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## SURVEY FOR TOXOPLASMOSIS IN WILD AND DOMESTIC ANIMALS FROM NORWAY AND SWEDEN

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**Abstract:** Fifty-nine of 1250 (4.7%) wild and domestic animals from Norway and Sweden had positive dye-test titers ( $\geq 1:8$ ) for antibody against *Toxoplasma gondii*. A dye-test titer of 1:8 (30-40 i.u.) or higher was detected in 3 of 732 small rodents (0.4%), 21 of 87 domestic cats (24%), 9 of 29 red foxes (*Vulpes vulpes*) (31%), 2 of 2 domesticated arctic foxes (*Alopex lagopus*), 12 of 99 red deer (*Cervus elaphus*) (12%), 5 of 8 roe deer (*Capreolus capreolus*) (63%) and in 7 of 34 wild rabbits (*Oryctolagus cuniculus*) (21%). Antibodies were not found in 29 shrews (*Sorex* spp.), 127 gulls (*Larus* spp.), 4 terns (*Sterna* sp.), 10 skuas (*Stercorarius* sp.), 68 domestic reindeer (*Rangifer tarandus*) and 21 wild reindeer.

Histologic examination of brain tissue from another 51 wild rabbits, on which serological data were not available, did not reveal cysts. Sero-conversion was not observed in laboratory mice inoculated with the same material. Infection with *T. gondii* was confirmed in two of the three sero-positive small rodents using a FA-technique. Cysts were not detected in the brains of another 55 rodents, of which 26 were sero-negative and 29 were not tested serologically.

### INTRODUCTION

Infection with *Toxoplasma gondii* has been recorded from a number of domestic species in Norway<sup>1,2,4,8,14,15</sup> but only a few surveys have been conducted. The veterinary importance of *T. gondii* in Norway has been clearly demonstrated especially as a cause of abortion in sheep.<sup>15</sup> Serologic evidence indicates that human infections with this parasite are widespread.<sup>7,10,11,13</sup> However, information on toxoplasmosis among free-ranging wild species is sparse<sup>9</sup> and a wildlife survey is entirely lacking. Thus, further research is needed to understand the epizootiology and epidemiology of toxoplasmosis.

The purpose of this investigation was to conduct a survey for toxoplasmosis in some animal populations from Norway, with special emphasis on small rodents. Three domestic species are included: cat, reindeer (*Rangifer tarandus*) and domesticated arctic fox (*Alopex lagopus*).

Also incorporated in this study are free-ranging wild mammals from one locality in southern Sweden.

### MATERIALS AND METHODS

#### Serology

Plasma or sera was collected from 1250 wild and domestic animals (Table 1). The geographic locations of the study areas are shown in Fig. 1. Heparin was used as anticoagulant in plasma samples. All specimens were inactivated at 56 C for 30 min. except the sera from reindeer (*Rangifer tarandus*), red deer (*Cervus elaphus*) and roe deer (*Capreolus capreolus*) which required longer inactivation (45 - 60 min.). Plasma or sera was tested by a microtitre modification of the Sabin-Feldman dye-test.<sup>11</sup> Sensitivity of the dye-test was determined by serial dilutions of the International Standard of Anti-*Toxoplasma* Serum.<sup>18</sup> A titer of 1:8 corresponded to a specific antibody concentration in the range of 30-40 i.u.

