjects do not respond to changes in independent factors, but to the environment. It is often provoked by an artificial environment. Field experiments have a higher authority in environmental experiments. In laboratory experiments, the problem of propagation in real situations may arise, because this type of research is carried out in an artificially created environment (Korsunsky, 1983).

Advantages

- strict control over all indicators, as well as the use of specialized equipment for measurements

Disadvantages

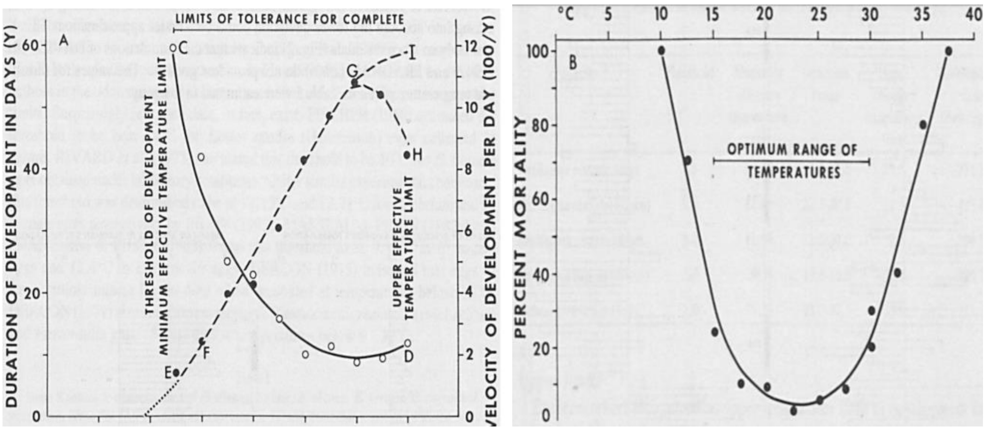
- the complexity of the reproduction of the results obtained in real life situations
- in artificial laboratory conditions it is practically impossible to simulate real life circumstances, only some of their fragments.
- errors in creating an experimental situation;
- difficulty of reproducing the results in a real life situation.
- the influence of the experimenter;
- isolation from the real situation.
- there is less control over extraneous variables that might bias the results. This makes it difficult for another researcher to replicate the study in exactly the same way.

An example of work carried out in the laboratory is the work of Pilon and Masseai (1984) entitled "The effect of temperature: revealed the effect of temperature on egg development in *Zygoptera*". This article outlines how much temperature affects the development of the *Zygoptera* embryo using two mathematical models. With the help of models, the relationship between the rate of development and temperature was established. Environmental strategies of the species studied were taken into account. The minimum and maximum temperature limits for embryo development were identified.

During the experiment, it became clear that there are lower and upper limits that limit the development of the embryo. They are shown on the diagram below under the designation 4A. The graph shows that if the temperature rises then the time spent on the development of the embryo decreases. This is verified by points C and D on chart

4A. Under the points E-H shows the speed of development. With an increase in temperature, the speed reaches the highest point (graph 4A, G). However, later a decrease in the efficiency of temperature (G-H) occurs (Pilon & Masseai,1984).

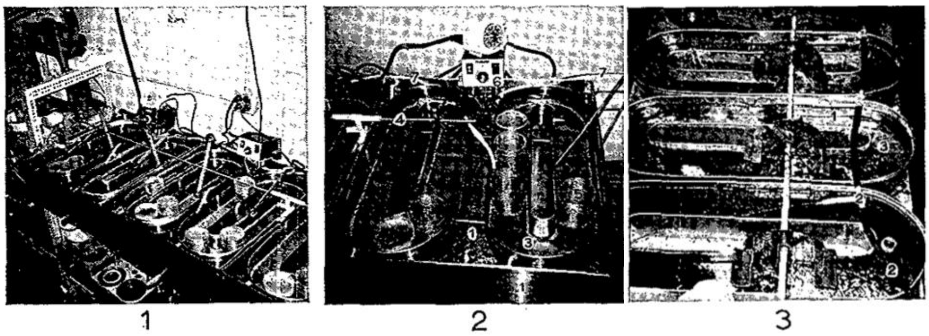
Graph 4B shows the dependence of the percentage of embryos that have developed over a short period of time from temperature (Peairs, 1927; Uvarov, 1931). It turned out that if the temperature exceeds the optimum range, the embryo development begins to be inhibited. Thus, too high temperatures adversely affect the development of eggs (Pilon & Masseai, 1984).



