

1-1-2003

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DEĞERLİ, SERPİL and ÖZÇELİK, SEMRA (2003) "The First Giardia Infection in Cattle Gall Bladder," *Turkish Journal of Veterinary & Animal Sciences*: Vol. 27: No. 5, Article 28. Available at: <https://journals.tubitak.gov.tr/veterinary/vol27/iss5/28>

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The First Giardia Infection in Cattle Gall Bladder

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Received: 30.05.2002

Abstract: Giardia is a ubiquitously distributed flagellate protozoon that inhabits the small intestine of its host. In this paper, Giardia infection is reported in the gall bladder of an adult cow. Faecal samples from asymptomatic dairy cows and calves from several farms in Zara, Sivas, were studied by direct examination and the zinc sulphate flotation techniques. No Giardia cysts were found in 106 samples. In addition, the gall bladders of cattle provided from the slaughterhouse in Sivas were tested by direct and zinc sulphate flotation technique examination. Giardia trophozoites were found in 1.7% of the 56 gall bladders tested. This is the first report of Giardia-infected adult cattle from Turkey. Giardia trophozoites were obtained from fluid and mucosal scrapings from the infected gall bladder. Trophozoites were then cultured in bile-supplemented TYI-S-33 medium and RPMI/1640 containing 10% foetal calf sera and 1% NCTC 135.

Key Words: Giardiasis, cattle, gall bladder

Sığır Safra Kesesinde Görülen İlk Giardia İnfeksiyonu

Özet: Giardia, konağının ince bağırsağına yerleşen ve her yerde görülebilen kamçılı protozoondur. Bu makalede, erişkin bir sığırın safra kesesindeki Giardia enfeksiyonu bildirilmektedir. Çalışmamızda, Sivas'ın Zara ilçesinde farklı çiftliklerden, asemptomatik sığır ve buzağılardan dışkı örnekleri toplandı ve direkt inceleme ve çinko sülfat yüzdürme yöntemi kullanılarak incelendi. 106 dışkı örneğinin hiçbirisinde Giardia kistleri bulunamadı. Ayrıca, Sivas mezbananesinden sığır safra keseleri elde edildi ve direkt mikroskopik inceleme ile incelendi. İncelenen 56 safra kesesi örneğinin 1'inde (% 1,7) Giardia trofozoitleri görüldü. Bu durum, Türkiye'de bildirilen ilk olgudur. İnfekte safra kesesinin sıvısından ve mukozal kazıntılarında Giardia trofozoitleri elde edildi. Daha sonra, safra ile tamamlanmış TYI-S-33 besiyeri ile, % 10 fetal dana serumu ve % 1 NCTC 135 içeren RPMI/1640 besiyerlerinde kültürleri yapıldı.

Anahtar Sözcükler: Giardiasis, sığır, safra kesesi

Introduction

Giardia intestinalis is a cosmopolitan parasite, characteristically more prevalent in children than in adults and more common in warm than cool climates. It is the most commonly diagnosed flagellate of the human intestinal tract (1).

According to numerous protozoological surveys carried out in various regions of Turkey, giardiasis has been found in 11.2% of the adult population and 13.8% of children. High prevalence was dependent on the age group examined, as well as environmental hygiene and climatic conditions (2)

G. intestinalis of human origin can infect other mammals (e.g. dogs, cats, and sheep) (3). Although it has not been found that Giardia of non-human origin can be

infectious to humans, there is some evidence that giardiasis may be a zoonosis (4).

In addition to duodenal involvement, the gall bladder may occasionally become parasitized by *G. intestinalis*. In such individuals, there may be associated gall bladder colic and jaundice, due to obstruction of the bile passages or irritation and oedema of Vater's ampulla (5).

Given that Giardia has the potential to cause clinical disease in cattle and to be transmitted to other animal species and humans, detection of the parasite in cattle may be of major epidemiological significance (6).

While isolated studies have reported giardiasis in cattle and sheep in Africa, Europe and North America, no extensive study of the prevalence of the disease in domestic ruminants has ever been reported.

We coincidentally encountered a cattle gall bladder that was larger than normal size during a study of cystic hidatidosis. When we examined the gall bladder we observed trophozoites of Giardia. We then investigated some 55 more cattle gall bladders obtained from a slaughterhouse. This is the first reported case in Turkey of Giardia infection in the gall bladder of adult cattle.