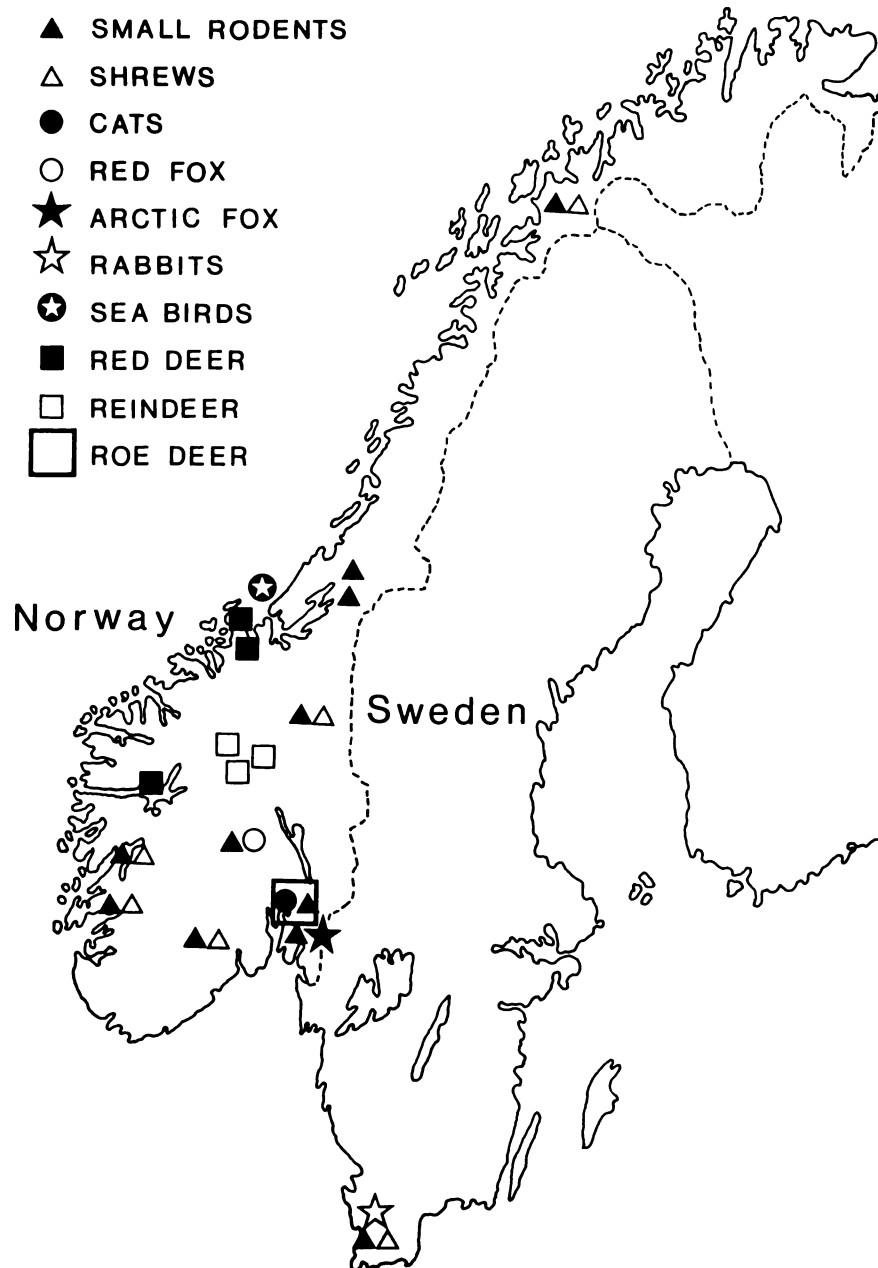


FIGURE 1. Localities in Norway and Sweden surveyed for toxoplasmosis in wild and domestic animals.

per ml undiluted serum/plasma. Titers refer to final dilutions.

### Parasitology

**Arctic fox (*Alopex lagopus*):** Twenty per cent tissue suspensions in sterile saline were made of liver, spleen, brain and diaphragm from one sero-positive arctic fox (titer 1:32). Five laboratory mice (NMRI(BOM) f spf) were each inoculated intraperitoneally with 0.5 ml of suspension from each organ. Mice were tested serologically six weeks later.

**Wild rabbits (*Oryctolagus cuniculus*):** Fifty-one rabbits, on which serologic data were not available, were examined: 2 - 3 cm<sup>3</sup> of the forebrain was removed, homogenized and examined for cysts by direct microscopic inspection.<sup>16</sup> Pools of four to five specimens were inoculated into laboratory mice as specified for the foxes.

**Small rodents:** The brains of all sero-positive small rodents were checked for cysts by direct microscopic inspection. Brain emulsions were further examined by a *Toxoplasma* specific immunofluorescence technique.<sup>12</sup> One sero-positive bank vole (*Clethrionomys glareolus*) was in a late stage of pregnancy. Her uterus, together with six fetuses, was homogenized and submitted to the same FA-test.

Serial histologic sections of the entire brain from 29 small rodents from Kviteseid, Norway, were stained with hematoxylin/eosin and examined microscopically for cysts. Serologic data were not available from these animals.

Brains of 26 sero-negative small rodents from Revinge in Skåne, Sweden, were examined for cysts by direct microscopic inspection.

## RESULTS

### Serology

Fifty-nine of 1250 (4.7%) wild and domestic animals from Norway and Sweden had positive dye-test titers ( $\geq 1:8$ ) for antibody against *T. gondii* (Table 1).

### Parasitology

Cysts were not detected by direct microscopic inspection of the brains of the three sero-positive small rodents. However, the presence of *T. gondii* was established in two of these animals using a FA-technique: trophozoites were found in the brain of a Norway lemming (*Lemmus lemmus*) and in the brain and the uterus containing fetuses of a bank vole.

Cysts were not found in histologic brain sections of 29 small rodents from Kviteseid, Norway.

*Toxoplasma*-like cysts were found in the brain of one sero-negative bank vole from Revinge in Skåne, Sweden. FA-results, however, were negative. Cystic stages were not observed in the brains of another 25 small rodents from the same locality.

Cysts were not found by microscopic inspection of brain material from 51 wild rabbits. Sero-conversion was not observed in laboratory mice inoculated with the same material.

The laboratory mice inoculated with tissue suspensions of brain, spleen, liver and diaphragm from one sero-positive arctic fox were negative by the dye-test.

