Abstract:

Štěpán Ryba, 2012

Molecular epidemiology of selected viral, bacterial and fungal disease of honeybees in the Czech Republic

Altogether, the six most common bee viruses which infect the honey bee (Apis mellifera) were monitored in the territory of the Czech Republic between 2006 and 2009. Parallel infections of viruses (DWV, ABPV and BQCV) in bee adults and parallel co-infection of viruses with fungal diseases caused by Nosema apis and Nosema ceranae were confirmed by PCR tests. A new sensitive method of detection of the originator of the American foulbrood (Paenibacillus larvae) from bee debris was developed for the practical use of detection of AFB disease in bee populations. Various approaches for the extraction of spores from bee debris and lyses of spores were compared. The sensitivity of PCR tests for the presence of Paenibacillus larvae in debris was compared with the classic cultivation method. The PCR method for the detection American foulbrood was further studied and developed to be more efficient. A new method, based on a matrix-like sample re-arrangement and a use of pooled samples, has been developed for testing 1000 samples in 35 PCR reactions. Another goal was to develop a robust and fast screening method for American foulbrood based on the cultivation test using paper sheets RIDA®COUNT with a specific cultivation medium, specific selection conditions for Paenibacillus larvae and chromogen visualization of the grown bacterial colonies. This method has been evaluated in the laboratory conditions and its sensitivity has been verified. Finally, the differential diagnostics of both microsporidia on a molecular level was implemented in the area of detection of bee fungal diseases and a possibility of microscopic differential diagnostics of Nosema apis and Nosema ceranae was confirmed. This microscopic method can be used in field conditions. The results of a microscopic examination were validated parallel with the PCR test.