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Source: Journal of Wildlife Diseases, 33(3) : 649-652

Published By: Wildlife Disease Association

URL: <https://doi.org/10.7589/0090-3558-33.3.649>

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## Serologic Survey for Toxoplasmosis in River Otters

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**ABSTRACT:** The prevalence of antibody titers to *Toxoplasma gondii* in river otters (*Lutra canadensis*) from eastern North Carolina (USA) was investigated in a cross sectional study. Sera from 103 live trapped river otters were tested for antibodies to *T. gondii* using a commercially available latex agglutination kit. Forty-six (45%) of the sera were positive with titers ranging from 1:16 to >1:2,048. Adult otters ( $n = 78$ ) had a seroprevalence of 47% and juvenile otters ( $n = 25$ ) had a seroprevalence of 39%. Significant differences were not found between the sexes within either maturity class. The high prevalence of antibodies to *T. gondii* indicates that many animals in this population were exposed to the organism. This suggests handling of otters may be a zoonotic concern for fur trappers.

**Key words:** Antibodies, *Lutra canadensis*, river otter, serology, serosurvey, *Toxoplasma gondii*.

*Toxoplasma gondii*, an obligate intercellular protozoan, infects humans, domestic and nondomestic mammals, and birds throughout the world (Dreesen, 1990). Domestic and wild felines are the definitive hosts, capable of shedding infective oocysts into the environment (Dubey and Beattie, 1988). In North America, wild mustelids such as mink (*Mustela vison*) (Franti et al., 1976; Tizard et al., 1976), fishers (*Martes pennanti*) (Tizard et al., 1976), striped and spotted skunks (*Mephitis mephitis*, *Spilogale putorius*) (Franti et al., 1976; Tizard et al., 1976), martens (*Martes martes*) (Tizard et al., 1976), and other aquatic mammals such as nutria (*Myocastor coypus*) (Howerth et al., 1994), muskrat (*Ondatra zibethicus*) (Smith and Frenkel, 1995) and beaver (*Castor canadensis*) (Smith and Frenkel, 1995) are reported with antibodies to *T. gondii*, indicating exposure and probable tissue infection.

It is unknown whether river otters (*Lutra canadensis*; Mustelidae) are a potential source of toxoplasmosis for humans or other animals exposed to infected otter tissue. A cross sectional study was conducted to determine the prevalence of antibodies to *T. gondii* in a population of live-trapped river otters from central eastern North Carolina (USA).

River otters ( $n = 103$ ; 49 adult males, 29 adult females, 13 juvenile males, 12 juvenile females) were live-trapped using modified and standard leg-hold traps of various sizes. The study site consisted of 10 counties in eastern North Carolina (37°08'N to 34°17'N, 75°53'W to 78°18'W). Otters were trapped by wildlife biologists and cooperating local trappers for the North Carolina Wildlife Resources Commission Otter Translocation Project (Raleigh, North Carolina, USA) from December 1995 through February 1996 in compliance with the Commission's Animal Care and Use Committee. Otter husbandry during the holding period has been described (Spelman et al., 1993). Maturity class, defined as adult >1 yr and juvenile <1 yr, was subjectively determined by body weight, tooth size and tooth wear.

The otters were weighed, then anesthetized with an intramuscular injection of either ketamine hydrochloride 10 mg/kg (100 mg/ml, Ketaset®, Aveco Co., Fort Dodge, Iowa, USA), ketamine hydrochloride in combination with midazolam hydrochloride 0.25 mg/kg (5 mg/ml, Versed®, Hoffman-LaRoche, Nutley, New Jersey, USA) (Spelman et al., 1993), or a combination of tiletamine hydrochloride and zolazepam hydrochloride 4 mg/kg (100 mg/ml total, Telazol®, Fort Dodge Labo-

TABLE 1. Prevalence of antibodies to *Toxoplasma gondii* in river otters from eastern North Carolina by age group.

	N	Positive titer	Prevalence (%)	Titer dilution								
				1:16	1:32	1:64	1:128	1:256	1:512	1:1,024	1:2,048	1:>2,048
Total	103	46	45	3	5	5	2	8	5	6	8	4
Adults	78	37	47	3	3	3	2	8	4	5	5	4
Juveniles	25	9	36	0	2	2	0	0	1	1	3	0

ratories Inc., Fort Dodge, Iowa, USA). Once anesthetized, the animals were placed in dorsal recumbency and monitored by rectal temperature, heart rate, respiration rate and pulse oximetry. Physical examinations were conducted, biological specimens taken, and trap wounds treated if present. Blood samples were taken from the jugular vein using a 12 ml syringe and 20 ga needle then placed in a serum separator vacutainer tube (Vacutainer®, Becton Dickson, Franklin Lakes, New Jersey, USA). The serum separator tube was allowed to stand at room temperature to clot for 30 min, then centrifuged for 10 min. The serum was aliquoted into three cryogenic vials (Nalge Co., Rochester, New York, USA) and placed on ice until frozen at  $-70^{\circ}\text{C}$  within 10 hr.