## DISCUSSION

This study demonstrated significant differences in the frequency of sero-positive animals between the three species of cervids examined. These observations may be attributed to differences in climate and presence of cats, affecting the survival and occurrence of oocysts, respectively. Negative results were obtained from 68 wild and domestic reindeer which graze in sparsely populated mountain regions at high altitudes where cats and other felids are absent. Red deer and roe deer, on the other hand, graze in the lowlands of coastal regions close to human settlements, where conditions are more conducive to infection. The highest frequency of sero-positive cervids was

TABLE 1. *Toxoplasma gondii* in wild and domestic animals from Norway and Sweden as determined by the Sabin-Feldman dye-test.

Species*	No. tested	No. positive	Per cent positive	Dye-test titer**				
				<1:8	1:8	1:16	1:32	1:64
Domestic cat ( <i>Felis domesticus</i> )	87	21	24	66	8	9	3	1
Red fox ( <i>Vulpes vulpes</i> )	29	9	31	20	4	2	2	1
Arctic fox (domesticated) ( <i>Alopex lagopus</i> )	2	2	100	0	—	—	2	—
Red deer ( <i>Cervus elaphus</i> )	99	12	12	87	8	4	—	—
Roe deer ( <i>Capreolus capreolus</i> )	8	5	63	3	1	—	2	1
Wild rabbit ( <i>Oryctolagus cuniculus</i> )	34	7	21	27	7	—	—	—
Bank vole ( <i>Clethrionomys glareolus</i> )	408	2	0.4	406	2	—	—	—
Norway lemming ( <i>Lemmus lemmus</i> )	40	1	3	39	1	—	—	—

\*The following species gave negative results (number of individuals in parentheses):

domestic reindeer - *Rangifer tarandus* (68), wild reindeer (21), grey-sided vole - *Clethrionomys rufocanus* (5), field vole - *Microtus agrestis* (46), root vole - *Microtus oeconomus* (17), wood mouse - *Apodemus sylvaticus* (136), yellow-necked field mouse - *Apodemus flavicollis* (10), house mouse - *Mus musculus* (70), shrews - *Sorex* spp. (29), common gull - *Larus canus* (1), herring gull - *Larus argentatus* (77), great black-backed gull - *Larus marinus* (3), lesser black-backed gull - *Larus fuscus* (46), arctic tern - *Sterna macrura* (4) and arctic skua - *Stercorarius parasiticus* (10).

\*\*The figures indicate the number of individuals reacting at each dilution. Titers refer to final serum/plasma dilutions. Titer 1:8 corresponded to a specific antibody concentration in the range of 30-40 i.u. per ml undiluted serum/plasma. Titers <1:8 were considered negative.

detected among the roe deer. Samples originated from the districts surrounding the Oslo Fjord, one of the most densely populated regions of Norway.

Similar observations have been made by other researchers. Waldeland<sup>14</sup> reported that in sheep from southwestern Norway toxoplasmosis was more prevalent on lowland pastures than on mountain pastures. In a serologic study of domestic pigs in the region surrounding the Oslo Fjord, Hellesnes *et al.*<sup>8</sup> found a higher prevalence of dye-test positive pigs in the coastal than in the inland zone. In California, Franti *et al.*<sup>5</sup> detected the highest prevalence of sero-positive carnivores, rodents and sheep in the coastal regions below 30 m elevation. From Austria, Werner *et al.*<sup>17</sup> stated that the near vicinity of human settlements is apparently positively correlated with a higher risk of infection with *T. gondii* in free-ranging wild animals. Present results are similar.

This study included 732 small rodents from eight species. To obtain a representative picture of the prevalence of toxoplasmosis among these animals, samples were taken at widespread localities representing several biotopes and varying degrees of contact with human activity and from different stages of the population cycle. Antibodies to *T. gondii* were detected in only three (0.4%) of these animals. The results, however,

may not necessarily give an exact expression of the prevalence of *Toxoplasma*. Chronically infected, but sero-negative individuals have been recorded from a number of species, including some small rodents;<sup>6</sup> thus, prevalence may be higher than indicated by serologic examination. Doby *et al.*<sup>3</sup> presented experimental evidence indicating that *Apodemus sylvaticus* and *Clethrionomys glareolus* are unable to maintain detectable levels of antibodies for long periods of time when infected with a less pathogenic strain of *T. gondii*. To obtain more conclusive information concerning the prevalence of infection, 58 small rodents were examined by parasitologic methods. The results suggested that prevalence may not be much higher than demonstrated by the serologic examination. It also should be stressed that the results pertain only to the trappable part of the small rodent populations under study. Animals with acute infections may succumb or remain in their nests and burrows.

A low rate of infection among small rodents may account for relatively high prevalences of antibodies against *T. gondii* in cats and foxes, since these animals may consume hundreds of small rodents during a year. The foxes frequently had access to wastes from slaughtered sheep and swine. This may represent another important source of infection.

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