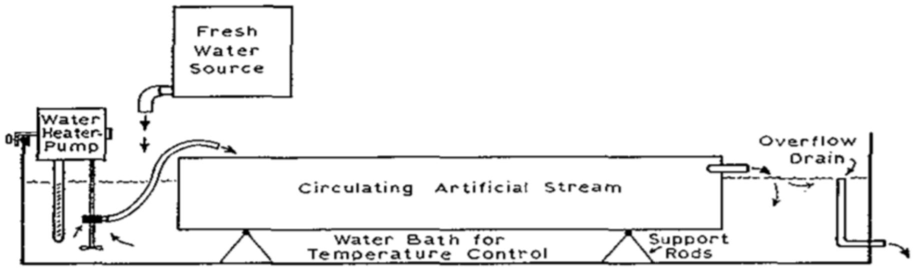
Picture 4: (A)Theoretical effect of temperature on the duration and velocity of development. (B) The dependence of the percentage of embryos that have developed over a short period of time from temperature (Pilon & Masseai,1984).

Another example of the use of a laboratory experiment to study aquatic insect species is the work written in 1968 by Nebeker and Lemke under the title "Preliminary studies on the tolerance of aquatic insects to heated waters". In this work, experimenters tested twelve species of aquatic insects to determine their deadly threshold when the water temperature rises. The research was carried out in laboratory. The lethal temperature was understood as the temperature at which the test insects of the test subjects died after 96 hours.

The laboratory experiment was carried out in special oval-shaped containers. They were immersed in a water bath, which is depicted in the illustrations under the numbers 1 and 2. With the help of a pump, which is shown in the illustration under the number 4 and using special blades (image number 3), the river flow was simulated (Nebeker & Lemke, 1968).



Picture 5: (1),(2) Special oval containers immersed in a water bath. Test Chembers. (3) Special blades with the help of which the river flow was modeled. Holding-Acclimating Chambers (Nebeker & Lemke, 1968).



Picture 6: The diagram shows a special pump with which the river flow was modeled. Diagram of test chamber shown in Picture5 (2) (Nebeker & Lemke, 1968).

The table below presents the results obtained after testing species for elevation to elevated water temperatures (Nebeker & Lemke, 1968).

In the course of the experiment, it became clear that such species as, for example, *Taeniopteryx maura*, *Ephemerella* subvaria and *Isogeiiiis frontalis*, are the most sensitive to temperature increases compared with other studied species. This can be explained by the fact that in nature the development and activity of these species does not begin at the moment when the temperature drops out. In contrast, *Ophiogomphus rifpinsulensis* and *Boyeria vinosa* (order *Odonata*) turned out to be the most stable in high temperatures. These types of dragonflies live in slow warm streams, therefore they are able to endure various extreme conditions (Nebeker & Lemke, 1968).

SPECIES TESTED	TL_m^{96} (°Celcius)
Taeniopteryx maura (winter stonefly)	21 °
Ephemerella subvaria (mayfly)	21.5°
Isogenus frontalis (stonefly)	22.5°
Allocapnia granulata (winter stonefly)	23 °
Stenonema tripunctatum (mayfly)	25.5°
Brachycentrus americanus (caddisfly)	29 °
Pteronarcys dorsata (stonefly)	29.5°
Acroneuria lycorias (stonefly)	30 °
Paragnetina media (stonefly)	30.5°
Atherix variegata (true fly)	32 °
Boyeria vinosa (dragonfly)	32.5°
Ophiogomphus rupinsulensis (dragonfly)	33 °

Picture 7: The results obtained after testing them for elevation to elevated water temperatures. Temperatures at which 50% of the test species died after 96 posure (TLm68) when acclimated at 10°C. for one week (Nebeker & Lemke, 1968).

