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# The XXIIIth Congress of the Polish Parasitological Society

4-7 September 2013, Szklarska Poręba-Piechowice

# ABSTRACTS

### **SESSION II**

# Faunistics investigations and ecology of parasites

#### Long-term spatiotemporal stability and dynamic changes in the haemoparasite community of bank voles (*Myodes* glareolus) in NE Poland

#### Anna Bajer<sup>1</sup>, Renata Welc-Falęciak<sup>1</sup>, Małgorzata Bednarska<sup>1</sup>, Mohammed Alsarraf<sup>1</sup>, Jolanta Behnke-Borowczyk<sup>2</sup>, Edward Siński<sup>1</sup>, Jerzy M. Behnke<sup>3</sup>

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Long-term field studies on parasite communities offer the best insight into the ecological and evolutional processes shaping host-parasite interactions. The aim of our study was to identify the principal factors regulating long-term trends in the haemoparasite communities of bank voles. In the years 1999, 2002, 2006 and 2010 three isolated populations of bank voles (n=880) were sampled in the Mazury lake district.

Overall, 90.8% of the bank voles harboured at least one of the species of haemoparasites studied. However, prevalence (all species combined) showed different temporal changes in each of the three sites. In voles from Urwitalt, prevalence increased consistently year by year, whereas in Talty the peak years were 2002 and 2006, and in Pilchy, prevalence oscillated without a clear pattern. Across the study, bank voles harboured a mean of 1.75±0.034 haemoparasite species and species richness remained stable with no significant between-year fluctuations or trends. However, each of the 5 constituent species/genera showed a different pattern of spatiotemporal changes. The overall prevalence of *Babesia microti* was 4.9% but this varied significantly between years peaking in 2006 and declining again by 2010. For *Bartonella* spp. overall prevalence was 38.7% and this varied with the year of study, but the temporal pattern of changes differed between the 3 sites. The overall prevalence of *Haemobartonella* spp. was 68.3% with an increase in prevalence with year of study in all 3 sites. *Hepatozoon erhardovae* had an overall prevalence of 46.8% but showed a marked reduction with each successive year of the study, and this was consistent in all three sites. The overall prevalence of *Trypanosoma* spp. was 15.4%, varying significantly between sites, but showing temporal stability.

Among the five studied blood parasites, the pattern of spatiotemporal changes differed depending on parasite species. For some parasites, the effect of host age was also confirmed as playing an important role. However, despite between-sites and between-years dynamics, overall the haemoparasite community remained quite stable over the 11 year period of the study.

### Environmental factors influencing infection with helminths of wild Polish horses

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The Polish horse (*Equus caballus gmelini* Ant. silvatica Vet. type) is a very characteristic, primitive breed which was fully developed and formed more by environment influence rather than human domestication. These horses are very resistant to lung diseases, gastrointestinal infections, and have the ability to adapt to climate and environmental changes. These horses are mostly afflicted with different types of helminths, mainly nematodes, trematodes and cestodes. Although those infections are usually asymptomatic, sometimes the mortality of the horses is high.

The aim of study was to determine the influence of seasonality, sex, the age of the horse and the environment on infection with helminths. The study was conducted in autumn 2011 and spring 2012 in the Research Station of the Polish Academy of Sciences in Popielno and Biebrza National Park. The study was conducted on 20 horses from Popielno and 18 horses from Biebrza. Fresh faeces samples were collected from each studied Polish horse avoiding any contamination from the ground. Further examination of the samples was performed in the Veterinary Hygiene Institute in Gdańsk-Oliwa to determine the precise faecal egg count (EPG – eggs per gram) in each sample. A modified McMaster method was developed to confirm the required specific parasitological examination. The results were analyzed with the following statistical tests: Kolmogorov-Smirnov test, Pearson chi square test, Fisher test, U-Mann Whitney test.

The most common parasites were *Trichonema* spp., *Strongylus* spp., *Dictyocaulus arnfieldi*, *Oxyuris equi* and *Parascaris equorum*. Neither season nor horse sex was found to have an influence on invasion prevalence. The prevalence of *Parascaris equorum* was significantly higher in Biebrza National Park compared to Popielno, however, in Popielno, a higher prevalence of *Strongylus* spp. was noticed. In young horses, the EPG was significantly higher than in older ones.

### *Neospora caninum* antibodies in moose (*Alces alces*) in north-east area of Poland

#### Aleksandra Cybulska, Katarzyna Goździk, Aleksandra Kornacka, Justyna Bień, Władysław Cabaj, Bożena Moskwa

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*Neospora caninum* infection causes disease in canids and cattle, and is considered to be a major cause of bovine abortions worldwide. The wildlife population has been identified as a reservoir and transmitter of the parasite. In Poland, antibodies to *N. caninum* were found in three species of wild ruminants: European bison (*Bison bonasus bonasus L.*), red deer (*Cervus elaphus*) and fallow deer (*Dama dama*) (Cabaj *et al.*, 2005; Goździk *et al.*, 2010; Bień *et al.*, 2012). The wild ruminants probably become infected by ingesting food or water contaminated by *N. caninum* oocysts excreted by canids in the area, with the infection being maintained by vertical transmission.

The purpose of this study was to investigate the seroprevalence of *N. caninum* in moose (*Alces alces*). Blood samples were collected from 7 moose hunted in the north-east of Poland during 2011–2012.

The samples were tested for antibodies to *N. caninum* by agglutination test (NAT) (Romand *et al.*, 1998), the commercial competitive screening enzyme linked immunosorbent assay (cELISA) (VMRD, USA) and enzyme-linked immunoassay (ELISA) (IDEXX Laboratories Inc., Westbrook, ME, USA) with some modifications (Bień *et al.*, 2012). Sera that gave a positive result were further investigated by immunoblot analysis to verify presence of antibodies. SDS-polyacrylamide gel electrophoresis and immunoblot analysis were performed according to Björkman et al. (1994) and Cabaj *et al.* (2005) using Bio-Rad System. A serum was stated as positive when the percentage of inhibition (%I) was equal or greater than 30% (c-ELISA) and OD > 0.2 (ELISA) (Bień *et al.*, 2012).

Antibodies to *N. caninum* were detected only in one moose from the Ełk area. The antibody titer was confirmed by NAT (1:1280), cELISA (I=91%) and ELISA (OD=0.736). Immunoblot analysis revealed seropositivity against immunodominant *N. caninum* antigens of 120, 70, 55, 35, 25 and 17 kDa. The serological results of the present study indicate that the moose in Poland are exposed to *N. caninum* infection. To our knowledge, this is the first report on the seroprevalence in moose living in a natural environment in Poland.

# The expansion of nematodes *Ashworthius sidemi* in wild ruminants in Poland

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Ashworthius sidemi, a nematode belonging to the family Trichostrongylidae is a primary parasite of the Asian deer, particularly sika deer (*Cervus nippon*), through which it was introduced to the European part of Russia and Ukraine, as well as Slovakia, the Czech Republic and France. Migrating red deer carried this parasite from the neighboring countries to Polish territory.

Until now, in Poland, this parasite has been recorded in European bison, red deer, roe deer and fallow deer. A moratorium on hunting elk was imposed in Poland in 2001, so obtaining materials for parasitological studies from these animals was very difficult. However, the later authorization of the Minister of Environment for elk hunting enabled the parasitological examination of the digestive tract of this species. As a result of post-mortem examinations of 10 elk, *A. sidemi* were found in abomasa, for the first time in Poland, in 2 specimens from Augustów Forest and Biebrza Marshes. The intensity of the invasions was 120 and 7 specimens respectively.

This finding of *Ashworthius sidemi* in elk indicates a further expansion of the focus of ashworthiosis in Białowieża northwards into the Biebrza Marshes and Augustowska Forest. The growth of the elk population and their tendency for long distance migrations can contribute to the spread of parasitosis over much greater distances than when deer is the host.

