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Seroprevalence of *Toxoplasma gondii* and *Trichinella spiralis* in North Carolina Black Bears (*Ursus americanus*)

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ABSTRACT: Serum samples from 143 hunter-killed black bears were collected during the 1996 and 1997 black bear hunting seasons in eastern North Carolina. All samples were tested for antibodies to *Toxoplasma gondii* by the modified agglutination test. Antibodies to *T. gondii* were present in 120 of 143 (84%) bears. Females had significantly higher titers than males (Wilcoxon rank sums test, $P = 0.045$), and titers increased with age (Jonckheere test, $P = 0.01$). Samples collected during 1996 ($n = 79$) were tested for antibodies to *Trichinella spiralis* by enzyme-linked immunosorbent assay. No samples were positive for antibodies to *T. spiralis*.

Toxoplasma gondii and *Trichinella spiralis* can cause severe disease in humans. Both can be acquired by consuming infective stages in undercooked meat. *Toxoplasma gondii* can also be contracted transplacentally or by consumption of food or water contaminated with oocysts (Dubey and Beattie, 1988). Historically, domestic animals have been considered the primary reservoir hosts of these parasites for humans, but wildlife also play a role as sources of both human and domestic animal infection.

Numerous point-source outbreaks of human trichinellosis have been traced to bear meat (Wand and Lyman, 1972), and recently black bears (*Ursus americanus*) from Pennsylvania were reported to have 80% seroprevalence for *T. gondii* (Briscoe et al., 1993). Viable *T. gondii* was isolated from the hearts of 10 of 22 seropositive bears (Dubey et al., 1995). However, the epizootiology of these parasites in black bears is not well understood. Opportunistic sampling of hunter-killed black bears in eastern North Carolina was undertaken to evaluate seroprevalence of *T. gondii* and *T. spiralis*. Hunters were also surveyed to investigate the disposition of the carcasses.

Blood samples were collected from the hearts of intact carcasses or from the body cavities of dressed carcasses of bears illed in Hyde County (35°43′N to 35°04′N, 75°45′W to '39'W) and Tyrrell County (36°00′N to 35°34′N, 75°55′W to '38'W), North Carolina. From 11 November through 16 No-ember 1996, 79 samples were collected (14 female, 65 male). rom 10 November through 15 November 1997, 64 samples ere collected (26 female, 38 male). Blood samples were laced in sterile 50-ml conical polypropylene centrifuge tubes nd stored on ice until transported and processed at North Car-lina State University, Raleigh, North Carolina. Blood was cen-ifuged and serum removed and frozen in polypropylene cry-preservation tubes at −20 C within 24 hr of collection. Gen-ler, weight (intact carcasses only), and location of kill were ecorded for each bear. A first premolar tooth was collected for ectioning and analysis of cementum annuli for age determi-nation (Craighead et al., 1970). In addition, in 1996 a brief erbal survey was conducted with each hunter to collect infor-mation on intended uses of the carcass, sharing of meat, and meat storage and cooking methods (n = 79 complete surveys).

Frozen serum samples were sent to the Parasite Biology and Epidemiology Laboratory of the United States Department of Agriculture (USDA), Beltsville, Maryland, for analysis. Sam-ples from 1996 and 1997 (n = 143) were tested for antibodies to *T. gondii* at dilutions of 1:25, 1:50, and 1:500 by the modified agglutination test (MAT) as described by Dubey and Desmonts (1987). Based on experience with MAT testing of human and animal sera, titers of ≥1:25 were considered positive (Dubey et al., 1994, 1995). Samples collected in 1996 (n = 79) were also tested for antibodies to *T. spiralis* by enzyme-linked im-munosorbent assay (ELISA) (Dubey et al., 1994, 1995). Posi-tive and negative controls were included in each test and all samples were run by a single individual.

The Wilcoxon rank sum test (Hollander and Wolfe, 1973) was used to compare the *T. gondii* titers in male and female bears. The Jonckheere test for ordered alternatives was used to assess the association between age and titer (Hollander and Wolfe, 1973).

None of the 1996 samples was positive for antibodies to *T. spiralis*, and, therefore, further testing was not pursued in 1997. Antibodies to *T. gondii* were found in 84% (120 of 143) of sera tested. Prevalence of antibodies at the various terminal dilutions was 3.5% at 1:25 (n = 5), 61% at 1:50 (n = 87), and 19.5% at 1:500 (n = 28), while 16% (n = 23) of samples were neg-ative. A significantly greater number of females than males were seropositive at the higher dilutions (Wilcoxon rank sums test, $P = 0.045$). Titers also increased with age (Jonckheere test, $P = 0.01$).

All hunters associated with the 79 bears sampled in 1996 completed questionnaires. Ninety-six percent of the hunters butchered the carcasses themselves and none wore gloves while skinning or handling the carcasses. All of the hunters surveyed consume the bear meat or share with others who do, and 20% prepare the meat in a manner that may be inadequate to inac-tivate infective stages of *T. gondii* or *T. spiralis,* such as pre-paring jerky or not cooking the meat thoroughly. Although no questions addressed the immunocompetency of the hunters sur-veyed, 3 volunteered information that they were immunocom-promised, 1 due to immunosuppressive therapy following organ transplantation, and 2 due to cancer chemotherapy.

