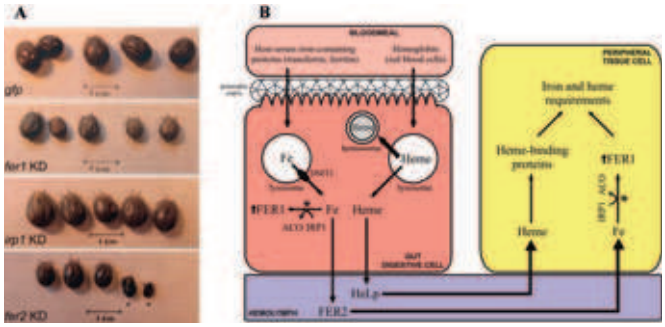
6. Section of Biological and Ecological Sciences

The section associated four workplaces whose research is focused on the mutual relations between organisms, between organisms and the environment and on the functional mechanisms in ecosystems with respect to the anthropogenic effects. The objective is to understand the key processes with the possibility of using the results in medicinal, biotechnological, veterinary and agricultural practice as well as as the bases for rational exploitation of the landscape. The research is also focused on animal biodiversity, vertebrate evolutionary ecology and adaptations of their behaviour, on the research of the evolution, structure and ecological role of plant biodiversity (from the genetic level through the level of organisms and communities to ecosystems) and on the study of the insect as a biological model as well as pest. Another area of interest includes the interactions of parasitic and symbiotic organisms, organism communities in the soil ecosystems, the functioning of the ecosystems of valley reservoirs and lakes, the study of the global cycle of carbon, energy and material flows through ecosystems, and the ecology of the landscape influenced by man. An important component of the research is the use of advanced methodologies in ecology, in particular the methods of molecular biology, of remote research of the Earth and of mathematic modelling with an emphasis on a systemic approach.

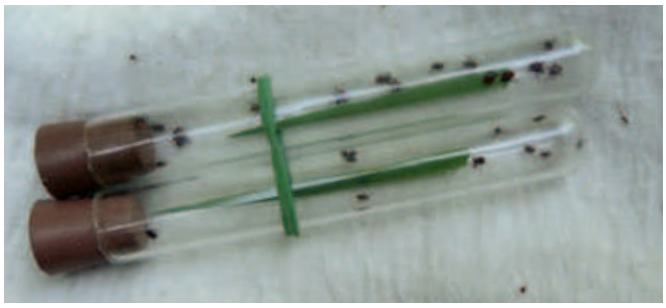
From the results from 2009, we present:

Discovery of a Tick Ferritin 2, a Novel Iron-Transporting Protein as a Candidate for Anti-Tick Vaccine
(Biology Centre)

During research on the hard tick (*Ixodes ricinus*), the vector of tick-borne encephalitis and Lyme borreliosis, and the ways it manages the surplus of iron originating from the host's blood, the authors discovered a so-far unknown protein designated as ferritin 2. Unlike the already earlier described ferritin 1, serving for intracellular ironstorage, the newly-discovered ferritin 2 is secreted into the body fluid (hemolymph) of the tick. The functions of ferritin 1, ferritin 2 and the iron-regulatory protein (IRP) were studied using a method of RNA interference, which makes it possible specifically to block their production. Surprisingly, it was demonstrated for the newly discovered ferritin 2 that its main role is in iron transport from the tick's digestive tract to other organs, mainly the salivary glands or ovaries. Impairment of the iron metabolism had a negative impact on reproduction and further development of the ticks. The most important result was the discovery that the suppression of the production of ferritin 2 led to a limitation of the ticks' ability to feed on the host, with more than half of the ticks dying during blood feeding on the host. A similar effect was achieved also during experimental vaccination of rabbits with recombinant ferritin 2, where the antibodies imbibed with the host blood efficiently blocked ferritin 2 in the tick's gut. This fact, together with a remarkable molecular difference from the ferritins of mammalian hosts, makes the tick ferritin 2 a promising candidate for an efficient 'anti-tick' vaccine limiting the ability of the ticks to feed and thanks to their weakening also the risk of the transmission of the pathogenic progenitors of infectious diseases. The potential veterinary use of ferritin 2 is protected by a Czech* as well as an international** patent claim. The pilot tests of this vaccine on cattle have already been performed within foreign cooperation and the results achieved greatly support further development towards commercialisation of this vaccine and its broad application, especially in the countries where economical losses caused by tick feeding in the livestock production are enormous.