A. sidemi is a typical parasite of cervids. The invasion occurs in not more than a few hundred nematodes, and does not seem to be pathogenic for them. However, in bison, which are the new hosts of this parasite, the invasion can reach a maximum intensity of tens of thousands of specimens. Such a high intensity of invasion of this blood-sucking nematode can cause inflammation of the gastrointestinal tract and chronic diarrhea, which, especially in young animals, may lead to cachexia, or even falls. However, although A. sidemi nematode infection has never been detected in domestic ruminants in previous studies, the possibility of infection of these animals grazing on pastures shared with wild ruminants cannot be ruled out. Therefore, regular monitoring of the status of parasitic invasions of both wild and domestic animals is necessary in the vicinity of previously identified foci of ashworthiosis.

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## Parasites of European beaver *Castor fiber* in north-eastern Poland

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In the period from April 2011 to November 2012, parasitological necropsies were performed of 48 European beavers from the Podlasie region. Helminths were isolated from the contents of the digestive tract of beavers, then they were fixed and determined to the species. In addition, blood samples and faeces from the beavers were examined. In total, 95.8% of the beavers were infected with parasites. A total number of six species of parasites were detected. *Stichorchis subtriqetrus* trematodes were found in 93.7% of the beavers. They were localized mainly in the cecum, less in the colon, and single juvenile trematode of this species were found in the small intestine. The intensity of the invasion ranged from a few to a few hundred specimens. In 68% of the beavers, 10 to a few thousand individual *Travassosius rufus* nematodes were present in the stomach. A few specimens of the nematode *Psilotrema castoris* was found in the small intestine of four beavers (8.3%). This is the first record of this species in Poland and the third of its discovery in the world.

Furthermore, in the small intestine of one beaver, 2 nematodes belonging to the species *Trichostrongylus capricola* were detected. Also, in one beaver pathological changes caused by the hydatid cestode *Echinococcus granulosus* were found to occur in the liver. In the examined beavers, inflammatory changes of the gastric mucosa caused by *Travassosius rufus* nematodes were also found and in cecum caused by trematode *Stichorchis subtriquertus*. These statements confirm our previous observations that these species are the most dangerous parasites of beavers in Poland. Besides the autopsy, coproscopic examinations using Baermann, flotation and decantation methods were performed of fecal samples collected from harvested beavers. Although all the results of the Baermann method were negative, the results of the flotation and decantation methods confirmed the necropsy findings. Also, using the flotation method, single oocysts of the coccidian *Eimeria sprehni* were detected in the faeces of one beaver. A Kingston and Morton blood test conducted did not reveal the presence of protozoa or microfilariae in the beavers.

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### The role of the gray heron (*Ardea cinerea*) in the spreading of parasitic nematodes

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The gray heron is widely distributed in the area of Europe, Asia and Africa. In Poland, the breeding grounds of herons are located near water bodies, usually together with cormorants or close to their breeding colonies. The grey heron feeds mainly on fish, but its diet also consists of amphibians, small mammals, insects, earthworms, molluscs and crustaceans. Parent birds feed nestlings with partly digested and regurgitated food. The infection with nematodes can occur very early, due to transmission of gastrointestinal-dwelling parasites from the infected adults. Such way of obtaining the nematodes of the genus *Contracaecum* was described for very young nestlings of cormorants (Kuiken et al. 1999, Kanarek 2011), and according to authors, it can cause high mortality among the young birds. No data is available on parasites of the grey heron nestlings.

Ten dead individuals of the young herons, collected under the nests in a breeding colony of cormorants near the Włocławek Reservoir were analyzed. Subsequent sections of the gastrointestinal tract (esophagus, stomach and intestine) were placed in separate crystallizers, sectioned and decanted in saline. Collected parasites were identified just after the decantation or in the material preserved in 70% ethanol supplemented with glycerol (5%). The prevalence of infection in the birds was 100%. Larvae of nematodes of the genus *Contracaecum* and *Porrocaecum* were detected only in the stomach of the young herons. Three individuals were infected both with the larvae and the adult nematodes of the genus *Contracaecum*. Mean intensity of infection with nematodes of *Contracaecum* spp and *Porrocaecum* spp. was  $9.3 \pm 5.2$  and  $3.1 \pm 2.03$  in the respective genera. Nematodes were not detected in the intestine, nor in the esophagus of the studied herons.

#### Parasites in the protected areas of the Tatra Mountains, Slovakia

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The northern part of Slovakia represents the main tourist and recreational area of the country. After the windstorm in 2004 that caused losses or changeovers of prime biotopes, the migration of existing wildlife is expected, as is the immigration of replacements, followed by growth of carnivorous predator populations. which represents a risk of spreading parasitic infections.

The aim of our work was to study the parasite circulation in ecosystems that undergo dynamic changeovers. The distribution of zoonotic agents in carnivore hosts was analysed, emphasizing the most important ones – echinococcosis and trichinellosis. The small intestines of carnivores were examined for the presence of *E. multilocularis* using a modified sedimentation and counting technique (Raoul et al., 2001). Muscle samples were examined for the presence of *Trichinella* spp. larvae by the artificial HCl-pepsin digestion method (Kapel and Gamble, 2000). Fecal samples were investigated using the coprological flotation method.

The presented research work reveals the presence of the *Echinococcus multilocularis* tapeworm, which causes severe alveolar echinococcosis in humans, in almost 40% of foxes from TANAP and adjacent areas. The parasite was also present in domestic cats, raccoon dogs and martens. Trichinellosis was harboured in over 19% of foxes, and was also detected in wolves, brown bears, lynxes, domestic cats and also in large proportion of predators from the family Mustelidae (19.4%). In 2012, a pilot study was initiated on the parasite fauna of the endemic sub-species alpine marmot (*Marmota marmota latirostris*). An examination of 30 fecal samples revealed the presence of *Eimeria* spp. oocysts (16.7%) and eggs of the *Ctenotaenia marmotae* tapeworm (53.3%). A long-term examination of carnivores from the protected areas of TANAP revealed that these wild predators play a significant role in environmental contamination with parasites and may contribute significantly to the spread of parasitic zoonoses. They might also play important roles in the structuring of parasite communities that provide opportunities for increased exposure of humans to parasitic infection.

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#### Occurrence of the third-stage larvae of *Aelurostrongylus abstrusus* (Nematoda: Metastrongyloidea) in slugs in Poland

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Aelurostrongylus abstrusus (Railliet, 1898) is a metastrongylide nematode parasite which infects the lungs of domestic cats and wild felids. The life cycle of *A. abstrusus* involves two hosts. In the cardio-respiratory system of the definitive host, the oviparous females produce eggs with first-stage larvae (L1s). The first-stage larvae (L1) are coughed up, swallowed and excreted to the environment in the host's faeces. The intermediate hosts are molluscan (snails or slugs), infected through penetration of the epidermis or ingestion of first stage larvae. After entering the intermediate host, L1s develop into third-stage larvae (L3) and encyst mainly in the intestinal ligament. Paratenic hosts (mice, frogs, toads, snakes, lizards, ducklings, chickens) become infected by the ingestion of infected snails or slugs. The definitive host is infected by the ingestion of the infected mollusc or the paratenic hosts.

Twenty-four specimens of *Arion lusitanicus* were collected by hand in the area of Pruszcz Gdański in September and November 2012. During examination of the body cavity and internal organs of *A. lusitanicus*, cysts were observed on the surface of the intestine. The cysts contained third stage larvae of *A. abstrusus*. Morphological and morphometric analyses were supported by molecular methods.

This study reports for first time the presence of *A. abstrusus* infective larvae in the invasive synantropic slug *Arion lusitanicus* (intermediate host), evidencing the veterinary importance of this mollusc in the transmission of *A. abstrusus*.