7. Conclusion

The aim of my bachelor thesis was to explore the key factors affecting the population dynamics of aquatic insects, critically evaluate the advantages and disadvantages of different strategies for overwintering, with the view of mortality and energy requirements and describe methods that can be used in monitoring the mortality during overwintering in different groups of insects.

I focused primarily on limiting factors of the aquatic environment in general (such as oxygen, temperature, pH, water flow and predators) which directly affect the survival of aquatic organisms.

Further I described the types of diapause in which insects survive the winter. It was concluded that diapause at the egg stage has more advantages than, for example, diapause at the imago stage. Insects overwintering in the egg stage are less susceptible to drought or cold and are more easily tolerated by these unfavorable conditions and therefore survive with greater likelihood during the winter.

I tried to come close to the problems of lack of oxygen in the environment (especially during winter) and to the problem of acidification of the water pollution of the atmosphere (acid rain), which has become very serious and led to acid stress on aquatic organisms.

Besides, I also described physiological adaptations of insects to winter cold, such as antifreeze proteins, postdiapause quiescence and others. It can be concluded that the strategy of cold hardiness has significant advantages. Obviously, this strategy allows insects to control freezing, protects against mechanical damage, and saves energy. Cold hardiness is a stable state of natural cryopreservation in winter and ensures insect resistance to ice and snow. However, cold hardiness is very dependent on the rate of change of temperature. The reason for this is that insects need time for changes in the body. Therefore, there is a high risk of insect death in case the temperature change has occurred quickly.

At present, we know very little about cryoprotectants, aspects of cold endurance, and winter cocoons. Only after more information about all aspects of physiological adaptations is known, can we set more complex tasks.

Besides, it would be useful to find out more information about the species that are supercooled in the presence of ice and for the species and that survive during freezing. At the moment we do not know to what extent the lifestyle, trophic level, habitat, temperature during winter, conditions during spring, phylogenesis and other factors have an effect on winter movements and endurance to cold.

At the end of the work I studied and described methods, that can be used to study the mortality of insects, advantages and disadvantages of the two main types of experiments. It was concluded that although field research is less controlled and accurate than laboratory research. However, many environmentalists prefer field conditions as laboratory experiments are more distant from real natural conditions than field ones.

Thus, in the further work, I would like to delve further into the impact of climatic conditions on the mortality of aquatic insects of some species through a field experiment. The experiment will enter 40 individuals, who will be held in insectarium. Individuals will be individually tagged and monitored during the entire period of overwintering. During the entire experiment will also be measured microclimatic conditions (temperature and insolation) using datalogger.

8. References

Akhutin A.V., 1976: History of the principles of physical experiment. From Antiquity to the XVII century. Moscow, 292 p.

Anholt B.R., 1995: Density Dependence Resolves the Stream Drift Paradox. Ecological Society of America, October, 2235-2239 p.

Asahina E., 1969: Frost resistance in insects, Advances in Insect Physiology. 6:1–49.

Baust J.G., Morrissey, R. E., 1976: The ontogeny of cold tolerance in the gall fly, Eurosta solidagenis (sic) J. Insect Physiol. 22:431–437.

Belyshev F., 2016: Fauna and ecology of dragonflies. Under total. ed. Mordkovich V. G. - Novosibirsk: Science.

Bennett,V.A., Sformo T., Walters K., Toien O., Jeannet K., Hochstrasser R., Pan Q., Serianni A.S., Barnes B.M. and Duman J.G., 2005: Comparative overwintering physiology of Alaska and Indiana populations of the beetle Cucujus clavipes (Fabricius): roles of antifreeze proteins, polyols, dehydration and diapause. Journal of Experimental Biology, 208, 4467–4477.

Benoit J.B., Denlinger D.L., 2007: Suppression of water loss during adult diapause in the northern house mosquito, Culex pipiens.Journal of Experimental Biology 210, 217–226.

Bey-Bienko G.Y., 1980: General entomology. 3rd edition., "High School", Moscow, 416 p.

Bogatov V.V., 1979: Methods for determining the drift distance of benthic invertebrates (using the example of the Bureya river). Ecology. № 4. S. 82-88.

