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# SEROLOGIC SURVEY FOR *TOXOPLASMA GONDII* IN GRIZZLY BEARS FROM ALASKA

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ABSTRACT: Blood samples were collected from 892 grizzly bears (*Ursus arctos*) in Alaska (USA) from 1973 to 1987. Sera were tested for evidence of exposure to *Toxoplasma gondii* by means of the modified agglutination test. Two hundred twenty sera (25%) had titers ≥25, the minimum threshold titer. Six hundred seventy-two sera (75%) had titers <25. Antibody prevalence ranged from 9% (18 positive of 196 tested) in southern areas to 37% (162 of 433 tested) in northern areas. There was no readily apparent explanation for these discrepancies in location-specific prevalence.

Key words: Alaska, grizzly bear, serology, Toxoplasma gondii, Ursus arctos.

#### INTRODUCTION

Toxoplasma gondii is a protozoan parasite with worldwide distribution (Dubey and Beattie, 1988). Clinical signs of toxoplasmosis in humans and other animals include lymphadenopathy, myalgia and neuralgia (Dubey and Beattie, 1988). Infection can cause pregnant domestic sheep and goats to abort (Dubey, 1994).

Domestic and free-ranging felids are the only recognized definitive hosts for T. gondii. The parasite multiplies in the gastrointestinal tract of cats. Infective T. gondii oocysts are excreted in feces. Other mammals can become infected by ingesting food or water contaminated with oocysts. The parasite multiplies in the gastrointestinal tract of these secondary hosts. The resulting developmental stages circulate via the blood and lymphatic systems. Tissue cysts form in various tissues. Ingestion of these cysts provides another form of transmission (Dubey, 1994). Grizzly bears (Ursus arctos) may be exposed to T. gondii via either route. Several test procedures have been employed to detect T. gondii antibody in mammal sera (Peterson et al., 1974; Kocan et al., 1986; Chomel et al., 1995). The modified agglutination test (MAT) is the most sensitive procedure for detection of latent T. gondii infections in black bears (Dubey et al., 1995).

Serum antibody prevalence of T. gondii

was 23% (25 positive of 110 tested) for moose (Alces alces) from southern portions of Alaska (USA) from 1974 to 1982 (Kocan et al., 1986), using the indirect hemagglutination test (IHA). Antibody prevalence was 28% in 1,572 Alaska Natives tested during the early 1970s (Peterson et al., 1974), using the indirect fluorescent antibody test and the IHA. Antibody prevalence was 18% (87 positive of 480 tested) for grizzly bears and 15% (six positive of 40 tested) for black bears (Ursus americanus) captured in Alaska from 1988 to 1991 (Chomel et al., 1995), using the latex agglutination test (LAT).

The objectives of the current project were to determine 1) the serum antibody prevalence of *T. gondii* in grizzly bears from Alaska and 2) the relationship between antibody prevalence and sex, age, location, and year of collection.

#### **MATERIALS AND METHODS**

Bear blood was collected by personnel of the Alaska Department of Fish and Game and the U.S. Fish and Wildlife Service during studies of bear population ecology (Fig. 1). Individual study areas were combined into three regions: (1) Northern Region composed of Northeast Arctic, Northwest Arctic, and Western Arctic; (2) Interior Region composed of Southcentral Interior, Central Interior, and Eastern Interior; and (3) Southern Region composed of Alaska Peninsula, Kodiak Island, and Southeastern Islands. Sera were stored temporarily at -12 C and subsequently at -40 to -50 C for up to

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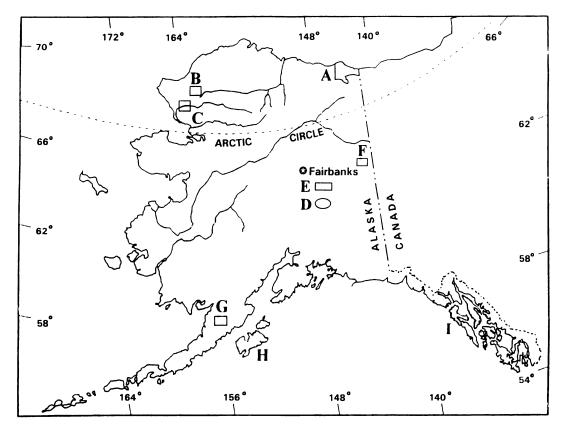


FIGURE 1. Location of collection sites for grizzly bears (*Ursus arctos*) included in *Toxoplasma gondii* serologic survey. A, Northeast Arctic (69° to 70°N; 141° to 146°W); B, Northwest Arctic (69° to 70°N; 158° to 162°W); C, Western Arctic (67°30′ to 68°30′N; 158° to 162°W); D, Southcentral Interior (62°30′ to 63°30′N; 146° to 148°W); E, Central Interior (62°45′ to 64°15′N; 146°30′ to 148°30′W); F, Eastern Interior (64°30′ to 65°N; 141° to 143°W); G, Alaska Peninsula (57° to 58°N; 156° to 157°W); H, Kodiak Island (57° to 58°N; 152° to 155°W); I, Southeastern Islands (57° to 58°N; 134° to 135°W).

22 yr until the time of testing. Ages of bears (in years) were determined by examining cementum annuli of premolar teeth (Craighead et al., 1970).

Sera were tested by means of the MAT (Dubey and Desmonts, 1987). Mercaptoethanol was incorporated with whole formalinized tachyzoites in the test procedure. Sera which agglutinated the antigen suspension at a serum dilution  $\geq 1:25$  were considered indicative of previous natural exposure to *T. gondii*. Sera with a titer  $\geq 25$  will be referred to as positive. All others will be referred to as negative.