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#### Stomach parasites of wild boars in the Lower Silesia district

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Wild boars feed on a varied diet consisting of plants and animals. As omnivorous scavengers, they eat almost anything they come across during foraging, including grass, nuts, berries, carrion, the nests of ground nesting birds, roots, tubers, insects and small reptiles or rodents. Wild boars inhabit nutrient-rich, deciduous and mixed forests nearby agricultural land, so they are an important link in the circulation of parasites between the forest environment (sylvatic) and the farm (livestock). The animals, that wild boar feed on are in many cases the intermediate hosts of numerous parasites. Dung beetles commonly found in the woods (e.g Anoplotrupes stercorosus or Trypocopris vernalis) are the intermediate hosts of parasitic stomach nematodes belonging to the Spiruroidea superfamily, represented by Ascarops strongylina and Physocephalus sexalatus, found in feral pigs in Lower Silesia in the period from November 2011 to December 2012. Tests for confirmation of the stomach parasites were conducted on 64 wild boars of both sexes aged from 8 months to 6 years, which were shot during hunts in the Zlotowek Forest Inspectorate (Lower Silesia). The stomachs of the wild boars were collected immediately after the animals were killed, packed in refrigerated containers and transported to the Division of Parasitology, where necroscopy of the internal organs was conducted. During the study, the parasites were found in the gastric mucosa in 20 cases. The parasites were the most frequently observed in the area of the fundus along the greater curvature. The morphological studies confirmed the existence of two species of parasites: Ascarops strongylina and Physocephalus sexalatus. No examples of mixed infection were found. Rectal faecal samples collected from killed animals confirmed the presence of parasite eggs stated at necroscopy. The intensity of the infection in affected individuals was varied and ranged from 1 to 28 parasites in the lumen of the stomach. Previous studies indicate that stomach infestations in wild boars are common in Europe (Fernandez-de-Mera et al. 2004, Humbert and Henry 1989). Pathological changes caused by the parasites are varied and depend on the degree of intensity of the invasion. In the case of extensive infection, frequent effects are ulceration, gastritis and digestive disorders. Clear inflammatory lesions of the gastric mucosa with local reddening were diagnosed only in two cases during the studies. Monitoring the presence of parasites in wild boars is justified, as they increasingly inhabit surroundings of farms and can be a potential source of parasites for livestock.

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# The prevalence of gastrointestinal parasites in raccoons (*Procyon lotor*) in the Lubuskie province (Poland) – with particular regard to *Baylisascaris procyonis*

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The aim of the study was to estimate the prevalence of gastrointestinal parasites in raccoons with particular regard to zoonotic parasites.

The gastrointestinal tracts (stomachs and intestines) from raccoons which were hunted or killed on the road were examined. Overall 56 raccoons from Lubuskie province (Poland) were examined. The small and large intestines were collected from all raccoons while the stomachs were only recovered from 34 specimens. The samples were examined with the use of sedimentation and counting technique (SCT) – the intestines and stomachs were examined separately. Samples of raccoon faeces were collected from the environment: defecation sites, latrine sites (localization: commune: Górzyca, district: Słubicki, province: Lubuskie). Samples were collected monthly in 2012 (January, February, March, April, May, June, September, October, November, December). Overall, 154 samples were obtained and examined with the use of the McMaster flotation technique, modified by Raynaud.

The following parasites were detected in the intestinal and stomach contents of raccoons: tapeworms *Mesocestoides* spp. (60% in intestines and 27% in stomach), Echinostomatidae flukes (32% and 16%) and nematodes *Capillaria* spp. (21% and 6%). Moreover, Acanthocephala were found in the intestines of 2 raccoons. The highest intensity of infection was observed in the case of *Mesocestoides* spp. The examination of faecal samples from the environment showed the following results: the most significant concerning zoonotic parasites being the detection of *Baylisascaris procyonis* eggs (mean 60 EPG). The eggs of this parasite were found in 3 samples of faeces collected in November and December, 2012. Furthermore, in some samples of faeces, the eggs of flukes, *Capillaria* spp., *Mesocystoides* spp. and coccidian oocysts were found. It is one of rare reports concerning *Baylisascaris procyonis* in Poland confirming the presence of this dangerous parasite in the Polish raccoon population.

The study was conducted as a part of grant No. NN 308 63193.

## Anisakidae of beluga (*Delphinapterus leucas*) from the Hudson Bay

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Beluga (*Delphinapterus leucas* Pallas, 1776) is a marine mammal living in the Arctic and subarctic waters. This species feeds mainly on fish, krill and other crustaceans (Copepoda, Amphipoda, Decapoda). Beluga is the final host for nematodes of the Anisakidae family. Anisakidae are parasites of marine organisms and are found in many geographical areas. The first intermediate hosts for Anisakidae are crustaceans, the second are fish, and the parasites reach sexual maturity in birds and marine mammals.

In our study 190 nematodes were collected from stomachs of 11 belugas harvested by northern Quebec Inuit. The material came from the Arctic regions of the Hudson Bay and Hudson Strait, northern Quebec. All belugas were infected, with an intensity of infection of 1–52 specimens. Morphologically, we identified three species: *Pseudoterranova decipiens* s. l., *Anisakis simplex* s. l. and *Contracaecum osculatum* s. l. Molecular analysis allowed us to detect sibling species. Using the PCR-RFLP method (restriction enzymes RsaI, TaqI, HhaI and XbaI), we identified four species: *Pseudoterranova bulbosa*, *Anisakis simplex* s. s. and *Contracaecum osculatum* C and A. We noted three developmental stages: L3 (159 individuals), L4 (16 individuals) and adults (15 individuals – 11 males and 4 females).

Beluga is predominantly the definitive host for. *P. decipiens* s. l. (prevalence 72%), *C. osculatum* s. l. (45.5%) and, to a lesser extent, *A. simplex* s. l. (36%).

## Strongylides (Nematoda; Strongylidae) of equids from the Askania Nova Biosphere Reserve, Ukraine

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Wild and domestic horses (Equidae) have been kept in the Askania-Nova Biosphere Reserve for more than 100 years. Nowadays, six species of equids – wild Przewalski's horses (*Equus ferus przewalskii*), Turkmenian kulans (*E. hemionus*), donkeys (*E. asinus*), Shetland ponies (*E. caballus*), plains zebras (*E. burchelli*) and Grevy's zebras (*E. grevyi*) are kept in the reserve. The aim of our work was to examine biodiversity and structure of the community of strongylid nematodes in six equid species from the Askania-Nova Biosphere Reserve by *in vivo* method.

Ninety equids (24 Przewalski's horses, 13 kulans, 16 donkeys, 14 ponies and 23 zebras) of various ages were included into the study. All animals were treated with macrocyclic lactone drug "Univerm" (0.2% aversectin C, Russia). Fecal samples (200 g each) were collected from each animal 24, 36, 48 and 60 hours after treatment. All expelled strongylid (62 418 specimens) were collected and identified.

In total 36 species of strongylids (9 species of subfamily Strongylinae and 25 of Cyethostominae) were found in six equid species. In the Przewalski's horses 31 strongylid species were collected, from 9 to 18 species (average  $14.5\pm2.5$ ) parasitizing one horse. In kulans 25 strongylid species were found, from 7 to 19 species (average  $14\pm3.6$ ) per one kulan. In donkeys 26 strongylid species were observed, from 6 to 16 species (average  $12.7\pm2.4$ ) per one donkey. In Shetland ponies 28 strongylid species were detected, from 8 to 24 species (average  $14.9\pm4.0$ ) parasitizing one host. In plains zebras 21 strongylid species were found, from 4 to 14 species (average  $8.5\pm3.2$ ) per one host. In Grevy's zebras 18 strongylid species were found, from 4 to 14 species (average  $9.0\pm3.3$ ) parasitizing one host. The general structure of the strongylid community of all equids, with the exeption of Grevy's zebras, was multimodal with dominant, subdominant, background and rare species. Grevy's zebras had bimodal structure of strongylid community ("core – satellite" mode), which is typical for animals that undergo frequent deworming programs. Bray–Curtis Cluster analysis revealed similarity of strongylid communities in both zebra species and donkeys, as well as similarity between strongylid community of Turkmenian kulans and Shetland ponies. Our current data confirm stability of ecological and parasitological conditions in the Askania-Nova Biosphere Reserve during last 40 years.

