

Pests

Insects (Insecta) are the largest phylum of arthropods. They play a very important positive role as well as negative in the functioning of ecosystems, e.g. bees are essential for the human economy, but a large group of insects can cause damage to crops. The vast majority of insects are herbivores. They feed on different parts of plants, causing the reduction or even complete loss of crops. Primary pests in forests are insects that attack strong and healthy trees. Lepidoptera includes butterflies and moths. While most of these first have a positive impact on the environment, whereas moths are a threat to forest plantations, orchards and agricultural areas. The damage is usually caused by the larvae, which feed intensively before metamorphosis.

The gypsy moth (*Lymantria dispar*) is a common forest pest, occurring also in parks and avenues. Its caterpillars feed on oak leaves, especially English oak, as this develops the earliest.

Baculovirus as biopesticide

The largest family of viruses infecting insects is the Baculoviridae family and the largest group of host insects are butterflies. The main characteristic of baculovirus is that one species of virus suitable for the insect, like a key in a lock, which is used for the production of biopesticides based on viruses from the Baculoviridae family. Such biological plant protection products are characterized by high selectivity because they act on strictly defined species without harming "by chance" beneficial insects, like bees. In addition, they are unable to infect human or other animals, which means that they are very safe and can be used on a large scale. Due to the fact that they are a natural part of nature, since they have evolved with their host-insects, are completely safe.

In Poland biopesticide used to protect plants is based on the infecting baculovirus pest of fruit trees, which is the codling moth (*Cydia pomonella*). Baculovirus, which is an active component of the preparation is *Cydia pomonella* Granulovirus (CpGV). In North America, where the gypsy moth is a huge problem, biopesticide called Gypchek™ is available containing baculovirus infecting this insect, i.e. LdMNPV (*Lymantria dispar* Multinucleopolyhedrovirus).

Aim of the project and basic research

Due to the excessive use of chemical pesticides in the world in recent decades, many research centers focused on finding biological plant protection products that are safe for the environment, act selectively and enough quickly are able to kill feeding pests. During the preliminary tests biological activity of LdMNPV isolates collected from three regions of Poland and the above mentioned American biopesticide was compared *in vivo* (i.e., in a living organism, which in this case are the gypsy moth caterpillars). We found that the baculovirus derived from Biebrzanski National Park (LdMNPV-BPN) has a significantly higher activity against the host than other isolates. In addition, in the course of further research it has been established by our team, that although LdMNPV-BPN infects gypsy moth, it has significantly different genetic structure (the nucleotide sequence of the genome) from other LdMNPV available in the NCBI database, as well as the Polish isolates. These results led to further research and the creation of this application.

The main objective of the project is to determine the role of three selected genes: *egt* (ecdysteroid UDP-glucosyltransferase), *hrf-1* (host range factor-1) and *vfgf* (viral fibroblast growth factor) in the increased biological activity of baculovirus that infects gypsy moth from the area of Biebrzanski National Park in Poland (LdMNPV-BPN). In the literature these selected genes function as potential virulence factors, which affect the virus speed of killing. In this proposal, it is planned to construct recombinant baculoviruses based on Polish reference LdMNPV isolate and exchanged *egt*, *hrf-1* and *vfgf* genes derived from LdMNPV-BPN. The constructed recombinants will be tested in gypsy moth cell line and *in vivo* in gypsy moth caterpillars with respect to its biological activity. Polish isolate LdMNPV-BPN having increased virulence relative to its host will now be described on the basis of the three proteins. Implementation of the project on one hand is to expand knowledge of genetics baculovirus, on the other, in the future this will allow for quick search for natural and most importantly safe crop protection agents.