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Echinococcus granulosus in Gray Wolves and Ungulates in Idaho and Montana, USA

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ABSTRACT: We evaluated the small intestines of 123 gray wolves (Canis lupus) that were collected from Idaho, USA (n=63), and Montana, USA (n=60), between 2006 and 2008 for the tapeworm Echinococcus granulosus. The tapeworm was detected in 39 of 63 wolves (62%) in Idaho, USA, and 38 of 60 wolves (63%) in Montana, USA. The detection of thousands of tapeworms per wolf was a common finding. In Idaho, USA, hydatid cysts, the intermediate form of E. granulosus, were detected in elk (Cervus elaphus), mule deer (Odocoileus hemionus), and a mountain goat (Oreamnos americanus). In Montana, USA, hydatid cysts were detected in elk. To our knowledge, this is the first report of adult E. granulosus in Idaho, USA, or Montana, USA. It is unknown whether the parasite was introduced into Idaho, USA, and southwestern Montana, USA, with the importation of wolves from Alberta, Canada, or British Columbia, Canada, into Yellowstone National Park, Wyoming, USA, and central Idaho, USA, in 1995 and 1996, or whether the parasite has always been present in other carnivore hosts, and wolves became a new definitive host. Based on our results, the parasite is now well established in wolves in these states and is documented in elk, mule deer, and a mountain goat as intermediate hosts.

Key words: Canis lupus, Echinococcus granulosus, survey, tapeworm, wolves.

Echinococcus granulosus is a minute tapeworm that is approximately 3 mm in length and requires two mammalian hosts to complete the life cycle. Genetic heterogeneity is common in *E. granulosus*, and at least 10 strains of the parasite have been identified (Jenkins et al., 2005). In North America, adult *E. granulosus* is found primarily in wolves (*Canis lupus*), coyotes (*Canis latrans*), and domestic dogs (*Canis lupus familiaris*). The intermediate form of the tapeworm is a hydatid cyst (metacestode), which is a fluid-filled cyst containing numerous immature forms of the tapeworm called protoscoleces. Hydatid cysts are found primarily in the lungs and liver of wild, even-toed, and domestic ungulates but will also develop in certain other animals, including humans. Hydatid disease in humans, caused by specific strains of E. granulosus, is an important zoonotic disease. Comprehensive reviews of E. granulosus and hydatid disease have been published (Thompson and Lymbery, 1995; Jones and Pybus, 2001; Jenkins et al., 2005).

In 1995 and 1996, 66 wolves were reintroduced from Alberta, Canada, and British Columbia, Canada, into central Idaho, USA (n=35), and into Yellowstone National Park, Wyoming, USA (n=31). At the end of 2007, there were an estimated 1,500 wolves in Idaho, Montana, and Wyoming, USA (Sime et al., 2008).

In North America, adult *E. granulosus* in wolves has been reported previously from Alaska, USA; Minnesota, USA; Alberta, Canada; British Columbia, Canada; Northwest Territories, Canada; Ontario, Canada; and Yukon Territory, Canada (Choquette et al., 1973, and reviewed by Leiby and Dyer, 1971; Jones and Pybus, 2001), but recent surveys within the past 20 yr are lacking. Hydatid cysts have been reported from moose (*Alces alces*), elk or wapiti (*Cervus elaphus*), mule deer (*Odocoileus hemionus*), white-tailed deer (*Odocoileus virginianus*), caribou (*Rangifer*)

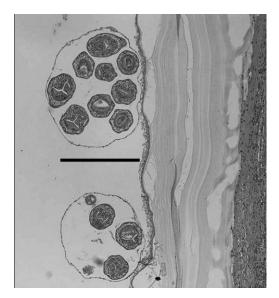


FIGURE 1. Small section of a hydatid cyst of *Echinococcus granulosus* from the lung of a mountain goat, illustrating the thick laminated wall and two internal brood capsules filled with protoscoleces. Hematoxylin and eosin stain. Bar= $500 \mu m$.

tarandus), and one mountain goat (*Oreamnos americanus*) (reviewed by Leiby and Dyer, 1971; Jones and Pybus, 2001). The purpose of this study was to determine the prevalence of adult *E. granulosus* in wolves in Idaho, USA, and Montana, USA, and to document the occurrence of hydatid cysts in wild ungulates.