## Helminthfauna of the common noctule *Nyctalus noctula* (Schreber, 1774) from the Wrocław area

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The study aimed to supplement and update the knowledge of the species composition of endoparasites of the common noctule (*Nyctalus noctula* Schreber, 1774) from the Wrocław area. The study was performed on helminths isolated from 80 individuals of *N. noctula* from an urbanized area of the city center. Helminths from the following groups were isolated: Digenea (Plagiorchiidae and Lecithodendriidae), Nematoda (Molineidae and Physalopteridae) and Cestoda (Hymenolepididae). A total of 10,240 parasites were found. Overall prevalence was 96.25%, with the average intensity of infection being 132.9. Three species of trematodes were detected: *Plagiorchis vespertilionis* (78.75%), *Prosthodendrium chilostomum* (80%), *Lecithodendrium spathulatum* (68.75%), one species of tapeworm *Vampirolepis skrjabinariana* (20%) and 2 species of nematodes: *Molinostrongylus skrjabini* (63.75%) and *Physaloptera myotis* (5%) – a new species for *N. noctula* in Poland. To analyse the helminth population and the structure of the helminth community qualitative and quantitative parasitological and ecological parameters were used.

## The parasite fauna of buzzards (*Buteo* spp.) in the territory of Slovakia – the results of an initial study

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Three raptor species from the genus *Buteo* occur in Slovakia – the common buzzard (*Buteo buteo*), roughlegged buzzard (*Buteo lagopus*) and long-legged buzzard (*Buteo rufinus*). The common buzzard is the most common raptor not only in Slovakia but also in Europe. The main diet component of these birds comprises small rodents, especially *Microtus arvalis*.

Our pilot study was focused on the detection of endoparasites in buzzards from Slovakia. We examined 53 common buzzards, 1 rough-legged buzzard and 1 long-legged buzzard. The raptors were subjected to parasitological dissection, the samples of pectoral muscles were examined for trichinellosis using an artificial digestion method and the fecal samples were examined using the standard flotation technique for the presence of parasite eggs and coccidian oocysts.

Parasitological dissection revealed 30% of birds to be infected with parasites from the class Trematoda, in 40% were found specimens of Cestoda, in 6% of individuals acanthocephalan parasites were found. The most common finding was nematode infection (64%).

Some of the identified tapeworms were identified as *Cladotaenia globifera*. In the following study, we will focus on identification of other parasite species.

Coprological examination detected 42.4% prevalence of *Capillaria* spp. Eggs of the genus *Cyathostoma* were found in one long-legged buzzard and intestinal coccidian oocysts were present in 15.2% of samples. Although the raptors are potential hosts of the *Trichinella pseudospiralis* parasite, no positive specimen was found within the present study. These are the first results of an initial study, which indicate that parasite fauna of raptors from the genus *Buteo* is quite diverse. The research is continuing and will contribute to the knowledge of the parasites of Falconiformes and Strigiformes.

The work was supported by a grant of the State Agency VEGA No. 1/0702/12.

#### A survey of the endoparasites of corvids in Slovakia and a new record of *Spiniglans sharpiloi* tapeworm in the Eurasian magpie (*Pica pica*)

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The Corvidae (order Passeriformes) belong to the most developed avian group. In Slovakia, they inhabit opened biotopes, in forests, lowlands and in mountains. Animal feed is the main component of their diet, especially during nesting, and particularly in juveniles. Later, in the late summer until winter this is replaced by crops. Within the present study, we investigated the occurrence of endoparasites in 33 corvids belonging to 4 species – 15 common ravens (Corvus corax), 11 rooks (Corvus frugilegus), 1 carrion crow (Corvus corone cornix), and 6 Eurasian magpies (Pica pica) found dead in the territory of Slovakia. All specimens were subjected to helminthological autopsy and presence of parasites was determined. A standard flotation technique was used for examination of faeces for the presence of parasite eggs and coccidian oocysts. An artificial digestion method was used for detection of Trichinella in pectoral muscle samples. The parasitological autopsy revealed the presence of gastro-intestinal helminths in only one common raven, in small intestine of which was detected one Cestoda specimen and two nematodes. Helminths were neither present in other common ravens nor in the examined rooks. The single carrion crow harboured cestodes in the small intestine. The Eurasian magpie was the most infected species with all six investigated individuals being infected with tapeworms and one of them also with Nematoda species. The tapeworms were identified as Passerilepis stylosa (Rudolphi, 1809), Passerilepis crenata (Gocze, 1782) from the family Hymenolepididae. In one of examined magpies, the presence of the Spiniglans sharpiloi tapeworm from the family Dilepididae described recently by Kornyushin et al. (2009) was identified. Herein we record the first finding of this parasite species in Slovakia. By the use of coprological methods, eight individual birds were found to contain coccidian oocysts which, due to previous freezing of samples, could not be identified to the species level. In two birds the sporadic occurrence of *Capillaria* spp. eggs was recorded, and in one magpie, sporocysts of Sarcocystis spp. were diagnosed. This finding is relatively rare, as birds from the family Corvidae were only recently affirmed as potential definite hosts for Sarcocystis genus (Gjerde and Dahlgren, 2010). Considering the feeding habits (especially scavenging) of corvids, using an artificial digestion method, the pectoral muscles of all birds were examined for the presence of Trichinella larvae. Despite the fact that this avian group may become a potential host for Trichinella pseudospiralis, no infected specimens were found.

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## *Neospora caninum* antibodies in free-ranging carnivores from Poland: preliminary studies

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*Neospora caninum* is an obligate intracellular protozoan parasite that can infect domestic and wild canids, ruminants and horses. Abortion and stillbirth due to neosporosis, especially in dairy cattle, have been reported worldwide. Domestic dogs, coyotes and wolves are definite hosts for the parasites. Many aspects of the life cycle of *N. caninum* are still unknown and the role of wildlife in the life cycle of *N. caninum* is still uncertain.

The study was performed on the meat juice from 344 wild animals from different regions of Poland. A commercial competitive ELISA (cELISA) from VMRD (USA) was used for detection of anti-*N. caninum* antibodies in wild carnivores according to the manufacturers' instructions. When the percentage of inhibition (%I) was equal to or greater than 30% (cELISA), the sample was regarded as positive.

The overall seroprevalence of *N. caninum* antibodies in examined samples was found to be 20.3%. Antibodies to *N. caninum* were found in 22.4% of 254 red foxes (*Vulpes vulpes*); in 12.5% of 8 Eurasian badgers (*Meles meles*); in 23.1% of 13 pine martens (*Martes martes*); in 25.0% of 12 raccoon dogs (*Nyctereutes procyonoides*); in 6.45% of 31 raccoons (*Procyon lotor*) and 15.3% of 13 American minks (*Neovison vison*). No antibodies were detected in 2 Eurasian otters (*Lutra lutra*) and 11 muskrats (*Ondatra zibethicus*). To our knowledge, this is the first report of antibodies to *N. caninum* in free ranging wild carnivores in Poland.

Care must be taken when comparing these results to studies based on a single test only. Further studies are necessary to confirm the existence of such sylvatic cycle in Poland. The existence of a possible sylvatic cycle could have important implications in both sylvatic and domestic cycles since they might influence the prevalence of infection.

This study was supported by the Polish-Slovak Joint Research Project for the years 2013-2015: Observation of serious protozooses (neosporosis and toxoplasmosis) in domestic and sylvatic cycle

## The occurrence of *Nematodirus* spp. in wild and domestic ruminants from southern Poland

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Infection with *Nematodirus* spp. can cause serious economic losses in ruminant livestock production, although particular species of the parasite may differ in their pathogenic effect on the host.

