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Surveillance for *Toxoplasma gondii* in the white-tailed deer (*Odocoileus virginianus*) in Ohio¹

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ABSTRACT. Serum samples from 147 white-tailed deer, *Odocoileus virginianus*, were collected at deer-check stations and Columbus area metropolitan park deer hunts during November and December of Ohio's 1996-97 and 97-98 hunting seasons. These samples were tested for *Toxoplasma gondii* antibodies using a modified direct agglutination assay. Forty-four percent (65/147) of the samples tested positive. Sixteen percent had a titer of 25, 16% had a titer of 50, 3% had a titer of 250 and 9% had a titer of ≥ 500 . Percentage of positive deer varied greatly between geographical locations. Fifty-five percent of 45 serum samples collected from Hocking County were positive while only 6% of the 18 deer sampled from Franklin County tested positive. No significant differences in infection rates were observed between sexes. This is the first report of *T. gondii* antibodies from a game animal in Ohio. Pregnant women should thoroughly cook venison before it is consumed to avoid complications from this pathogen.

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INTRODUCTION

The intracellular protozoan *Toxoplasma gondii* (Luo and others 1995) is widespread in humans and many other species of warm-blooded animals (Dubey and Beattie 1988). Because it has low host specificity, *T. gondii* will infect most mammals (Bakht and Gentry 1992), including white-tailed deer and other ungulates such as black-tailed deer (*Odocoileus hemionus*), California mule deer (*O. hemionus californicus*), Rocky Mountain mule deer (*O. hemionus hemionus*) and southern mule deer (*O. hemionus fuliginatus*) (Chomel and others 1994). Ohio's deer population numbers approximately 500,000 and, on average, hunters harvest 158,000 each season (Ohio Division Wildlife 1997). Although *T. gondii* antibodies have been found in white-tailed deer from Pennsylvania, New York, Florida (Humphreys and others 1995), Minnesota (Vanek and others 1996) and Alabama (Lindsay and others 1991), no surveys have been performed in Ohio.

Toxoplasma gondii is transmitted in three ways: ingestion of food or water containing oocysts from infected cats, ingestion of undercooked meat containing tissue cysts (Dubey 1996), and through congenital transmission from a pregnant woman to her developing fetus (Engeland and others 1996). *Toxoplasma gondii* is found encysted in tissues of wild animals and, more frequently, in domestic animals, such as cats. Meat from wild game (Vanek and others 1996) and ingestion of shed oocysts from cat feces (Engeland and others 1996) are the two most likely sources of human infection. Approximately 30% of the human US population has *T. gondii* antibodies, and infection is even more prevalent in Europe and Central and South America with rates of 50% to 80% (Gardiner and others 1988), due to the common practice of eating undercooked meat (Dubey 1996).

Human infection with *T. gondii* often causes a disease called toxoplasmosis if the host is immunocompromised (Dubey and Beattie 1988). Toxoplasmosis can result in infant mortality, severe mental retardation (Vanek and others 1996) and vision loss when children are infected *in-utero* (Dubey 1996). Between 0.1 and 0.8% of all pregnant women acquire toxoplasmosis and approximately 40% of these transfer it to their fetuses (Bakht and Gentry 1992). Of the babies born, 26% are sub-clinically affected, 10% clinically affected and 3% die (Gardiner and others 1988). Toxoplasmosis also causes fatal encephalitis in immunocompromised patients. An estimated 3-20% of all AIDS patients die of encephalitis caused from toxoplasmosis (Dubey 1996). Our survey was conducted to determine the prevalence of *T. gondii* antibodies in white-tailed deer in central Ohio so potential risk, from ingestion of undercooked meat, could be evaluated.

MATERIALS AND METHODS

During the 1996 and 1997 fall Ohio hunting seasons, blood (5 to 15 ml) was collected from each of 147 deer at check stations in Delaware, Fairfield and Portage counties and during several Franklin County Metropolitan Parks deer management hunts. These county check stations accept deer from adjacent counties and were used as they have a historically high census. Blood was removed with a pipette from the thoracic cavity, placed in screw top polypropylene vials and refrigerated until transported to The Ohio State University, Columbus, OH. Blood was then centrifuged, serum removed and stored (-20° C) until tested.

Sera were examined for *T. gondii* antibodies by a direct agglutination test (Dubey and Desmonts 1987) at the Acarology Laboratory, Ohio State. Formalin fixed whole tachyzoites, obtained from the Institute de Puericulture in Paris, France provided by J. P. Dubey (Parasite Biology and Epidemiology Laboratory, Beltsville, MD), functioned as the antigen substrate. Sera were diluted at 1:25, 1:50, 1:250 and 1:500 in a pH 7.2

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phosphate-buffered saline. Each well of a 96-well u-bottom plate received 25 µl of diluted sera and 25 µl of antigen containing 2.5 ml of antigen diluting buffer, 35 µl 2-mercaptoethanol, 50 µl Evans blue dye solution and 0.15 ml of antigen. Positive and negative domestic pig controls (provided by J. P. Dubey) were included on each plate. Plates were then sealed with tape, incubated (37° C) overnight and read by the same individual. A blue button at the bottom of the well was read as negative while a diffuse color was positive. A chi-square goodness of fit test was utilized for all statistical analysis.

