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Fisrt report of indigenous *Dermacentor reticulatus* populations in Belgium and preliminary study on associated Babesiosis pathogens

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Background: The occurrence of indigenous clinical cases of canine (Losson *et al.,* 1999) and equine (Mantran *et al.,* 2004) babesiosis in Belgium during the last two decades suggested that the vector of the pathogens responsible for these diseases, *Dermacentor reticulatus,* could be present in Belgium. Recent reports indicated an expanding geographical distribution of *Dermacentor reticulatus* in Western Europe but until now it was uncertain that indigenous *Dermacentor* populations were established in Belgium.

Methods: Four different locations throughout Belgium, identified as potential *Dermacentor reticulatus* sites, were monitored by flagging during 2010. Ticks were stored in 100% ethanol immediately after trapping and morphologically identified. Tick DNA extraction was performed according to a proteinase K protocol (20mg/ml) following by an ethanol precipitation and to avoid false-negative results, an additional PCR targeting the tick 16S rRNA gene was performed. Only tick-DNA positive samples were further analyzed for the presence of *Babesia* spp. A *Babesia* spp. genus-specific PCR was applied based on the amplification of the 18S rRNA gene.

Results: Two different tick species were identified, *Ixodes ricinus* and *Dermacentor reticulatus*. A total of 282 *D. reticulatus* adult ticks were collected from the 4 sites. Ticks were found mainly from early March until the end of May with a peak of activity in March.

Four out of 188 tick extracts remained negative for the 16S rRNA gene PCR even after diluting the samples 10 and 100 X. The remaining 184 DNA extracts yielded negative results for *Babesia*.

Conclusions: This is the first record of indigenous questing populations of *D. reticulatus* in several areas of Belgium. The low number of ticks examined for the presence of *Babesia* spp. specific DNA did not allow us to conclude about the carrier status of *B. canis canis* by this tick species. Additional studies should be carried out in order to define more accurately the distribution and vectorial capacity of this tick species in Belgium.