The aim of the study was to recognize the species composition of *Nematodirus*, and their infection level in domestic ruminants, as well as the potential role of wild cervids in the transmission of these nematodes. During the years 2009–2012, the small intestines of 103 animals (20 sheep, 33 cattle, 39 roe deer, six red and five fallow deer), slaughtered or hunted in southern Poland, were dissected. Altogether, three species of *Nematodirus* were noted, with the prevalence of 19.4%. *Nematodirus battus* was recorded in 15.0% of sheep examined, *N. filicollis* occurred in 10.0% of the sheep and in one of five fallow deer studied, whereas *N. europaeus* was found in 20.5% of roe deer. None of the examined red deer or cattle were infected.

It appears that the wild cervids play a minor role in the transmission of *Nematodirus* sp. Special attention should be paid to the confirmation of the occurrence of the highly pathogenic species – *Nematodirus battus,* recently reported sporadically only in coproscopic examinations. The presence of this species merely in livestock (sheep) may suggest its introduction with animals imported for breeding.

# Helminths of Northern fur seals (*Callorhinus ursinus* L., 1758) on St. Paul Island, Alaska, USA

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Northern fur seals (NFS) (*Callorhinus ursinus* L., 1758) are parasitized by more than 20 species of helminths of four groups – Nematoda, Cestoda, Trematoda and Acanthocephala. The studies performed during July–August 2011 and 2012 were aimed to examine the abundance and biodiversity of helminths parasitizing the Northern fur seals on St. Paul Island, Alaska. In total, 502 NFS males (3–4 years old) were examined during the annual Aleut subsistence harvest. Gastro-intestinal tracts were collected from 406 NFSs. All helminths collected from skins and gastro-intestinal tract (in total, 3,827 specimens of nematodes, 6,183 cestodes, 578 trematodes, 483 acanthocephalans) were fixed in 70% ethanol and identified by their morphology.

All NFSs examined were infected with gastrointestinal helminths. The prevalence of NFS infection by gastric nematodes was 71.9%; intensity –  $10.5\pm14.2$ . Four species of three genera – *Anisakis (A. simplex), Contracaecum (C. osculatum)* and *Pseudoterranova (P. decipiens, P. azarazi)* were found. Prevalence of NFS infection with cestodes was 98.3%; intensity –  $15.7\pm13.9$ . *Diphyllobothrium pacificum* was the dominant species (prevalence=97%); *Diplogonoporus violettae* was found in 6.6% NFSs. The prevalence of NFS infection with acanthocephalans was 39.4%; intensity –  $3.1\pm3.2$ . Seven species of genera *Corynosoma (C. strumosum, C. alaskensis, C. semerme, C. similis, C. validum, C. villosum)*, and *Bolbosoma (B. nipponicum)* were found. Trematodes were found in 36.2% of the NFS examined; *Phocitrema fusiforme* was the dominant species (prevalence=78%), *Pricitrema zalophi* and *Stictodora* spp. were observed in 5.5% and 6.4% of NSF infected respectively. The filarial nematode *Acanthocheilonema odendhali* was found in 18% of NFSs with an average intensity of  $1.32\pm0.83$ .

A comparison of current data with that of previous researchers reveals changes in the gastrointestinal parasite community structure in NFSs during the last decades which, in our opinion, are connected with changes in NFS diet. Further studies are necessary to confirm the relationship between parasite community structures and changes of NFS foraging behavior.

#### Lyperosomum sp. from hedgehog Erinaceus concolor

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According to Panin (1984), the genus *Lyperosomum* (Looss, 1899) contains 33 species described mainly from birds. Only two species of *Lyperosomum* i.e. *L. soricis* and *L. transcarpathicus* was recorded from insectivores as *Sorex* spp. and *Crocidura* spp. Twenty-five immature dicrocoeliid flukes were recovered from the gall-bladder of a hedgehog *Erinaceus concolor* killed in a road accident in the Pruszków area. Identification based on morphology was not possible due to the lack of developed gonads. Only molecular analysis gave a chance to sign parasites belonging. For two specimens, the partial 28S rDNA gene (1200bp fragments) was sequenced. The obtained sequence was compared with two (only two records of *Lyperosomum* 28S rDNA are available) sequences from *L. collurionis* and *L. transcarpathicus* deposited in GeneBank under the respective accession numbers AY222143.1 and AF151943.1. The 28S rDNA sequences from the hedgehog and sequence of *L. collurionis* were identical. Flukes found in the hedgehog probably belong to the *L. collurionis* or a closely related species. This is the first record of a "bird *Lyperosomum* species" isolated from a hedgehog.

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#### Identification of tetraphyllidean larvae from *Notothenia rossii* to *Onchobothrium antarcticum* Wojciechowska, 1990 by molecular analysis (Admiralty Bay, King George Island, Antarctica)

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Cestodes of the order Tetraphyllidea are parasites of chondrichthyans. Tetraphyllidean larvae have been found by many authors, in Antarctic and Sub-Antarctic bony fishes (Rocka 2003). The first report on the occurrence of adult cestodes parasitizing the Antarctic shark, given by Linstow (1907), described a new species, Phyllobothrium dentatum (Tetraphyllidea). Studies on parasites of the Antarctic skates began in 1990 when the first tetraphyllidean species, Onchobothrium antarcticum Wojciechowska, 1990 from Bathyraja eatonii (Günther, 1876) was described. This cestode has trilocular bothridia armed with a pair of hooks situated on the anterior margin of the anterior loculus (Wojciechowska 1990). Antarctic bony fishes (intermediate hosts for Tetraphyllidea) are infected with 5 forms of cestode larvae (cercoids). The larvae of the Tetraphyllidea differ from each other with morphology of scolices: there are larvae with bothridia subdivided into one, two and three loculi, some bothridia are subcylindrical in shape and other bothridia are undivided with hook-like projections. Only one species of the family Onchobothriidae, Onchobothrium antarcticum, has been described in the Antarctic, and cercoid with trilocular bothridia was assigned to this cestode species (Wojciechowska 1990). Ten cercoid larvae, obtained from Notothenia rossii, and three adult specimens of Onchobothrium antarcticum isolated from Bathyraja eatonii were examined. A partial sequence of cytochrome c oxidase subunit I of three adult specimens and four cercoids were identical. This result confirms the taxonomic affiliation of the cercoids with trilocular bothridia to Onchobothrium antarcticum.

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#### Temporal and spatial distributions of fish parasites from Oder estuary (Poland)

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The first parasitological studies of fish from the Pomeranian Bay and Polish waters of the Oder River estuary were carried out by Kozikowska (1957). In the subsequent years (until 2012) 2453 fishes of 17 species from the Pomeranian Bay and the Szczecin Lagoon were examined. Parasitological examination focused on: the skin, oral and nasal cavities, eye lens, vitreous humor, gills, heart, gonads, liver, spleen, kidney, gallbladder, gastrointestinal tract, swim bladder, peritoneum and blood.

The parasites component communities comprised 73 species belonging to: Protozoa, Myxosporea, Ciliata, Fungi, Monogenea, Digenea, Cestoda, Nematoda, Acanthocephala, Arthropoda, Hirudinea and Coccidia. The dominant parasites were the digeneans of the following genus/species: *Sphaerostomum globiporum* (Rudolphi, 1802) Szidat, 1944; *Apophallus donicus* (Skrjabin et Lindtrop, 1919) Price, 1931; *Apophallus muehlingi* (Jägerskiöld, 1899) Luhe, 1909; *Bucephalus polymorphus* Baer, 1827; *Tylodelphys clavata* (von Nordamann, 1831) Diesing, 1850; *Bunodera luciopercae* (Műller, 1776) Stiles et Hassal, 1898; *Azygia lucii* (Műller, 1776) Lühe, 1909; *Crepidostomum farionis* (Műller, 1784) Lühe, 1909; *Diplostomum* spp.; *Deropristis inflata* (Molin, 1859) Odhner, 1902; *Ichthyocotylurus variegatus* (Creplin, 1823); *Ichthyocotylurus platycephalus* (Creplin, 1825); *Nicolla skrjabini* (Ivanitzky, 1928) Dollfus, 1959; *Brachyphallus crenatus* (Rudolphi, 1802) Odhner, 1905.