RESULTS

Sixty-five of the 147 (44%) white-tailed deer sampled during the 1996 and 1997 hunting seasons exhibited antibody presence to *T. gondii* (Table 1). Antibodies were found in sera from deer obtained in 7 of the 8 counties sampled: Delaware, Fairfield, Franklin, Geauga, Hocking, Perry and Portage. Percent positive from each county varied, ranging from 6-100%, although sample sizes from several counties were small (see Table 1). The percentage of positive animals from each county is depicted in Figure 1. In Franklin County, where 15 deer came from 2 metroparks, (High Banks and Sharon Woods), only 1 sample tested positive. This proportion of 6.7% was significantly lower than the proportion found positive in the other counties sampled ($\chi^2 = 6.39$, $df = 64$, $p < 0.05$). There was no significant difference for antibody levels between male 22/58 (37%) and female 43/89 (48%) deer ($\chi^2 = 0.86$, $df = 64$, $p > 0.05$).

DISCUSSION

In adult animals *T. gondii* infection occurs primarily through ingestion of oocysts shed by feline feces and



FIGURE 1. Geographical location and percent antibody positives for *T. gondii* in Ohio deer collected during 1996-97 as shown in Table 1.

through the consumption of infected meat (Dubey and Beattie 1988). Because deer are wild herbivores and do not spend significant amounts of time near domestic cats, one would expect infection rates to be low. However, the results of this study demonstrate that a significant proportion (44%) of hunter-killed deer have had an infection with *T. gondii*. High sero-positivity has been

TABLE 1

Number of deer tested, percent positive, and Ohio county where *T. gondii* antibodies were detected in deer sera.

County	No. Tested	No. Sero Positive (≥25)	% Sero Positive (≥25)	No. Positive at titer of				
				<25	25	50	250	(≥500)
Delaware	28	11	39%	17	2	4	3	2
Fairfield	61	29	48%	32	14	7	2	6
Franklin	17	1	6%	16	0	1	0	0
Gauga	1	1	100%	0	0	1	0	0
Hocking	29	16	55%	13	6	8	0	2
Knox	2	0	0%	2	0	0	0	0
Perry	1	1	100%	0	1	0	0	0
Portage	8	6	75%	2	0	2	0	4
Totals	147	65	44%	82	23	23	5	14

reported in deer and other herbivorous food animals (Vanek and others 1996; Stewart and others 1995; Dubey 1985; Dubey, Rickard and others 1992; Dubey, Brown and others 1992; Lindsay and others 1991; Chomel and others 1994; Brillhart and others 1994; Humphreys and others 1995) and represents an often unrecognized risk associated with eating undercooked meat. Although no isolation was attempted in this study, *T. gondii* tachyzoites have been isolated from tissues of naturally infected deer and domestic cattle (Lindsay and others 1991; Dubey 1992). Antibody presence indicates present or past exposure to the pathogen.

Since deer do not eat meat, the only source of infection is through oocysts shed in feline feces. Oocysts persist in nature for several months (CDC&P 1998). They have been identified on vegetation (Dubey and Beattie 1988) and in soil (Dubey and others 1995). The sole wild feline in Ohio is the bobcat, *Lynx rufus*. Although the bobcat's population is increasing, it is considered rare and has been positively identified in only 16 of the 88 counties (Stoll 1997). Bobcats are established in two counties where deer sera were collected: Fairfield and Hocking. The bobcat may have been the source of infection for antibody positive deer in those counties. Positive serum samples collected from the other six counties must have acquired infection through oocysts shed from infected domestic or feral felines. The low sero-positive percentage in Franklin County (6%) is unexpected since isolated deer herds in metro parks (High Banks and Sharron Woods) should have had more contact with domestic cats.

Pathogenic effects resulting from toxoplasmosis in immunocompetent adults are usually undetected (Feldman 1982). However, severe outbreaks from eating uncooked meat have occurred (Choi and others 1997). Unilateral chorioretinitis and lymphadenopathy (usually in the neck and head) have been observed when immunocompetent patients ingest large amounts of *T. gondii* tachyzoites (McCabe and others 1987; Choi and others 1997). In immunocompromised patients, infection usually begins with fatigue and general malaise and may be associated with fever and rashes lasting up to two weeks (Dubey and Beattie 1988). Congenital infection with *T. gondii* can yield extremely pathogenic effects on the fetus depending on the time the mother is infected. Congenital infection occurs most easily during the third trimester and pathogenic effects are not severe. Although much less likely, congenital infection during the first trimester can result in abortion (Remington and Desmonts 1983).

Approximately 158,000 deer were killed by hunters in Ohio during 1996 most likely for consumption (ODW 1997) and 44% of the deer tested in this study had antibodies to *T. gondii*. Therefore, individuals ingesting deer meat from Ohio should take appropriate precautions to avoid infection. Freezing (-12.37° C) (Kotula and others 1991), exposure to at least 67° C for 3 minutes (Dubey and others 1990), or exposing meat to gamma irradiation (0.5 kGy) (Dubey 1996) kills tachyzoites. Microwave cooking is not an effective method because food is not cooked evenly (Dubey and Beattie

1988). To avoid infection from cat feces, hands should be washed and air-borne particles avoided when emptying litter. Pregnant women should take special precautions by not emptying litter pans, eating well-cooked meats and not allowing domestic cats outside where they may acquire new infection.

To our knowledge, no other serological or isolation evidence of *T. gondii* has been reported from wild game animals in Ohio. Despite our small sample size in several counties, as shown in Table 1, antibodies to *T. gondii* are present in Ohio deer. Precautions to avoid infection should be followed when processing or ingesting meat from all deer. Further studies are needed to better understand the epizootic/epidemic nature of this disease in game animals and families of hunters who consume wild meats.

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The 1998 Paper of the Year Award

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