Tick ferritin 2: a novel iron-transporting protein and candidate anti-tick vaccine.
Panel A: The influence of RNA interference on the ticks' ability to feed on the host. GFP – the group injected with the control (GFP) two-fibre RNA; fer 1 KD – the influence of the suppression of the intracellular ferritin 1; IRP KD – the influence of the suppression of the iron-regulating protein (IRP); fer2 KD – the influence of the suppression of the secreted ferritin 2.
Panel B: Model scheme of the metabolism of iron in ticks.



The phases of the research at which males and females are studied separately.
(Photo: Archives of the Institute of Parasitology of the Biology Centre)

The collection of ticks in the field
(Photo: Archives of the Institute of Parasitology of the Biology Centre)

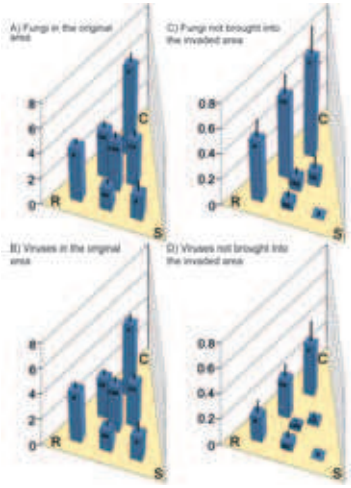
Based on the results attained and the work of the laboratory of Pedro L. Oliviera (Rio de Janeiro), the authors anticipate two different paths of heme (blood pigment) metabolism and non-heme iron from the host's blood. The elimination of ferritine 2 leads to a disruption of the transmission of non-heme iron, which is important for instance for the function of enzymes in the respiratory chain. Moreover, it leads to the accumulation of toxic iron in the tick's gut. This significantly disrupts the ability of the tick to blood feed and further develop.

Hajdušek, O. – Sojka, D. – Kopáček, P. – Burešová, V. – Franta, Z. – Šauman, I. – Winzerling, J. – Grubhoffer, L.: Knockdown of proteins involved in iron metabolism limits tick reproduction and development. Proceedings of the National Academy of Sciences of the United States of America. Vol. 106, (2009), pp. 1033–1038.
** Czech patent application: Kopáček, P. – Hajdušek, O.: 'Feritin 2 pro imunizaci organismu proti klíšťatům.' Industrial Property Office of the Czech Republic, PV 2008-402 (25 June 2008).*
*** International Patent Application: Kopáček, P. – Hajdušek, O.: 'Ferritin 2 for the host immunization against ticks.' Industrial Property Office of the Czech Republic, PCT/CZ2009/000085 (18 June 2009).*

Synergy in Plant Invasions: Fast-Growing Species Experience Greater Enemy Release
(Institute of Botany)

It is known that plant species do not bring with them into a new region most of their enemies which regulate in a natural way the size of their populations in the area of their original spread and thus keep them from dominating the other plants. The idea that this mechanism is one of the important causes of invasions is generally accepted. The study conducted is the first to show that the amount of the enemy release depends on the type of plant. The analysis of the fungal and viral diseases of 243 plant species of European origin which are invasive in the United States revealed that two mechanisms generally considered as the main causes of the invasions of non-indigenous plants, hence the high level of resources and escape from enemies, work in synergy. Fast-growing plants, adapted from the areas of their original spread to damp and nitrogen-nutrient-rich sites, hence a habitat with high levels of resources, are more predisposed to fungal and viral diseases (see the figure on the next page). In the invasion of a new area, however, these species lose many more of these pathogens than plants from resource-poor sites, which helps their spread. This result contributes to the explanation of why plant invasions are the most frequent in an environment rich in nutrients and other resources which is usually created by human activities. Invasive, fast-growing species thus actually gain a double advantage – an increased amount of resources allows them to dislodge from the communities slow-growing plants but also fast-growing indigenous