The digeneans Apophallus donicus and Apophallus muchlingi were the most common (prevalence 50–100%) in sander, perch, vimba, bleak, rudd, white bream and ruffe. Brachyphallus crenatus and Azygia lucii in flounder were rarely recorded (prevalence 0.8%). The smallest number of representatives of the parasites was found in: fungus-like organisms – 1 species (Glugea acerinae Jírovec, 1930 from ruffe), Monogenea – 1 species (Dactylogyrus amphibothrium Wagener, 1857 from ruffe), leeches – 1 species (Piscicola geometra Linneus, 1761 from asp), and Coccidia – 1 species (Eimeria anguillae Leger & Hollande, 1922 from eel). Among all of the examined fishes the eel has been studied most thoroughly. The prevalence ranged from 1.9% (Ergasilus gibbus von Nordmann, 1832 and Argulus foliaceus Linnaeus, 1758) to 88.7% (Myxidium giardi Cépède, 1906).

# Seroprevalence of *Toxoplasma gondii* in wild birds in north-western Poland

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Data concerning *Toxoplasma gondii* infection in wild birds in Poland are sparse. In the present study, serum samples from 55 birds of 7 species from north-western Poland were examined for the presence of *T. gondi*-specific IgG antibodies using a direct agglutination test. Antibodies to *T. gondii* were found in 21 (38.2%) of the 55 birds, including 4 of 5 common pochards (*Aythya ferina*), 2 of 10 tufted ducks (*A. fuligula*), 5 of 15 greater scaups (*A. marila*), 4 of 15 common coots (*Fulica atra*), 5 of 6 velvet scoters (*Melanitta fusca*), and 1 of 3 common scoters (*M. nigra*). These results indicate that water ecosystems are contaminated with *T. gondii* oocysts.

## Seroprevalence of *Toxoplasma gondii* in cervids farmed in north-western Poland

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*Toxoplasma gondii* is one of the most common parasites of humans and animals, but there is little information about its occurrence in farmed cervids. The aim of this study was to determine the seroprevalence of *T. gondii* in red deer (*Cervus elaphus*) and fallow deer (*Dama dama*) bred in a farm located in north-western Poland. A total of 313 blood samples were collected from red deer and fallow deer during 2011–2012. The samples were screened for *T. gondii*-specific antibodies using a direct agglutination test according to the manufacturer's instructions. Negative and positive controls were included in each test. Sera with a titre of 1:40 and higher were considered positive. The seroprevalence of *T. gondii* was higher in red deer than in fallow deer. The high prevalence of *T. gondii* antibodies found in the present study indicates that meat from farmed cervids, particularly red deer, should be regarded as a potential source of infection for humans. Moreover, the seroprevalence data in these grazing animals reflected environmental contamination with *T. gondii* oocysts; therefore preventive measures should be taken to prevent contamination of pastures, hay, and surface water with feces of cats.

#### Intestinal parasites in soil samples from latrines discovered during archaeological excavations in Kożuchów, Poland

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Archaeoparasitology is a relatively new and interdisciplinary branch of knowledge. The studies have provided important information on human and animal parasitic infections as well as on sanitary conditions, and social and dietary behaviours of hosts in the past. The aim of our study was to examine latrine soil samples for the presence of parasites. From an archaeological site (Kożuchów), 22 samples were collected from three latrines dating from the 13th to the 16th centuries. The rehydrated and fixed samples were examined using coprological and immunological methods. *Ascaris* and *Trichuris* eggs and *Cryptosporidium* oocysts were identified in soil samples derived from all the latrines, whereas *Taenia* eggs were found only in samples originating from the 15th century latrine.

## Intensifying synanthropisation of animals and dispersion of zoonotic parasites

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Synanthropisation (gr. syn – together, anthropos – man), that is *living together with man*, is the phenomenon which increases with the increasing number of people in the world: in the last century, this number has increased four times. The demographic explosion, and the human pressures on the environment associated with it, have resulted in the natural areas of animals shrinking and the boundary between the natural and anthropogenic environment blurring. Consequently, some animals overcome their fear and penetrate into the sub-optimal conditions that prevail in the vicinity of man.

Out of many species of animals that tend to synanthropisation, in Europe the most important for the transmission of parasites to humans are the red fox, wild boar, raccoon (an invasive, alien species) and some species of birds. They dispersed faeces throughout the environment, often containing eggs and/or oocyst of zoonotic parasites (for example: *Toxocara* spp., *Echinococcus multilocularis, Ascaris* sp. *Trichuris* sp., *Baylisascaris procyonis, Cryptosporidium* sp., *Giardia* sp.). While faeces in soil and water are decomposed in a short time the infective form of parasites can survive there for several months or years. They are potential sources of zoonotic infections which are dangerous for people (toxocarosis, baylisascariosis, echinococcosis, ascariosis, cryptosporidiosis, giardiosis). Although nowadays, it is difficult to determine the role played by synanthropic and domesticated animals in the biological contamination of the environment (for example dog, red fox and wolf can be infected with the same species of parasites) the development of molecular examinations (genotyping) provides promising results for obtaining an answer.

Sustained migration of people from rural to urban areas and changes of human behaviour (for example, limitation of hunting) intensify the process of synurbanisation i.e. penetration of urban areas by wild animals. There, we can expect that the problem of transmission of zoonosis from wild animals to people will continue to increase due to the tendency to expand the green areas of cities, such as parks, squares, open recreation and sports fields. This expansion favours synanthropisation and exposure to infection by spending more time in the open areas.

## Ashworthius sidemi: a new blood-sucking abomasal nematode of cattle

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Ashworthius sidemi Schultz, 1933 is a blood-sucking abomasal nematode considered to be as pathogenic as *Haemonchus* spp. It is mainly a parasite of sika deer, which was introduced into many countries of the former Soviet Union, Slovakia and the Czech Republic. The first *A. sidemi* found in Poland was reported in 1997 in European bison from the Bieszczady Mountains (Dróżdż *et al.*, 1998). After that, this nematode infection was observed in red and roe deer in the Bieszczady region, and in European bison from the Białowieża Primeval Forest (Dróżdż *et al.*, 2002). In 2012 *A. sidemi* was found in fallow deer in an enclosure located in the Dulowa Primeval Forest (Kowal *et al.*, 2012) and in elk (in press).

Based on very high prevalence of A. sidemi in wildlife, Dróżdż et al. (2002) suggested that nematode should also be able to infect cattle and sheep fed on the same meadows or pastures. The main interest of the study was to confirm this hypothesis. Faecal samples were collected from cattle grazing on the same pastures with the wildlife in the Białowieża Primeval Forest buffer zone and Strzałowo Forest District (Mazurian Lake area). Approximatelly, 200 faecal samples were placed in Petri dishes and incubated at 25°C for 7–14 days. DNA of L3 infected larvae was identified to the A. sidemi level according to Ljunggren and Goździk (2008). The amplified genomic DNA samples were analysed on 2% agarose gels stained with GelRed in TAE buffer, at 100 V. The gels were visualised using the Kodak 1D<sup>TM</sup> Electrophoretic Documentation and Analysis System. A segment of approximately 406 bp was excised from agarose gel and purified using the NucleoSpin Extract II (Macherey-Nagel, Germany). DNA was sequenced in the forward and backward directions by Genomed (Warsaw, Poland). The sequence was edited using Vector NTI Advance<sup>TM</sup>, version 10 (Invitrogen, USA). BLAST searches were performed in order to compare the sequence with those in GenBank. DNA of L3 A. sidemi larvae was found in cattle faecal samples in four out of eleven examined herds. The obtained sequences matched the sequences already published in GenBank, showing very high similarities with the isolate from European bison (accession number EF467325). To the best of our knowledge, this is the first report on A. sidemi in cattle. There is a need for further monitoring of the current spread of A. sidemi in wildlife and domestic animals.

