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## Seroprevalences of antibodies to *Neospora caninum* and *Toxoplasma gondii* in zoo animals

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### Abstract

*Neospora caninum* is an apicomplexan parasite that causes neuromuscular disease in dogs and abortions in cattle. Little is known about the prevalence of antibodies to this parasite in zoo animals. Sera from 556 animals, from 13 Czech and Slovak zoos were tested for antibodies to *N. caninum* and *Toxoplasma gondii* by indirect fluorescent antibody test. Antibodies to *N. caninum* were found in 31 of 556 zoo animals (5.6%), representing 18 of 114 species tested: Eurasian wolf (*Canis lupus lupus*), Maned wolf (*Chrysocyon brachyurus*), fennec (*Vulpes zerda*), cheetah (*Acinonyx jubatus*), jaguarundi (*Herpailurus yagouaroundi*), Eurasian lynx (*Lynx lynx*), Indian lion (*Panthera leo goojratensis*), fisher (*Martes pennanti*), blackbuck (*Antelope cervicapra*), European bison (*Bison bonasus*), lechwe (*Kobus leche*), African buffalo (*Syncerus caffer caffer*), eland (*Taurotragus oryx*), sitatunga (*Tragelaphus spekei gratus*), Thorold's deer (*Cervus albirostris*), Eastern elk (*C. elaphus canadensis*), Vietnam sika deer (*C. nippon pseudaxis*) and Père David's deer (*Elaphurus davidianus*). Titres ranged from 1:40 to 1:2560. The highest prevalence 50% was found in family mustelidae of the order carnivora. Antibodies to *T. gondii* were detected in 193 of 556 zoo animals (34.7%) representing 72 of 114 species tested, with titres ranging from 1:40 to 1:40960. The highest prevalence 100% was found in families: hyaenidae, mustelidae, ursidae and viveridae of the order carnivora. The results of this study indicate that zoo animals have more exposure to *T. gondii* than to *N. caninum*. It is the first report of seroprevalence of antibodies to *N. caninum* in European zoo animals.

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### 1. Introduction

*Neospora caninum* and *Toxoplasma gondii* are apicomplexan parasites that may cause clinical

disease in domestic and wild animals (Dubey and Beattie, 1988; Dubey, 2003). Little is known about the pathogenicity of *N. caninum* infection in wildlife, however, fatal neosporosis was reported in two black tailed deer (*Odocoileus hemionus columbianus*) from California, one full-term stillborn Elds' deer (*Cervus eldi siamensis*) from Paris zoo, two full-term stillborn

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twin calves of lesser kudu (*Tragelaphus imberbis*) from German zoo, one 16-day-old white rhinoceros calve (*Ceratotherium simium*) from the National zoological gardens of South Africa and juvenile fallow deer (*Dama dama*) from Switzerland (Woods et al., 1994; Dubey et al., 1996; Peters et al., 2001; Williams et al., 2002; Soldati et al., 2004).

Antibodies to *N. caninum* were found in 6 wildlife species (white-tailed deer, moose, raccoon, gray fox, coyot, wolf) from USA (Lindsay et al., 1996, 2001a, 2001b, 2002; Dubey et al., 1999; Gondim et al., 2004), 10 wildlife species (opossum, 5 species of brocket, Maned wolf, Chiloes fox, crab-eating fox, pampas fox) from South America (Patitucci et al., 2001; Melo et al., 2002; Yai et al., 2003; Canón-Franco et al., 2004; Tiemann et al., 2005), 7 wildlife species (European bison, Alpine ibex, chamois, red deer, roe deer, red fox, brown hare) from Europe (Buxton et al., 1997; Simpson et al., 1997; Wolfe et al., 2001; Ferroglio et al., 2001a; Ferroglio and Rossi, 2001b; Ferroglio and Trisciuglio, 2003; Cabaj et al., 2005), 9 wildlife species (zebra, eland, Thompson gazelle, impala, buffalo, warthog, lion, cheetah, spotted hyena) from Africa (Cheadle et al., 1999; Ferroglio et al., 2003) and 1 free-ranging species (dingo) from Australia (Barber et al., 1997). Dubey et al. (2003) reported *N. caninum* antibodies in 7 marine mammals species (sea otter, Pacific harbour seal, sea lion, ringed seal, bearded seal, Atlantic bottlenose dolphin, walrus).