species, because they are regulated in their natural habitat by the activity of their natural enemies. This discovery helps us understand better the dramatic invasions of some plant species and indicates that the current global changes like the disruption of the landscape by mankind and its enrichment with nutrients will continue to be accompanied by massive invasions of plant species in various parts of the world. The results of this study, moreover, confirm that the distinction between species based on their geographical origin (a principle that is sometimes questioned) is justified by the differences in the behaviour and characteristics of the indigenous and non-indigenous species.



Synergism in plant invasions: fast-growing plants experience greater enemy release. The average number of species of fungal and viral pathogens discovered in plants with various types of life strategies in the original area in Europe (A, B) and the number of these pathogenic species which plants escaped from after importation into North America (C, D). The competitive (C) and ruderal (R) strategies are typical for fast-growing species; the species able to tolerate stress (S) grow more slowly.
(Adopted from Blumenthal et al., Proc. Natl. Acad. Sci. USA 106: 7899–7904, 2009)

Cooperation took place with partners within the Research Coordination Network on Integrating the Ecology and Evolution of Invasions (NSF, USA).

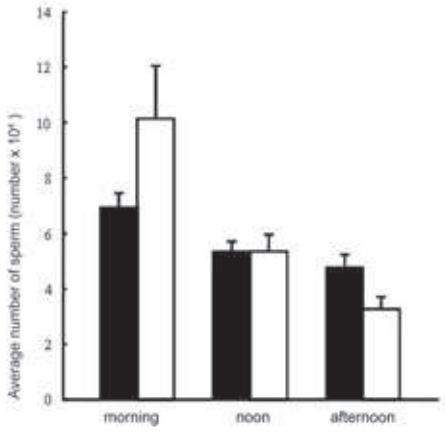
Blumenthal, D. – Mitchell, C. E. – Pyšek, P. – Jarošík, V.: Synergy between pathogen release and resource availability in plant invasion. Proceedings of the National Academy of Sciences of the United States of America. Vol. 106, (2009), pp. 7899–7904.
Pyšek, P. – Hulme, P. E.: Invasion biology is a discipline that's too young to die. Nature. Vol. 460, (2009), p. 324.

Sexual Selection in the Bitterling Fish
(Institute of Vertebrate Biology)

The spawning of the bitterling fish. A male (on the right) and a female bitterling near a mussel into whose gill chambers the fish lay their eggs.
(Photo: Archives of the Institute of Vertebrate Biology)

Sexual selection explains the mechanisms and processes influencing the unequal reproductive success among individuals within a population. Its study can explain a wide range of apparently disadvantageous behavioural patterns or morphological adaptations but can also offer important background information for ideal pairing in livestock breeding, which will increase production (growth rate) and minimise offspring mortality. In the research, the authors focused on four aspects of sexual selection and found the following: (1) the viability, growth rate and survival of offspring were significantly higher when females were allowed to select a mate. The most likely mechanism was olfactory choice based on a complex of immune genes. (2) The reproductive success of males differed according to type of environment. In a highly competitive environment, males who invested in colouration signalling aggression and the higher production of sperm were successful. However, those males also impeded the females' ability to choose their mate. On the contrary, females had more opportunities to express their choice of a suitable partner in an environment with a lower level of competi-

tion. (3) Male–male competition for fertilisation of the fish eggs may lead to sperm depletion in the short and long terms (hours and weeks) and hence considerably increase the number of unfertilised eggs. (4) The successfulness of natural reproduction in the bitterling fish is primarily influenced by the flow-rate regime and ambient temperature. The higher water temperatures in European rivers recorded in the last few years along with the strong anthropogenic influence on the character of rivers (weirs, dykes) are likely responsible for the population increase and invasion of the bitterling in most of Europe.