A generalized linear model (McCullagh and Nelder, 1989) with a logit link and a binomial distribution was used to analyze the relationship between serologic test results and four host parameters: 1) age, 2) sex, 3) location, and 4) year of collection. Location and sex were categorical variables. Age and year of collection were continuous variables. The four host pa-

rameters were entered into the model sequentially in order of importance. The significance of each factor was determined by the following criteria: 1) degree to which the factor improved the fit of the model and 2) amount by which the factor increased the likelihood of a positive serologic test result. Significance ( $\alpha=0.05$ ) was determined by comparing the increase in the log-likelihood to a chi-square distribution with the appropriate degrees of freedom. Quadratic terms were added for the continuous variables. All second-order interactions were evaluated.

### **RESULTS**

Two hundred twenty sera had titers ≥25. Twenty-four samples (3%) had a titer of 25. One hundred thirty-two (15%) had a titer of 50. Sixty-four (7%) had a titer

Table 1. Serum antibody prevalence of *Toxoplasma* gondii in grizzly bears (*Ursus arctos*) from Alaska, 1973 to 1987.

Region	Location	Preva- lence <sup>a</sup>	Per- cent
Northern	Northeast Arctic	60/208	29
	Northwest Arctic	80/166	48
	Western Arctic	22/59	37
	Subtotal	162/433	37
Interior	Southcentral	25/152	16
	Central	15/98	15
	Eastern	0/8	0
	Subtotal	40/258	16
Southern	Alaska Peninsula	5/18	28
	Kodiak Island	11/151	7
	Southeastern Islands	2/27	7
	Subtotal	18/196	9

<sup>&</sup>lt;sup>a</sup> Number positive/number tested.

≥500. Six hundred seventy-two sera (75%) were negative. Serologic test results were not confirmed by means of either histologic examination or attempted isolation of *T. gondii* from bear tissues.

The host parameter with the greatest predictive value was location (Table 1). The order of the remaining parameters was as follows: age, year of collection, and sex. Quadratic terms for age and year of collection were not significant. No second-order interactions were significant. The final model included location, age and year of collection. Parameter estimates were as follows: (1) Intercept = 3.6213, (2) South = 0.00, (3) Interior = 0.8514, (4) North = 1.8369, (5) Age = 0.0847, and (6) Year = -0.0829.

Probability of a positive serologic test result can be predicted by the following formula:

Probability = 
$$\frac{e^f}{1 + e^f}$$

where  $f = 3.6213 + (0.00, if South) + (0.8514, if Interior) + (1.8369, if North) + (Age <math>\times$  0.0847) - (Year  $\times$  0.0828). The probability that an individual serum would be positive 1) increased at northern latitudes, 2) increased with the age of the

bear, and 3) decreased during the study period.

#### DISCUSSION

Antibody prevalence had a distinct geographic pattern. Prevalence was low in bear populations from southern regions of Alaska. Prevalence was high in northern regions (Table 1). Similar geographic patterns have been reported for brucellosis in bears and respiratory viruses in caribou (Rangifer tarandus) (Zarnke, 1991). No explanation for this pattern is readily apparent.

Prevalences reported here for Northern and Interior Regions were higher than those reported previously for grizzly bears (Chomel et al., 1995). These discrepancies may be due to the different test methods employed in the two surveys. The MAT which was employed in the current survey is more sensitive than the LAT which was used in the previous study (Dubey et al., 1995). No bear tissues were examined histologically to confirm *T. gondii* exposure.

The source of *T. gondii* exposure for grizzly bears is unknown. Felids infected with *T. gondii* shed infective oocysts in feces. Bears could be exposed via ingestion of food or water contaminated by felid feces. Domestic cats (*Felis domesticus*) are uncommon in rural Alaska villages. In most areas of mainland Alaska, feral domestic cats do not survive outside of established communities. Therefore, domestic cats are not believed to represent a major source of exposure for bears.

Lynx (Lynx canadensis) are found throughout most of the mainland. However, lynx are absent from both Kodiak Island and the Southeastern Islands. In addition, lynx are rare north of the Brooks Range (68°N) (Bee and Hall, 1956). The absence of lynx from the two southern areas coincides with low antibody prevalence in bears. However, the low population density of lynx in the northern areas is in direct contrast to the high *T. gondii* antibody prevalence in bears. Therefore, lynx are apparently not the primary source of

exposure for bears in the Northern Region.

Grizzly bears in the northern areas of Alaska are presumably exposed to *T. gondii* via ingestion of infected meat. Potential sources of *T. gondii* include moose, caribou, arctic ground squirrels (*Citellus undulatus*) and other bears. Caribou herds in both the northeast and northwest arctic migrate to more southerly areas on a seasonal basis. In the southern portion of their range, these caribou have opportunity to interact with lynx. Perhaps these caribou herds serve as a continuing source of *T. gondii* introduction to the Northern Region.

Antibody prevalence was directly related to age of bears. This is a common pattern for many host species and disease agents. Apparently, opportunity for exposure to *T. gondii* is routinely available. As a bear ages, its cumulative likelihood for exposure increases.

There was a minor decrease in antibody prevalence during the years of the survey. This factor was the least statistically significant of those which were evaluated. No obvious behavioral or environmental changes could be identified which might have been related to this minimal decline in prevalence.

Humans occasionally eat grizzly bear meat. Results of the current study indicate that bear meat could serve as a source of *T. gondii* exposure for humans and other mammals. In order to reduce the potential for transmission of *T. gondii* to humans, bear meat should be thoroughly cooked prior to consumption.

Grizzly bear population dynamics are stable in most areas of Alaska. Results of the current study provide no indication that *T. gondii* has a negative effect on grizzly bear populations.

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