Since reports of prevalence of *N. caninum* in zoo animals are limited to two studies in felids from South Africa and North America (Cheadle et al., 1999; Spencer et al., 2003), one report in canids from South America (Vitaliano et al., 2004) and one report in cervids from South America (Tiemann et al., 2005), prevalence of neosporosis in other mammal species is largely unknown. To date, no study reported seroprevalence of *N. caninum* in animals from European zoos.

There are many reports on toxoplasmosis in zoo animals. For example, fatal toxoplasmosis was recorded in captive dik-dik and manul from zoo in USA (Dubey et al., 2002; Riemann et al., 1974a, 1974b), in lions from zoo in Africa (Ocholi et al., 1989) and in Siberian tiger from zoo in Belgium (Dorny and Franssen, 1989). In the Czech Republic, fatal toxoplasmosis was recorded in saiga and nilgais antelope from Prague and Chomutov zoos (Sedlák et al., 2004).

Antibodies to *T. gondii* were found for example in 6 felids from zoo in Brazil (Silva et al., 2001) and in nine felids and other 104 exotic mammals from zoo in USA (Lappin et al., 1991; Riemann et al., 1974a, 1974b). In the Czech Republic, serologic assay in zoo animals was not done yet.

The present study was done to determine the prevalence of antibodies to *N. caninum* in zoo animals in Czech and Slovak zoos. Additionally, we determined the prevalence of antibodies to another apicomplexan parasite *T. gondii* in these animals.

## 2. Materials and methods

Sera from 556 zoo animals (114 mammal species or subspecies) were obtained from 12 Czech zoos, 1 Slovak zoo and 4 small private exotic breeds in the Czech Republic. Samples were collected by private veterinaries for State Veterinary Institute Prague during years 1995 till February 2005. The majority of sera samples had been collected from clinically healthy zoo animals. Sera were centrifuged and frozen at  $-20^{\circ}\text{C}$  until they were examined in the year 2005.

Serum samples were examined for antibodies against *N. caninum* and *T. gondii* by an indirect fluorescence antibody test (IFAT) using a commercially available NC-1 tachyzoite slides (VMRD, Pullman, USA) and Sevatest toxoplasma NIFR (Sevapharma, Prague, Czech Republic). Species-specific conjugate were used: anti-monkey IgG (Sigma-Aldrich s.r.o., Prague, Czech Republic) for cercopithecidae, anti-human IgG (Sevapharma) for hominidae, anti-dog IgG (Sigma) for canidae, anti-cat IgG (Sigma) for felidae, hyaenidae, mustelidae, otariidae, ursidae and viveridae, anti-horse IgG (VMRD) for equidae, anti-llama IgG (VMRD) for camelidae, anti-swine IgG (Sigma) for suidae, anti-deer IgG (KPL) for cervidae, anti-bovine IgG (VMRD) for bovidae except genus *Ammotragus*, *Budorcas*, *Capra*, *Hemitragus*, *Nemorhaedus*, *Oreamnos*, *Ovibos*, *Ovis* and *Pseudois*, in which anti-goat IgG (VMRD) was used. The sera were diluted in a two-fold series starting at 1:20 as a basic dilution. The titre  $\geq 1:40$  was considered positive. Continuous peripheral fluorescence was considered specific (Conrad et al., 1993; Paré et al., 1995).

*N. caninum* positive and negative dog and cattle sera (VMRD) were used as controls. Sera from

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