

The immune system of spiders is based on innate, non-specific reactions at humoral and cellular level, mainly induced by hemocytes. At each stage of life cycle spiders are constantly exposed to environmental stress, but at the beginning of ontogenesis, developing embryos are protected by a physical barrier – egg sac made of spider silk, which isolates them to some extent from the external environment. Simultaneously with the individual development progress, a functional immune system must be formed, which will make the embryos independent of the passive protection provided by the cocoon and egg shells. In this way, juveniles emerging from the cocoon can rely on their own immune responses, stimulated by the action of biotic and abiotic environmental factors.

In order to complete the knowledge on spider immune potential during ontogenesis, this paper has been divided into two main parts, concerning the immunity of spiders at the embryonic and post-embryonic period of development. To compare the range of passive and active immunity of embryos, representatives of two species of spiders were selected: *Parasteatoda tepidariorum* (Theridiidae) and *Pardosa lugubris* (Lycosidae) - differing in their biotope, strategy hunting and of parental care.

In the first part of the research, it was checked whether at the early stages of individual development *P. tepidariorum* and *P. lugubris* spiders have an active immunological potential, which is measurable at the level of expression of genes encoding selected antimicrobial proteins (AMPs), the presence of low-molecular proteins on the protein profile and at the concentrations of selected conservative AMPs detected by immunodetection. The spiders were also exposed to a simple form of immunostimulation by isolating embryos from egg sac. The obtained results indicate that embryos of *P. tepidariorum* and *P. lugubris* spiders do not rely solely on a passive cocoon protection, but also have their own active immunological potential. They show both constitutive and stimulated activity and functionality of the immune system, even before being able to leave the egg sac. *P. lugubris* appearing to have a higher immune potential, which may be related to the potentially higher exposure of *P. lugubris* embryos to the pathogens and cocoon-damaging factors, which related to the maternal care strategy.

In order to obtain a more complex knowledge of the spider's immune system functionality during the progressing individual development, this research was supplemented with the second experimental set-up. The impact of different degrees of immunostimulation of selected markers of innate immunity of spiders in later life stages was assessed. It has been shown that

the level of analyzed innate immune response markers depends on the intensity of immunostimulation. The immune system of both immature and mature females is stimulated only in response to the highest concentration of used immunostimulants (foreign body + infection of Gram-positive bacteria *Bacillus subtilis*). Immature and mature spiders differ in their immune potential, characterized by the expression of genes encoding proteins related to the immune system, the level of synthesis of HSP, lysozyme and other low-molecular proteins with a potential immune function, as well as to the percentage of live, apoptotic and dead hemocytes.

In this study, it was shown that the innate immunity of *P. tepidariorum* differs between individual developmental phases, both at the early and late stages of ontogeny. Interspecies differences in the functioning of the spider immune system over the course of individual development are dependent on parental investment strategies, as well as on the biotope and lifestyle of the studied spiders. It is manifested by a different allocation of energy resources between processes related to the reproduction and immunity. Moreover, embryos of semelparous spiders (reproducing once, occasionally twice in a lifetime) are characterized by a higher immunological potential compared to embryos of iteroparous spiders (reproducing many times during their lifetime), which is associated with a higher parental investment in relatively few offspring produced by *P. lugubris* females compared to *P. tepidariorum*. Results in this research will fill the gap in the knowledge about the development and activity of the spiders' immune system throughout their ontogeny.