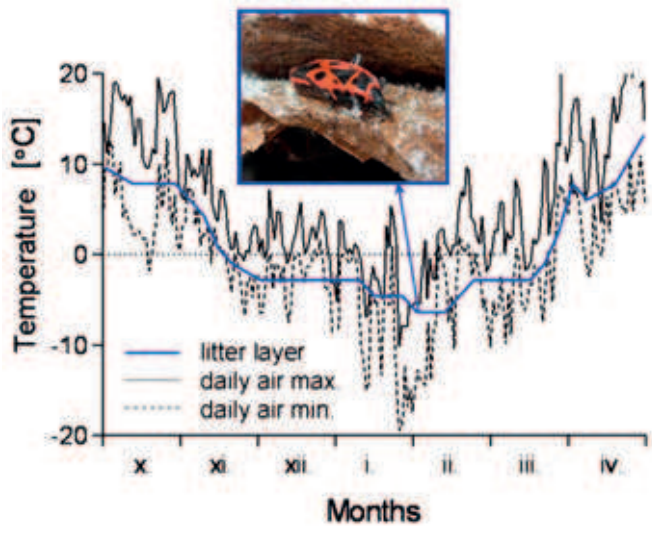


The volume of sperm in the ejaculate changes depending on the sperm competition. The daily fluctuation of the average amount of sperm in the ejaculate of the males of the bitterlings exposed to sperm competition (white column) and the control group without sperm competition (black column). Morning (9–10 a.m.), Noon (12–1 p.m.), Afternoon (3–5 p.m.).
(Photo: Archives of the Institute of Vertebrate Biology)

Cooperating entities: University of St Andrews (GB), University of Leicester (GB), University of Lodz (PL)

Reichard, M. – Ondračková, M. – Bryjová, A. – Bryja, J. – Smith, C.: Breeding resource distribution affects selection gradients on male phenotypic traits via sexual selection: experimental study on lifetime reproductive success in the bitterling fish (Rhodeus amarus). Evolution. Vol. 63, 2 (2009), pp. 377–390.
Konečná, M. – Jurajda, P. – Reichard, M.: River discharge drives recruitment success of the European bitterling (Rhodeus amarus) in a regulated river in Central Europe. Journal of Fish Biology. Vol. 74, 7 (2009), pp. 1642–1650.
Smith, C. – Pateman-Jones, C. – Zieba, G. – Przybylski, M. – Reichard, M.: Sperm depletion as a consequence of increased sperm competition risk in the European bitterling (Rhodeus amarus). Animal Behaviour. Vol. 77, 5 (2009), pp. 1227–1233.
Casalini, M. – Agbali, M. – Reichard, M. – Konečná, M. – Bryjová, A. – Smith, C.: Male dominance, female mate choice and intersexual conflict in the rose bitterling (Rhodeus ocellatus). Evolution. Vol. 63, 2 (2009), pp. 366–376.

Heat-Shock Protein Expression as a Part of the Complex Adaptation for Cold Tolerance in the Insect, *Pyrrhocoris apterus*
(Biology Centre)



Overwintering *Pyrrhocoris apterus*. The body temperature of the diapausing heteroptera, *Pyrrhocoris apterus*, drops in the long term to temperatures below zero during their overwintering in a layer of leaf litter.
(Photo: Archives of the Institute of Entomology of the Biology Centre)