Although human trichinellosis is usually attributed to the in the prevalence of trichinellosis in the U.S. swine population has been documented from about 1.4% at the turn of the cen-tury to 0.125% by the 1970s (Zimmerman, 1977). During the same period, the role of wildlife, particularly bears, as a major sylvatic reservoir of the disease and important source of human infection was recognized (Bailey and Schantz, 1990). Between 1982 and 1986, 14.6% of trichinellosis cases in the United States were traced to infected bear meat (Bailey and Schantz, 1990). The reported prevalence of *T. spiralis* in North American black bears ranges from 1.8% in Pennsylvania to 15.6% in Montana (Worley et al., 1991; Dubey et al., 1994).

The lack of antibodies to *T. spiralis* in this study was not anticipated. The sample size was relatively small and limited in geographic distribution and may not have had sufficient power to detect low seroprevalence. Alternatively, the absence of an-tibodies in black bears may reflect the historically low preva-lence of *T. spiralis* in the North Carolina swine herd and the low numbers of commercial swine operations in the region sur-veyed (Cowen et al., 1990). Domestic swine have long been considered the major source of infection for wildlife that scav-enge improperly dumped swine carcasses or scraps and refuse at garbage dumps. However, recent surveys of wildlife in Eu-rope and the United States suggest that a sylvatic cycle of *Trichinella* infection operates largely independent from the do-mestic pig cycle (Pozio et al., 1992; Snyder et al., 1993). These studies also suggest that the dominant *Trichinella* species in wildlife may be sylvatic genotypes *Trichinella* T5 and T6 (Wor-ley et al., 1990; Zarlenga et al., 1991).

Toxoplasmosis is a serious disease of humans and livestock, although infections in immunocompetent individuals are rare. Congenital infectious can cause disease in the developing fetus, resulting in hydrocephaly, mental retardation, and other birth defects (Dubey and Beattie, 1988). Reported seroprevalence, by a variety of tests, for *T. gondii* in North American black bears has ranged from 8% to 80% across different regions of the country (Binninger et al., 1980; Briscoe et al., 1993). The re-sults of this study are comparable to those recently reported for Pennsylvania black bears using the identical serologic test (Briscoe et al., 1993) and confirm that black bears have the highest seroprevalence of any animal or human population test-ed to date in the United States. While no testing for parasite burdens was undertaken for this study, previous work has shown the MAT to be the most consistent and reliable serologic test in black bears (Dubey et al., 1994, 1995). Sensitivity and specificity of the MAT in bears have not been fully evaluated but are established for pigs (Dubey, 1997; Dubey et al., 1997).

As omnivores, black bears consume both forage and foraging animals, as well as carrion and garbage. This may contribute to their high *T. gondii* seroprevalence, although the prevalence in North Carolina and Pennsylvania black bears is much higher than reported in other wild omnivores. A 1990 survey of free-ranging wild hogs (*Sus scrofa*) in the Great Smokey Mountains National Park, reported 31% seroprevalence by MAT (Diderrich et al., 1996). A serosurvey of raccoons (*Procyon lotor*) from 7 states reported a mean prevalence of 50%, also by MAT (Dubey et al., 1992). The positive relationship between titer and the age of the bears suggests that the high seroprevalence may be due to cumulative exposure and supports similar observations from other studies (Briscoe et al., 1993; Zarnke et al., 1997). Signifbias introduced by the hunters. Larger, heavier bears are favored targets, and accordingly male bears are overrepresented in the sample. The larger female bears that are also favored by hunters tend to be older and, as previously demonstrated, titers increase with age.

The hunter survey suggests that bear hunters and those who handle and consume bear meat should be educated about the potential risks of exposure to *T. gondii* and how to minimize them. Such education could be accomplished by inclusion of a pamphlet on safe carcass handling with hunting licenses or dissemination of similar material to hunting clubs or hunting supply retailers. Important points to address include: (1) wearing gloves while butchering carcasses, (2) washing hands thoroughly with soap and water after handling bear meat, (3) cooking bear meat until welldone (to an internal temperature of 66 C for at least 3 min), and (4) burying viscera and carcass scraps to reduce scavenging by other animals.

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Hosts Manipulated by One Parasite Incur Additional Costs from Infection by Another Parasite

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ABSTRACT: Manipulation of host phenotype by parasites often serves to increase the predation rate of definitive hosts on intermediate hosts. For intermediate hosts, the indirect consequences of manipulation may extend beyond the direct increase in predation, however. Metacercariae of the trematode *Curtuteria australis* encyst in the foot of New Zealand cockles, *Austrovenus stutchburyi,* and stunt its growth, rendering cockles incapable of burrowing into the sediments. Here, we show that cockles manipulated by *C. australis* are 5 times more likely to be infected by the castrating sporocysts of another trematode than normal, nonmanipulated cockles. Our results indicate that the consequences for *C. australis*-manipulated cockles are far more important than a simple increase in the risk of predation