Buskirk M.J.,1992: Competition, cannibalism, and size class dominance in a dragonfly. Oikos 65: 455-464.

Chernyshev V. B., 1996: Ecology of insects. Publishing House of Moscow State University, 304 p.

Chown S. L., Nicolson S. W., 2004: Insect physiological ecology: mechanisms and patterns. Oxford University Press, Oxford

Clements A. N., 1963: The Physiology of Mosquitoes. Pergamon Press, Oxford, New York.

Colinet H., Renault D., Hance T., Vernon, P., 2006: The impact of fluctuating thermal regimes on the survival of a cold-exposed parasitic wasp, Aphidius colemani.Physiological Entomology 31, 234–240.

Copeland R. S., Craig G. B., 1990: Habitat segregation among treehole mosquitoes (Diptera: Culicidae) in the Great Lakdes region of the United States. Annals of the Entomological Society of America 83, 1063-1073.

Corbet P.S., 1980: Biology of Odonata. Annual Review of Entomology 25:189-217.

Cossins A., Bowler K., 1987: Temperature Biology of Animals. London: Chapman and Hall, 339 p.

Daborn G.R., 1971: Survival and mortality of coenagrionid nymphs (Odonata: Zygoptera) from the ice of an aestival pond. Canadian Journal of Zoology 49, 569–571.

Danks H.V., 1971: Overwintering of some north temperate and arctic Chironomidae. I. The winter environment. Canadian Entomologist 103, 589–604.

Danks H. V., 1978: Modes of seasonal adaptation in the insects: I. Winter survival. Article in The Canadian Entomologist 110, 1167-1205.

Danks H.V., 1987: Insect Dormancy: an Ecological Perspective. Biological Survey of Canada (Terrestrial Arthropods), Ottawa.

Danks H.V., 1991: Winter habitats and ecological adaptations for winter survival. In: Lee, R.E. and Denlinger, D.L. Insects at Low Temperature. Chapman and Hall, New York, pp. 231–259.

Danks H.V., 2002: Modification of adverse conditions by insects. Oikos, 10–24.

Danks H.V., 2005: Key themes in the study of seasonal adaptations in insects I. Patterns of cold hardiness. Applied Entomology and Zoology 40, 199–211.

Danks H.V., 2006: Insect adaptations to cold and changing environments. CanadianEntomologist 138, 1–23.

Danks H. V., 2007: Aquatic insect adaptations to winter cold and ice. Trowbridge, UK: Cromwell Press.

Denlinger L., Lee E., 2010: Low Temperature Biology of Insects. Cambridge University Press, 2010.

Dejours P., 1975: Principles of Comparative Respiratory Physiology. North-Holland Publishing Company, Amsterdam, p. 253.

Dotath H., 1981: The effects of the 1978/79 interim on the Populations of *Sympecma fusca* in north-western Lower Lusatia (*Odonata, Lestidae*). Ent. Ber., 2, 49-52.

Duman J.G., 2001: Antifreeze and ice nucleator proteins in terrestrial arthropods. Annual Review of Physiology 63, 327–357.

Eichler W., 1951: Anopheles — Observations in South Russia. Riv. Malariology 30, 29-38.

Evans K.W., Brust R.A., 1972: Induction and termination of diapause in Wyeomyia smithii (Diptera: Culicidae), and larval survival studies at low and subzero tempera-tures. Canadian Entomologist 104, 1937–1950.

Frisbie M.P., Lee R.E. Jr., 1997: Inoculative freezing and the problem of winter sur-vival for freshwater macroinvertebrates. Journal of the North American Benthological Society16, 635–650.

Gehrken U., 1989: Diapause termination in eggs of the stonefly Arcynopteryx compacta (McLachland) in relation to dehydration and cold hardiness. Journal of Insect Physi-ology35, 377–385.

Gehrken U., Sømme, L., 1987: Increased cold hardiness in eggs of Arcynopteryx compacta (Plecoptera) by dehydration. Journal of Insect Physiology 33, 987–991.

Gilyarov M. S., 1986: Biological Encyclopedic Dictionary. Second ed., Soviet Encyclopedia, 864 p., ISBN 5-85270-002-9.

