

Examination of pathogens affecting wolves in Southern Poland – a preliminary results

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The wolf (*Canis lupus*) is the largest predator from Canidae family living in Europe. The wolf has been protected throughout Poland since 1998. Now, according to official data, there are about 1200 wolves in Poland. They are settled permanently in the east, north-east and south part of the country and are re-introduced to the Western Poland. It is worth to add that wolves are natural hosts for a wide range of intestinal parasites and some of these parasites may also infect domestic animals as well as humans. The aim of this preliminary study was to examine intestinal parasites and to estimate the prevalence of protozoan parasites with particular emphasis of *Neospora caninum* using microscopic, serological and molecular methods. Faecal samples were collected during winter months in 2015/2016 in Lower Silesian Voivodship. Tissue samples were possessed from animals which were vehicle killed or found dead in the Subcarpathian Voivodeship. Wolf seats were examined microscopically for the presence of eggs or oocysts of intestinal micro- and macroparasites using flotation method. Muscle samples were tested for the presence of *Trichinella* nematode. Each sample was examined individually by enzymatic digestion method according to standard protocol. DNA was isolated from all faecal samples according to protocol for isolation of genomic DNA from stool. For molecular examination of apicomplexan parasites three different PCR protocols were employed; first with primer set amplifying the entire intragenic region between 18S and 28S rRNA ribosomal DNA, second PCR based on small subunit ribosomal RNA gene and third PCR protocol using primers spanning noncoding NC-5 region applied to detect *N. caninum* DNA. In coprological survey the following gastrointestinal helminth eggs have been found: *Alaria* sp., *Capillaria* spp., *Trichuris* sp., taeniid tapeworms, *Ancylostoma* spp. and *Moniezia* spp. Oocysts of *Eimeria* spp. were detected in all of the tested samples. Using molecular techniques two samples were classified as positive in first PCR and two when second protocol was used. None of the samples were classified as positive in PCR protocol using *Neospora* specific primers. The digested muscle samples were examined under a microscope for the presence of *Trichinella* larvae. *Trichinella* spp. larvae were found in all of the examined samples. Our results are consistent with previous findings on intestinal pathogens affecting wolves and with a high *Trichinella* spp. prevalence in wolves.