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## *Trichinella spiralis* in road-killed raccoon dogs (*Nyctereutes procyonoides*)

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Trichinellosis is still one of the most important food-borne parasitic zoonoses in Poland. The epidemiological studies in Poland revealed sympatric occurrence of three *Trichinella* species: *T. spiralis*, *T. britovi*, and *T. pseudospiralis*. They are characterised by completely different circulation observed in natural environment (sylvatic cycle) and synanthropic environment (domestic cycle), and by different hosts. It is the main reason why trichinellosis is still to be considered as a threat to public health.

The aim of the study was to determine *Trichinella* prevalence in the population of wild raccoon dogs (*Nyctereutes procyonoides*). In 2012 muscle samples were collected at irregular intervals from carcasses of 39 raccoon dogs that had been killed accidentally by automobiles on roads of the Warta Mouth National Park between the towns Kostrzyn and Słońsk.

For isolation of *Trichinella* muscle larvae (TML), the muscle samples of about 200g were digested by HCl-pepsin solution. Of all raccoon dogs examined in the study two were infected with *Trichinella*. The TML were identified at species level by isolation of genomic DNA using EURyx kit, and by genomic PCR with primers: oTSR1 and oTSR4 (according to the conditions described in 1999 by Appleyard et al., Journal of Parasitology, 85: 556-559).

#### New records of nematodes imported by migratory passerines

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According to the literature data, 125 nematode species have been recorded from the birds nesting in Poland; 44 (35.5%) of these species have been found in Passeriformes. It has been reported that 36 species of passerines (21.7% of the Polish fauna) were infected with nematodes (Okulewicz 1997; Pojmańska *et al.* 2007; Okulewicz A. and Okulewicz J. 2008).

The author's helminthological collection of Passeriformes, obtained during a comprehensive study of the parasite fauna of birds of Lower Silesia conducted since the 1970s, contained unidentified alcohol-preserved and glycerol-cleared nematodes. The nematodes were collected from fairly rare three species of birds: Red-backed Shrike *Lanius collurio* (2 infected), Eurasian Golden Oriole *Oriolus oriolus* (1 infected) and Yellow Wagtail *Motacilla flava* (9 infected). All the birds were adult, obtained in spring months (April-May).

Viguiera euryoptera (Habronematidae) was found in the glandular stomach of Lanius collurio and Microtetrameres inermis (Tetrameridae) in the glandular stomach of Oriolus oriolus. In Motacilla flava birds, Acuaria subula (Acuariidae) was found under the cuticle of gizzard, Diplotriaena ozouxi (Diplotriaenidae) in air sacs and Microtetrameres inermis in the glandular stomach.

All the recorded nematode species (*V. euryoptera*, *A. subula*, *D. ozouxi*, *M. inermis*) represent the order Spirurida and are specific to passerine migratory birds. *V. euryoptera* species shows a strict specialisation: it is found only in birds of the genus *Lanius*.

The main food of the examined birds (*Motacilla flava*, *Lanius collurio*, *Oriolus oriolus*) are insects and their larvae, which are sometimes intermediate hosts of nematodes. The birds are likely to become infected in their wintering grounds because after arrival in the spring (April and May) they were found to carry only adult nematodes.

Life cycles of *Viguiera euryoptera* and *Diplotriaena ozouxi* are not documented, but it is known that the cycles of *Acuaria subula* and *Microtetrameres inermis* are complex; intermediate hosts are orthopterans.

This is the first record of *Acuaria subula* from *Motacilla flava* in Europe; *Viguiera euryoptera* and *Diplotriaena ozouxi* are new to the Polish fauna. Additionally, *Microtetrameres inermis* is recorded for the first time from *M. flava* and *Oriolus oriolus* in Poland.

#### Parasites of roe deer (*Capreolus capreolus*) from the Zlotowek Forest Inspectorate (Lower Silesia), Poland

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Coproscopic examinations of European roe deer (*Capreolus capreolus*) were conducted in September and October 2012 in the Zlotowek Forest Inspectorate (Lower Silesia). Rectal faecal samples were collected from 41 animals after planned hunting carried out in the Forestry, and were examined using Wilis-Shlaaf's flotation, McMaster and Baermann method.

Gastrointestinal, bronchopulmonary helminths and coccidia (*Eimeria*) were found in 39 out of 41 investigated fecal samples (95%). The greatest extent of infection was observed for gastrointestinal nematodes of the family Trichostrongylidae – 95%, with the average number of eggs in a single fecal sample being 94. The presence of a few nematode eggs from *Trichuris* spp. (22%) and *Nematodirus* spp. (9.7%) types was also noted. Larvae of *Dictyocaulus viviparus* lungworms were found in 17% of the fecal samples, numbering from 2 to 25 (average 6) per sample. Oocysts of coccidia of the genus *Eimeria* were present in 27% of stools, with an average number of 18 in a single sample.

According to the scientific data, a massive invasion of gastrointestinal parasites, especially nematodes, is having an impact on the size of the cervidae population, including roe deer. The negative effects of the parasites are manifested in disorders of reproduction, inhibition of growth of juveniles, reduced immunity and animal health, as well as lowering the quality of carcasses and antlers. Roe deer parasites are also a potential source of invasion for domestic ruminants. The population of roe deer has steadily increased in recent years, and the fields and meadows are their living space as well as its food base, leading to pollution of the environment with dispersive forms of parasites, including gastrointestinal roundworms. Hence this constitutes a real risk for animals, especially those kept in the grazing system. Based on the results obtained from the coproscopic examinations, it seems reasonable to continue the systematic parasitological monitoring of free-living wild animals. This will not only assess the efficacy of prophylactic measures, but it will also help to develop and implement a comprehensive program to improve the health of the roe deer population.

#### First record of the avian acanthocephalan *Polymorphus minutus* (Polymorphidae, Acanthocephala) parasitizing a raccoon (*Procyon lotor* L.)

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The adult forms of *Polymorphus minutus* are the rather common helminths which are noted quite often in many water bird species, whereas the larvae are found in scuds, which are the intermediate host. The wide host spectrum of this acanthocephalan encompasses birds recruited from many different families and orders, which allows the species to be described as an unspecialized parasite (generalist). Cases in which the hosts for this parasite are not birds are very rare and relate only to Arctic foxes (*Alopex lagopex*) from Alaska.

During this study on parasites of raccoons from the Czech Republic, 33 individuals were examined. In the intestines of two raccoons, 46 cystacanths were noted and identified as *Polymorphus minutus*. The research area from where the host material had been gathered can be described as bushy meadows placed on the outskirts of the river supplying the water complex of the fishponds. As this is a habitat for many wetland birds, the conditions are suitable for the existence and growth of this parasite. Examples of host barriers being overcome on the class level are rather rare for the parasites and all the identified acanthocephalan individuals are in the larval stage. Considering the context in which they were found, it seems that this record could be accidental. Since about thirteen species of acanthocephalan were noted in the raccoons, this first European record expands the list of the potential parasites for this host.