Gordeev M.I., Perevozkin V.P., 1997: Genetic and ecological effects of prédation by water spiders Argyroneta Aquatica on larvae of anopheles and culex mosquitoes. Russian Journal of Genetics. Publisher: Pleiades Publishing, T. 33. № 5. 704-709 p.

Harabiš F., Dolný A., Šipoš J., 2012: Enigmatic adult overwintering in damselflies: coexistence as weaker intraguild competitors due to niche separation in time. Population Ecology 54:549-556.

Hayashi F., 1994: Life-history patterns in 15 populations of Protohermes (Megaloptera: Corydalidae): Effects of prey size and temperature. In: Danks, H.V. (ed.) Insect Life-cycle Polymorphism: Theory, Evolution and Ecological Consequences for Seasonality and Diapause Control. Kluwer Academic Publishers, Dordrecht, pp. 227–243.

Hauer F.R. and Stanford J.A., 1982: Bionomics of Dicosmoecus gilvipes (Trichoptera: Limnephilidae) in a large western montane river. American Midland Naturalist 108, 81–87.

Hinton H.E., 1960: A fly larva that tolerates dehydration and temperatures from –270°C to +102°C. Nature, 188,336–7.

Holmstrup M., Westh P., 1994: Dehydration of earthworm cocoons exposed to cold: a novel cold hardiness mechanism. Journal of Comparative Physiology B: Biochemical Systemic and Environmental Physiology 164, 312–315.

Holmstrup M. Zachariassen K.E., 1996: Physiology of cold hardiness in earthworms. Comparative Biochemistry and Physiology A: Comparative Physiology 115, 91–101.

Irons J.G., Miller L.K., Oswood, M.W., 1993: Ecological adaptations of aquatic macroinvertebrates to overwintering in interior Alaska (USA.) subarctic streams. Canadian Journal of Zoology 71, 98–108.

Irwin J.T., Lee R.E., 2003: Cold winter microenvironments conserve energy and improve overwintering survival and potential fecundity of the goldenrod gall fly, Eurosta solidaginis.Oikos 100, 71–78.

Kalutsky A. L., 2005: Insects hibernation. Svirel, Moscow, № 2, 7-9p.

Kim M., Robich R.M., Rinehart, J.P., Denlinger D.L., 2006: Upregulation of two actin genes and redistribution of actin during diapause and cold stress in the northern house mosquito, Culex pipiens. Journal of Insect Physiology 52, 1226–1233.

Kiselev V.N., 1998: Fundamentals of Ecology: Textbook allowance. Minsk University, 367p.

Korsunsky V., 1983: General methods of teaching biology. M. Education, 4th edition, Moscow.

Lampert W., Sommer U., 1997: Limnoecology: The Ecology of Lakes and Streams: Special features of aquatic habitats. New York: Oxford University Press.

Lancaster J., Hildrew A.G., 1993: Flow refugia and the microdistribution of lotic invertebrates. Journal of the North American Benthological Society 12, 385–393

Lee R.E., Denlinger D.L., 1991: Insects at Low Temperature. Chapman and Hall, New York, pp. 17–46.

Manger, R., Dingemanse, N. J., 2009: Adult Survival of *Sympecma paedisca* (Brauer) during hibernation (*Zygoptera: Lestidae*). Odonatologica, 38, 55-59.

Miller A.M., Golladay S.W., 1996: Effects of spates and drying on macroinvertebrate assemblages of an intermittent and a perennial prairie stream. Journal of the North American Benthological Society 15, 670–689.

Nebeker A.V., Lemke A. E., 1968: Preliminary studies on the tolerance of aquatic insects to heated waters. National Water Quality Laboratory Federal Water Pollution Control Administration Duluth, Minnesota 55804.

Olsson, T.I., 1983: Seasonal variation in the lateral distribution of mayfly nymphs in a boreal river. Holarctic Ecology 6, 333–339.

Oswood M.W., Miller L.K., Irons, J.G., 1991: Overwintering of freshwater benthic macroinvertebrates. In: Lee R.E., Denlinger D.L. Insects at Low Temperature. Chapman and Hall, New York, pp. 360–375.