Aliquots of serum were thawed and screened for the presence of antibodies to *T. gondii* using a commercially prepared indirect latex agglutination kit (Toxotest-MT

“Eiken”, Tanabe USA, Inc., San Diego, California, USA). The kit uses inactivated *T. gondii* antigen coated latex particles that agglutinate when exposed to *T. gondii* specific antibodies. Sera were diluted 1:16 in buffer solution and assayed in duplicate wells following manufacturer’s instructions. Sera positive at 1:16 were serially diluted from 1:16 to 1:2,048 and retested to determine antibody titers. A titer  $\geq 1:16$  was considered positive for *T. gondii* specific antibodies. While the test was not specifically validated for use in otters, agglutination assays are generally not species-specific. The seroprevalence of *T. gondii* antibodies were compared by maturity class (adult, juvenile), by sex within the maturity class, and by county of capture using Chi-square analysis (Glantz, 1992) considering an  $\alpha \leq 0.05$  as significant.

The seroprevalence of *T. gondii*-specific antibodies was 45% (46/103 otters) (Table 1). Positive titers ranged from 1:16 to greater than the maximum tested dilution of 1:2,048. Antibodies were slightly more common in adults, but statistically significant differences were not found between the maturity classes or between sexes within each maturity class. Among counties with positive antibody titers, there were no significant differences in antibody prevalences (Table 2). Sufficient sample size was not available from each county to rigorously examine geographic data pattern by county within the study area. However, a pattern of higher prevalence along the Neuse River flowing into the Pamlico Estuary emerges on geographical inspection of the data. The Neuse River/Estuary system encompasses approximately one sixth

TABLE 2. Prevalence of antibodies to *Toxoplasma gondii* in river otters from eastern North Carolina by county of capture.

County of capture	Number tested (N)	Positive titer $\geq 1:16$ (N)	Prevalence <i>T. gondii</i> antibodies (%)
Beaufort	10	7	70
Carteret	7	0	0
Craven	12	9	75
Edgecomb	1	0	0
Hyde	9	6	67
Lenoir	3	2	67
Onslow	17	6	35
Pamlico	25	9	36
Pender	17	6	35
Wayne	2	1	50

of the population of North Carolina and has experienced a large population growth rate over the past 10 yr (North Carolina Division of Environmental Management, 1993).

Transmission of *T. gondii* occurs transplacentally, after ingesting raw or undercooked infected meat, or drinking water contaminated with oocysts (Dubey, 1994a). Wildlife can serve as a source of human infection if infected tissues are ingested or from poor hygiene when dressing killed game or fur bearing animals. Discarded meat scraps and viscera can contain *T. gondii* and result in tissue infection when ingested by scavenger animals (Briscoe, 1993; Dubey, 1994b). When definitive felid hosts eat infected meat, the wildlife cycle is continued by the passage of oocysts into the environment resulting in contamination of food and water supplies for other wildlife, livestock or humans.

Although river otters are primarily piscivorous and aquatic, they opportunistically eat birds and mammals (Greer, 1959; Serfass et al., 1990). The prevalence of *T. gondii* antibodies in the population of river otters we studied is comparable to the prevalence of antibodies published for terrestrial mustelids such as mink (55%) and fishers (41%) (Tizard et al., 1976), but is considerably higher than those reported in aquatic herbivores like nutria (7%) (Howarth et al., 1994), muskrat (17%) or beaver (7%) (Smith and Frenkel, 1995). Because the carnivorous dietary habit of the otter includes birds and mammals, similar to terrestrial mustelids such as mink and fishers, the likelihood of their exposure to *T. gondii* is greater than that of the aquatic herbivores.

We did not attempt to isolate *T. gondii* from exposed individuals and the experimental design of the present study precluded determination of rising titers which would indicate current infection. However, the organisms can be isolated by bioassay from the majority of other host species when serum antibodies are detected

(Dubey and Beattie, 1988). The river otter is hunted for its pelt in North Carolina. The *T. gondii* antibody prevalence of 45% in otters from eastern North Carolina found in this study suggests trappers could be at risk of contracting toxoplasmosis if caution is not exercised. Skinned otter carcasses should be handled and disposed of properly to prevent spread of the disease to humans or scavenger animals.

The authors thank the North Carolina State University veterinary student otter team volunteers including A. Koenig, C. Smith, L. Reiff, L. Rozear, S. White, J. Bowman, M. Crosier, J. Neal, E. Brumback, J. Geyer, W. Giambo, L. Harrison, M. Roher, S. Rogers, K. Sakamoto, J. Smith and J. Ward for their assistance in specimen collection; L. Reynolds and M. Spilken for technical assistance; and C. Harms for review of the manuscript.

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*Received for publication 2 September 1996.*