On 23 January 2006, a mountain goat was found dead in Elmore County in central Idaho, USA (43°49'N, $115^{\circ}06'$ W). The carcass was emaciated and partially scavenged. Two round, fluid-filled cystic structures, approximately 5 cm by 4.5 cm were removed from the lungs and placed in 10% formalin for evaluation. The cysts were sectioned at $5 \,\mu m$ and stained in hematoxylin and eosin for microscopic evaluation. Microscopic evaluation of the cysts indicated a laminated cyst wall and numerous internal brood capsules that were up to 520 µm in diameter, and each brood capsule contained groupings of 2-14 tapeworm protoscoleces with prominent refractile hooks and calcareous cor-

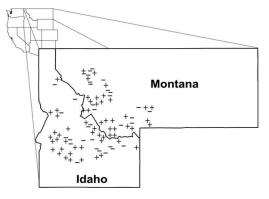


FIGURE 2. Locations of 90 wolves from Idaho, USA, and Montana, USA, that were evaluated for *Echinococcus granulosus*. + = positive wolves, - = negative wolves.

puscles (Fig. 1). Based on morphologic characteristics, the cysts were identified as hydatid cysts, the intermediate form of *E. granulosus*. This finding, therefore, stimulated a survey of gray wolves, the usual definitive host of *E. granulosus*, in Idaho, USA, and Montana, USA.

A total of 123 intact small intestines from wolves collected in Idaho, USA (n=63), and Montana, USA (n=60), in 2006 to 2008, were evaluated for E. granulosus. Ninety wolves with known collection locations are plotted in Figure 2. Wolves from Idaho, USA, with age and sex data included 21 adult males, 12 adult females, 10 subadult males, and five subadult females (n=48). Wolves from Montana, USA, with age and sex data, included 18 adult males, 0 adult females, eight subadult males, and nine subadult females (n=45). Some wolves had only age or sex data. Wolves were collected primarily by personnel in the Wildlife Services, US Department of Agriculture, when wolves were involved in livestock depredation situations. Other wolves were found dead, were illegal kills, or had an unknown cause of death. Intestines were removed intact by field or laboratory personnel and frozen. For evaluation, each small intestine was thawed, opened in water in a dish pan, and rinsed thoroughly. After excess supernatant was decanted,

intestinal contents were preserved in 10% formalin for a total volume of 700 ml until they were evaluated for tapeworms. A 35ml aliquot (5%) was examined under $30 \times$ magnification using a dissecting microscope, and the numbers of E. granulosus were counted. We used the following scale for *E. granulosus* numbers recovered from a 5% aliquot: 0; +=<100; ++=100 to 1,000; and +++=>1,000. When thousands of tapeworms were detected in the aliquot, the +++ designation was used without counting all the tapeworms. Representative adult specimens of E. granulosus (US National Parasite Collection [USNPC] 100986 for Montana-infected wolves and USNPC 100987 for Idaho-infected wolves) were deposited in the USNPC (Agricultural Research Service, US Department of Agriculture, Beltsville, Maryland, USA). Chi-squared P values were calculated using software Epi Info (Dean et al., 1994) to determine significant differences among proportions of positive wolves in Idaho and Montana, male and female, and adult and subadult wolves. Wolves were considered adults at 2 yr of age.

Echinococcus granulosus was detected in 39 of 63 wolves (62%) in Idaho, USA, and 38 of 60 wolves in Montana, USA $(63\%; \chi^2, P=0.87)$. In both states, 41 of 63 adult wolves (65%) were positive, compared with 20 of 34 subadult wolves (58%; χ^2 , P=0.54). In both states, 37 of 63 male wolves (59%) were positive compared with 33 of 49 female wolves (67%; χ^2 , P=0.35). The detection of thousands of tapeworms per wolf was a common finding. Numbers of E. granulosus recovered were similar in both states. In Idahoinfected wolves, 21 of 39 (54%) had >1,000 tapeworms per 5% aliquot; 4 of 39 (10%) had 100 to 1,000 tapeworms; and 14 of 39 (36%) had <100 tapeworms. In Montana-infected wolves, 20 of 38 (53%) had >1,000; 6 of 38 (16%) had 100 to 1,000 tapeworms; and 12 of 38 (32%) had <100 tapeworms. Other parasites detected without counting the worms were *Taenia* spp. in 71% of all wolves and ascarids (*Toxocara* sp. or *Toxascaris* sp.) in 8% of all wolves. During the study period, hydatid cysts, the intermediate form of *E. granulosus*, were detected in elk, mule deer, and a mountain goat (Fig. 2) in Idaho, USA, and in elk in Montana, USA.