Body temperature of the overwintering temperate-latitude insect individuals drops below 0°C often and for long periods (see the figure above). Numerous insect species survive temperature extremes at which no vertebrate organism could survive. The key for survival success lies in the seasonal transition into the diapause state and the activation of a complex of adaptions, which result in increased

cold and frost tolerance. The employees of this institute have studied this complex on the model heteroptera species, *Pyrrhocoris apterus*. Entering diapause is a case of deep phenotypic change, which is based on an alteration of gene transcription. Among others, also the transcription of genes coding the heat shock proteins from the Hsp70 kDa family is changed. The authors investigated the ability of the heteroptera to react to the exposure of both high and low temperatures by accelerating the transcription of the genes coding these proteins. They cloned and sequenced the fragments of the genes for the inducible and cognate forms of Hsp70 kDa heat shock protein. The abundance of mRNA transcripts was then monitored using quantitative real-time PCR and the abundance of the protein products using the Western blot analysis. The abundance of transcripts as well as the protein of the inducible form were significantly up-regulated in response to both heat and cold shocks. The authors prepared a (95 bp-long) dsRNA for the inducible form. The injection of the dsRNA into the heteroptera prior to their exposure to the temperature shock RNAi method caused an elimination of the heat- and cold-shock-induced transcriptional response. The RNAi predictably entirely prevented repair of damage caused by high temperatures. It was newly determined that the elimination of the Hsp70 kDa transcription also significantly lowers the ability of the heteroptera to repair damage caused by cold shock. The results attained have thus proved that the transcriptive activation of the Hsp70 kDa heat shock protein is an important component of the adaptive complex of cold tolerance in the insect, *P. apterus*.

Koštál, V. – Tollarová-Borovanská, M.: The 70 kDa heat shock protein assists during the reparation of chilling injury in the insect, Pyrrhocoris apterus. PLoS ONE. Vol. 4 (2009), e4546, pp. 1–9.
Tollarová-Borovanská, M. – Lalouette, L. – Koštál, V.: Insect cold tolerance and repair of chill-injury at fluctuating thermal regimes: role of 70 kDa heat shock protein expression. Cryo-Letters. Vol. 30 (2009), pp. 312–319.

Other notable results:

1. Identification of a single peridinine sensing Chl-a excitation in reconstituted PCP complex by crystallography and spectroscopy *(Biology Centre)*
2. Polyphasic characterisation and taxonomic revision of planktonic cyanobacteria of *Anabaena* spp. (Nostocaceae) *(Biology Centre)*
3. Effect of overwintering cattle on the structure and activity of soil microbial community, which ensures nitrogen transformation in soils through the denitrification process, with regard to N_2O emissions *(Biology Centre)*
4. Biological invasions: Europe on a crossroad? *(Institute of Botany)*
5. The ecological, taxonomic and evolutionary consequences of genome duplication *(Institute of Botany)*
6. Timing of flowering is a non-trivial result of selection pressure from pollinators and herbivores *(Institute of Botany)*
7. Survival strategies of plants in disturbed environments *(Institute of Botany)*
8. Carotenoid ornaments, sexual selection and immunogenetics in passeriformes *(Institute of Vertebrate Biology)*
9. Low-frequency electromagnetic fields generated by power lines disturb magnetic orientation of ungulates *(Institute of Vertebrate Biology)*
10. Thermal acclimation of the swimming capability of newt larvae: the effect of daily temperature fluctuations during embryogenesis *(Institute of Vertebrate Biology)*
11. Effect of summer flood on carbon deposition of CO_2 in wetlands *(Institute of Systems Biology and Ecology)*
12. Photosynthesis in silico. Understanding complex behaviour from molecules to ecosystems *(Institute of Systems Biology and Ecology)*
13. Magnetically modified microbial cells as intelligent whole-cell biocatalysts *(Institute of Systems Biology and Ecology)*
14. Surface temperature change of spruce forest as a result of bark beetle attack: remote sensing and GIS approach (*Institute of Systems Biology and Ecology*)
15. Structure of the motor subunit of type I restriction--modification complex EcoR124I *(Institute of Systems Biology and Ecology)*