Intracellular bacterial parasites from the *Anaplasma* genus in wild game animals in Poland

Anna W. Myczka, Zdzisław Laskowski

Department of Ecology and Evolution of Parasitism , The Witold Stefański Institute of Parasitology, Polish Academy of Sciences, Warsaw

INTRODUCTION. The spread of pathogenic microorganisms in the environment is mostly dependent on the presence of natural reservoirs of these microorganisms in environment. Wild game animals are serious problem because they are a reservoir of microorganisms difficult to detect and control. A wild animals are potentially reservoirs of *Anaplasma* spp.

Anaplasma sp. is Gramm-negative bacteria, which is an obligatory intracellular parasite of animals and humans. Anaplasma sp. is the etiological agent of anaplasmosis (GA – granulocytic anaplasmosis) – which is a zoonosis caused by direct contact with sick animal or indirectly by ticks (tick-borne disease).

This work aims to detect and identify bacterial parasites in tissues from wild animals: wild boar (Sus scrofa), roe deer (Capreolus capreolus), red deer (Cervus elaphus).

MATERIALS AND METHODS. The tested material were liver and spleens from: wild boars, roe deers and red deers. The material was collected during the hunting season of 2017/2018 in the Strzałowo Forest District. Spleens and livers of twenty random animals (from each genus), were selected for the study, and analysed.

The DNA was isolated using the MiniKit100 (Syngen) according to the manufacturer's protocol. Isolated DNA was used to PCR and to nested-PCR (semi-nested). The PCR products were separated on an agarose gel. Positive samples were purified and send to sequencing.

RESULTS. Analysis of samples from roe deer and red deer using the classical variant of PCR has allowed the detection of DNA of the expected size (in order) in three and one samples. Detection of parasitic bacteria in samples from wild boars using the basic variant of PCR did not give any positive results.

Further analysis based on nested-PCR allows detection of DNA of the predicted size in 5 wild boars for 20 samples, the prevalence was 20%. In the case of roe deer, for 20 tested individuals, in 19 samples *Anaplasma* sp. were found. In the case of red deer the predicted size of DNA was found in 14 individuals.

Two sequencing samples were selected from each test group. Attempts from red deer and wild boar showed 100% similarity to the sequence of *Anaplasma phagocytophilum* deposited in the NCBI GeneBank database. In the case of roe deer, the samples obtained showed 99.7% similarity to the sequence of *Anaplasma phagocytophilum* derived from red deer and wild boar.

CONCLUSIONS. Wild game animals in Poland are carriers of intracellular bacterial parasites from the *Anaplasma* genus. In wild boars, red deer and roe deer, the presence of *Anaplasma* phagocytophilum, which is potentially pathogenic to humans, has been detected. This means that game animals in Poland are a natural reservoir of intracellular bacterial parasites potentially pathogenic to humans.

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