### Helminth parasites of wolves (*Canis lupus* L.) from north-western Poland

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The forests of north-western Poland, although very preferable for wolves, have only recently been recolonized by these carnivores. Up to now, research on wolf parasites has been conducted in the eastern part of the country, where the species population is quite numerous and stable. The newly established packs of wolves from four forest complexes west of the Vistula river have been examined for parasites: 1) Noteć Forest (PN) – one of the biggest forest complexes in the region, home for three packs of wolves,  $5\Box 7$ individuals each; 2) Tuchola Forest (BT) – no less than two packs, 6 wolves in one of them and no less than 4 in the other; 3) Lower Silesia Forest (BD) - one of the biggest forest complexes in Europe, with four stable packs of wolves and one border pack with Germany; 4) Drawa National Park (DPN) – placed in the centre of the Drawa Forest, with one pack of wolves (up to 5 individuals). Parasite diagnostics was conducted by coproscopy with the use of a modified flotation method incorporating centrifugation (1500 rpm/5 min.). The analysis of 227 faecal samples (PN-84, BT-46, BD-54, DPN-43) led to identification of the eggs and oocysts of 9 parasite taxa: Alaria alata (9.69%; 5.95% PN; 17.39% BT; 5.56% BD; 13.95% DPN), Taeniidae gen. sp. (34.36%; 34.52% PN; 45.65% BT; 31.48% BD; 25.58% DPN), Capillariidae gen. sp. (25.55%; 30.95% PN; 28.26% BT; 27.78% BD; 9.30% DPN), Trichuris vulpis (2.20%; 3.57% PN; 4.35% BT). Ancylostoma sp. /Uncinaria sp. (32.60%; 35.71% PN; 43.48% BT; 11.11% BD; 41.86% DPN), Toxocara canis (7.49%; 8.33% PN; 8.70% BT; 13.95% DPN), Toxascaris leonina (0.88%; 2.38% PN), Physaloptera sp. (2.46%; 1.19% PN; 4.35% BT; 6.98% DPN) and coccidia (26.43%; 28.57% PN; 39.13% BT; 16.67% BD; 20.93% DPN).

# The structure and distribution of the parasite assemblages of roaches (*Rutilus rutilus* L.) along the Odra River against the background of environmental factors

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The main objective of the study was to trace the multi-directional effect of environmental factors, both natural and anthropogenic, on the distribution and structure of roach (Rutilus rutilus L.) parasite assemblages along the Odra River. A total of 451 roach were caught in seven sites along the whole (854 km) course of the Odra and were subjected to full parasitological section. In all, 68 parasite taxa were isolated and identified; they represented nine higher taxa: Monogenea, Aspidogastrea, Digenea, Cestoda, Nematoda, Acanthocephala, Hirudinea, Crustacea and Bivalvia. The results indicate the effect of environmental factors on the parasite communities associated with roach in the Odra. In a few cases, the observed relationships were fairly clear and explainable, based on the biology, life strategies and developmental types of the analysed groups of parasites. This was especially true of the level of industrialisation and water quality in its broad sense, with respect to nearly all the analysed assemblages, where the effect was mainly executed through the intermediate hosts (aquatic invertebrates), free-living larval stages or, in the case of ectoparasites, directly on adult stages of parasites. The effect of pollution was most often manifested "negatively", that is decreasing the indices of occurrence to greater or lesser degrees, for example, Digenea, Cestoda, Acanthocephala, Crustacea, bivalve glochidia or intestinal or heteroxenic parasites, endoparasites or crustacean- or mollusc-transmitted parasites. In a few cases the effect was "positive" increasing the indices of occurrence to varying degrees, as seen in, for example, Monogenea or oligochaete-transmitted parasites. Among the remaining factors affecting the roach parasite communities, riverbed partitioning, vicinity of major bird refuges or fish farms, as well as flow velocity can be mentioned. The riverbed partitioning differentiated the indices of occurrence in Digenea and probably also Aspidogastrea. The neighbourhood of bird refuges affected mainly allogenic Digenea and Cestoda, while the presence of fish farms affected the community of Digenea. The flow velocity mainly influenced the distribution of bivalve glochidia and, to a lesser extent, the assemblage of Monogenea. The neighbourhood of the estuary and sea water was also an important factor for the roach parasite community of the lower section of the Odra. The roach inhabiting the lower section of the river migrate to coastal waters and assume parasites from the group of generalists which occur in typically marine hosts. The results indicate also that the relationship between the environmental factors and the parasite assemblages are rarely simple, unequivocal and easy to interpret. Various biotic factors often interact with each other or with abiotic conditions. Their synergistic action may be expressed as weakening the end effect, cancelling each other's influence, or partially limiting the effect of one of the factors.

# Parasitological monitoring of the wild and domestic horses (*Equus przewalskii*, e. *Caballus*) in the Chernobyl exclusion zone (Ukraine) during 2012–2013 years

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There are fifty seven free-living Przewalski's horses (*Equus przewalskii*) kept in Chernobyl exclusion zone (CEZ), Ukraine, since 1998. Simultaneously, 6 domestic horses are bred in this territory in private stables. There is an exchange of parasites between wild and domestic horses grazing on the same grassland. Parasitic fauna is one of the factors which may influence the mortality rate in animals. It is, therefore, especially important to investigate ecto- and endo- parasite diversity in free-living and domestic animals in the reserve.

In this study, we investigated gastro-intestinal parasites and tick burdens of wild horses living in the natural reserve conditions in CEZ, Ukraine, using parasitological methods. The study was carried out in the years 2012–2013 in CEZ, Ukraine. Twenty nine naturally infected wild Przewalski's and domestic horses (23 and 6 respectively) with different EPG level were examined in 2012–2013. Faecal egg counts were performed using the McMaster technique with sensitivity of 25 eggs per gram (EPG). A total of 35 faeces samples of domestic horses were tested from November 2012 to April 2013. Of the surveyed domestic horses the last was treated in March 2012 with 0.2% aversectin C.

In total, 88.6% of domestic horses were infected with strongylids; the mean egg output was 496 EPG (50 –1625), 20% of horses had *Parascaris* eggs (282.1 EPG, 25–1000) during period November 2012 – April 2013. The investigated domestic horses were treated in February 2013 with pirantel 300 mg + prazikvantel 20 mg. All Przewalski's horses were infected with strongylids (EI=100%); the mean egg output was 268.5 EPG (25–750), 22.2% of horses had habronematides eggs (25 EPG). Only one horse had mixed invasion (strongylids+habronematides). Gastro-intestinal parasites of the wild and domestic horses included strongylides, parascarides and habronematides eggs in the tested faeces samples.

The ticks burden was carried out using the method of mechanical collection from horses and natural environment in the CEZ in September 2012. All collected ticks were *Dermacentor reticulatus*.

# Intestinal parasites of raccoon dog (*Nyctereutes procyonoides*) from western Poland

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The raccoon dog is considered the most successful invading mammal species in Europe. This species has a negative influence on biodiversity in ecosystems and may play an important role in transmission of viral, bacterial, and parasitic diseases. Studies of parasitological fauna of this invasive species living in our country may be essential for health of humans and native species of animals.

The main goal of this study was to examine raccoon fecal samples for the presence of intestinal parasites and assess the health hazard for humans, as well as domestic and wild animals. A total of 51 raccoon dog fecal samples were collected from two sites in western Poland. Fecal samples were examined using coprological and immunological methods. In examined samples *Isospora* and *Cryptosporidium* oocysts as well as *Toxocara* and *Trichuris* eggs were found.

### *Cryptosporidium* sp. oocysts in freshwater bivalves from Malta Lake, Poznan

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Clams can concentrate waterborne pathogens and can be used for sanitary assessment of water quality. The aim of our study was to examine bivalves from Malta Lake in Poznan for the presence of *Cryptosporidium* oocysts and *Giardia* cysts.

A total of 114 freshwater bivalves of two species were collected in November 2012 during complete drainage of the water reservoir. Detection of oocysts was performed using microscopic methods and a commercially available IFA kit. *Cryptosporidium* oocysts were found in 12 (10.5%) of the 114 analyzed samples. Therefore, our results suggest that the studied bivalve species are reservoirs of protozoan parasites and may be useful indicators of water contamination with the fecal material of humans or other animals.