

ABSTRACT

Immunodeficient honey bee (*Apis mellifera*) colonies suffer from broad range of parasites including eukaryotic protozoa. Despite this fact, the eukaryotic parasites are still poorly documented in the Czech Republic. The presence of eukaryotic parasites (*Nosema ceranae*, *Nosema apis*, *Crithidia mellificae* and *Apicystis bombi*) was observed in different apiaries in the Czech Republic. The samples were taken in 9 apiaries in 53 beehives during the 2014/2015 season. From each beehive, 10 adult of honey bees were taken from the peripheral comb in triplicate. DNA was isolated from every sample of honey bees. The parasites were detected by polymerase chain reaction (PCR) with specific primers. The treatment fall of parasitic mite *Varroa destructor* was obtained from beekeepers for season of 2014. *Crithidia mellificae* was detected by 5 types of specific primers (SEF, SER; SSU, SSU rRNA, Cyt b, Tryp cyt b) and positive amplicons were cloned and sequenced. The obtained sequences were compared with GeneBank and showed similarity from 98-100% to sequences of *Lotmaria passim* (Trypanosomatid). *Crithidia mellificae* was not detected. *L. passim* had prevalence of 79,2% and is reported in the Czech Republic for the first time. Primer Tryp-cyt b is recommended for the routine detection of *L. passim*. *Nosema ceranae* was detected in 47 apiaries with the total prevalence of 86,8%, while *Nosema apis* was detected in 4 samples with prevalence of 5,7% only. It correspond the global replacement of *N. apis* by *N. ceranae*. Presence of *Apicystis bombi* was not confirmed using specific primers in PCR. The interaction among the parasites, parasitic infection and colony collapse and presence of parasites with co-infection of mite *V. destructor* were analyzed by correlation tests. The correlations among the occurrence of observed eukaryotic parasites were not found. The treatment fall of *V. destructor* was correlated with colony losses in winter 2014/2015 and with the presence of *Nosema ceranae*. Results are discussed with possible impact of eukaryotic parasites on honey bee colony losses in immunodeficient honey bees.

Keywords: honey bee, *Apis mellifera*, *Nosema ceranae*, *Nosema apis*, *Crithidia mellificae*, *Apicystis bombi*, *Lotmaria passim*, *Varroa destructor*, PCR, Microsporidia, prevalence, collapse, co-infection