To our knowledge adult E. granulosus or hydatid cysts in Idaho, USA, or Montana, USA, have not been reported in the literature. The mountain goat, as an intermediate host of E. granulosus, has only been reported once before in one animal from Alaska, USA (Rausch and Williams, 1959) and likely is an uncommon intermediate host. To determine whether E. granulosus is being transmitted in Idaho and Montana, USA, we conducted a survey of the major definitive host for E. granulosus, the wolf. Earlier studies in Canada and in Alaska and Minnesota, USA, have reported prevalences of E. granulosus in wolves from 14% to 72% (Leiby and Dyer, 1971; Choquette et al., 1973, McNeill et al., 1984). Our results of a 63% prevalence are similar to most other studies in wolves.

There are generally two recognized biotypes of E. granulosus in North America (Rausch, 1986, 2003; Thompson and Lymbery, 1990). The northern or sylvatic biotype generally has a canine definitive host (wolf, dog) and a cervid intermediate host (moose, elk, deer, caribou). It is characteristically found above latitude 48° north in the Holarctic zones of tundra and boreal forest. The human infection produced by the northern biotype is relatively benign (Rausch, 2003). Most human cases of hydatid disease have been detected in indigenous peoples who hunt wild cervids or are reindeer herders with dogs. In humans, the cysts are primarily in the lungs, and the cysts often have poorly developed cyst walls, few or no protoscoleces, and often the infections resolve via rupture and expulsion (Meltzer et al., 1956; Wilson et al., 1968). Therefore, the probability of fatal human infections resulting from the wolf-ungulate cycle in Idaho, USA, and Montana, USA, is very low. The northern biotype is differentiated biologically and ecologically from the domestic biotype, and currently, there is no precedent for cross-infection to domestic livestock (Rausch, 1986).

In North America, the domestic biotype circulates in a synanthropic cycle primarily with dogs as the definitive host, and domestic ungulates, especially sheep, as intermediate hosts. It is mainly associated with the sheep-rearing areas in the southwestern United States (Arizona, California, New Mexico, and Utah), but human cases are uncommon and are generally restricted to high-risk groups who have close contact with sheepherding dogs (Schantz, 1995). It has been speculated that the origin of E. granulosus in the western United States was Australian sheep dogs imported into Utah, USA, in 1938 (Crellin et al., 1982). From Utah, USA, the infection apparently spread to adjoining states. The source of E. granulosus in Idaho, USA, and Montana, USA, is unknown, but several scenarios are possible. Wolves that naturally colonized northwestern Montana, USA, from Alberta, Canada, and British Columbia, Canada, may have initiated the infections in ruminants and transplanted wolves, or E. granulosus may have been present in other carnivores, wild ungulates, or domestic sheep before introduction of wolves, and wolves became a new definitive host. Another possibility is that the transplanted wolves initiated the infection in southeastern Montana, USA, and Idaho, USA, and were the source of the current infections being detected. Translocated wolves were treated with praziquantel before they were released in Idaho, USA, and Montana, USA (Johnson, 2001), but there is the possibility that all tapeworms were not eliminated from the treatments.

Domestic dogs are potential definitive hosts for the northern and the domestic biotypes of *E. granulosus*. Results from our study indicate a 63% prevalence in wolves in Idaho, USA, and Montana, USA. We are not aware of any reports of E. granulosus in dogs in Idaho, USA, or Montana, USA, or hydatid cysts in sheep or domestic ruminants in these states. Further evaluations of domestic dogs will determine whether dogs in these states will become infected with this biotype and whether they will also spread E. granulosus to other areas in the western states, where it is currently not present. Because the northern biotype has low pathogenicity in humans, the human health hazard potential is not as important as the domestic biotype, which has more serious ramifications in humans. Further work documenting the potential spread of E. granulosus and characterizing the strains or variants of E. granulosus found in these wolves and ungulates is warranted because strain identification has important implications regarding transmission, zoonotic potential, and control.

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