Pajunen V.I., Jansson, A., 1969: Dispersal of the rock pool corixids Arctocorisa carinata(Sahlb.) and Callicorixa producta (Reut.) (Heteroptera: Corixidae). Annales Zoologici Fennici 6, 391–427.

Paterson C.G., 1971: Overwintering ecology of the aquatic fauna associated with the pitcher plant Sarracenia purpurea L. Canadian Journal of Zoology 49, 1455–1459.

Pilon J. G., Masseau M. J., 1984: The effect of temperature on egg development in *Zygoptera*: preliminary discussion Faculté des Arts et des Sciences, Université de Montréal, Montreal., Canada.

Radkevich, V.A., 1998: General ecology. Minsk High School, 159 p.

Sawchyn W.W., Gillott, C., 1974: Life history of three species of Lestes (Odonata: Zygoptera) in Saskatchewan. Canadian Entomologist 106, 1283–1293.

Scholander P.F., Flagg W., Hock R.J. and Irving L., 1953: Studies on the physiology of frozen plants and animals in the arctic. Journal of Cellular and Comparative Physiology Supplement 1, 56 pp.

Shvanvich B.N., 1949: The course of general entomology. Publishing house "Soviet science", M.L., 900 p.

Spence J.R., Andersen N.M., 1994: Biology of water striders: interactions between systematics and ecology. Annual Review of Entomology 39, 101–128.

Spielman A., 1962: The influence of rainfall upon the abundance of Cuban Hippelates (Diptera: Chloropidae). Ann. Ent. Soc. Amer. 55 (1): 39 – 42.

Storey K.B., Storey J.M., 1992: Biochemical adaptations for winter survival in insects.Advances in Low-temperature Biology 1, 101–140. JAI Press, London.

Stubbs E., Steven J., 1983: British Hoverflies: An Illustrated Identification Guide (2nd ed.). London: British Entomological and Natural History Society. pp. 253. ISBN 1-899935-03-7.

Tauber M. J., Tauber C. A., 1974: Dietary influence on reproduction in both sexes of five predacious species (Neuroptera). Canadian Entomologist. 106, 921–925.

Tauber M.J., Tauber C.A., 1986: Seasonal Adaptations of Insects. Cornell University and Sinzo Masaki Hirosaki University. New York: Oxford University Press.

Trunova T. I., 2007: Plant and low-temperature stress / T. I. Trunova .– M .: Nauka.- 54 p.

Van de Bund W.J., Groenenkijk D.,1994: Seasonal dynamics and burrowing of littoral chironomid larvae in relation to competition and predation. Archiv für Hydrobiologie 132, 213–225.

Vinogradova E. B., 2007: Diapause in aquatic insects, with an emphasis on mosquitoes. In: V. R. Alekseev et al. (eds.), Diapause in Aquatic Invertebrates. Springer. pp. 83–113.

Wharton D.A., 2003: The environmental physiology of Antarctic terrestrial nematodes: a review. Journal of Comparative Physiology B 173, 621–628.

Wiggins G.B., Winchester N.N., 1984: A remarkable new caddisfly genus from northwestern North America (Trichoptera, Limnephilidae, Limnephilinae). Canadian Journal of Zoology 62, 1853–1858.

Yakhontov V.V., 1964: Ecology of insects. Publishing house: M.: Higher school, 459p.

Zachariassen, K.E., Husby, J.A., 1982: Antifreeze effect of thermal hysteresis agents protects highly supercooled insects. Nature 298, 865–867

9. Appendix

Appendix 1: Mosquitoes prepared to spend the winter in less frozen corners in the «Kazachy Stan» cave, Chelyabinsk Region, Russia
Photo: Evgeny Chibilyov


Appendix 2: Overwintering mosquitoes in the cave of the river Piusa, Estonia.
Photo: Arne Ader


Appendix 3: *Culex* overwinter in the basement in the city of Artyomovsky, Russia.
Photo: Natalya Sharova


Appendix 4: Adult Culex endure winters under snow
Photo: Sergey Toronto