Materials and Methods

Fresh faecal samples ($n = 106$) were collected from the rectums of cattle on several farms in Zara, Turkey. To avoid cross-contamination, new gloves were used for each sample. One collection was made for each animal. The faecal material was stored at $-20\text{ }^{\circ}\text{C}$ until examination.

The cattle gall bladders were provided from the slaughterhouse in the province of Sivas in Turkey. All samples ($n = 56$) were checked by direct examination for the presence of Giardia trophozoites. The infected adult cow taken to the slaughterhouse for sacrificing, was about 350 kg in weight, and the gall bladder was taken for careful examination as it was larger than average due to the infection. The gall bladder measured approximately 20×15 cm and was determined to contain a litre of fluid. This fluid was collected by injection and immediately investigated under a microscope by placing a drop on a slide.

In addition, mucosal scrapings were obtained with a glass slide from the gall bladder of the cow infected with Giardia, and trophozoites were observed to be actively moving. The trophozoites were then stained with Giemsa.

The gall bladder fluid was directly transferred to culture tubes containing TYI-S-33 medium and RPMI/1640 (containing 10% foetal calf sera, 1% NCTC 135, glutamine and HEPES) and was incubated at $37\text{ }^{\circ}\text{C}$. In addition, antibiotic added bile fluid was also incubated, and the condition of the trophozoites was also observed in this media.

Results

Faecal samples ($n = 106$) provided from cattle on different farms locations in Zara, were tested by zinc flotation technique for giardiasis. No adult cattle or calves tested positive for Giardia cysts. In addition, cattle gall bladders provided from the slaughterhouse in Sivas were

tested by direct examination. Giardia trophozoites were found in 1.7% of the 56 gall bladders.

Under light microscope observation, Giardia trophozoites were seen to be active in the gall bladder fluid and mucosal scrapings. Trophozoites were counted in a haemocytometer chamber and were recorded as 5×10^5 trophozoites/ml. Trophozoites within the samples were cultured in filtre-sterilized TYI-S-33 medium and supplemented with 1 ml penicillin every 2 h for the first 8 h and then daily for the next 2 days. In addition, trophozoites were cultured in RPMI/1640 containing 10% foetal calf sera and 1% NCTC 135. After 24 h, trophozoites started to detach from the culture tube surface and die. By the seventh day of cultivation no living trophozoites were seen.

The mucosal scrapings were evaluated using an Olympus light microscope, and the trophozoites were stained with Giemsa. These trophozoites had typical claw-hammer-shaped median bodies. The detected Giardia trophozoites were identified as belonging to the *G. duodenalis* group. The presence of a typical claw-hammer-shaped median body of the trophozoites further supports our conclusion that the organism found in the domestic ruminants was *G. duodenalis* as described by Filice (7).

Discussion

In Turkey, no detailed report on the investigation of the prevalence of giardiasis in cattle has previously been presented. However, it has been reported that giardiasis is a major cause of diarrhoea in calves. It was postulated that domestic ruminants might be a reservoir for human infection and vice-versa, thus classifying giardiasis a zoonothroponotic disease (8,9).

Faecal samples from asymptomatic dairy cows and calves from a farm in Denmark were examined by sucrose gradient flotation technique by Iburg et al. (6). Giardia cysts were found in 7.6% of the 92 samples.

Buret et al. (4) conducted an investigation to assess the prevalence of giardiasis in domestic ruminants, and the prevalence of infection was found to be 10.4% in cattle. They postulated that the infection rate was significantly higher in young animals than in adult cattle. The high prevalence of giardiasis in sheep and cattle reported in this study and the similarities of Giardia

samples found in these animals to the parasite from humans suggest that domestic ruminants may play a role as a reservoir for human infection and disease.

Occasionally the gall bladder may become parasitized by *Giardia* in humans and other mammals. We found *Giardia* trophozoites in the gall bladder of an adult cow. This is the first such report in Turkey. In addition, faecal samples (n = 106) provided from cattle in different farms in Zara were tested by the zinc flotation technique for giardiasis. No adult cattle and calves tested positive for *Giardia* cysts.

The prevalence of *G. intestinalis* infections was determined in Western Canadian and Western Australian dairy calves by O'Handley et al. (10). Some 58% of Western Australian calves and 57% of Western Canadian calves tested positive for *Giardia*.

In various districts of Switzerland, 815 calves were randomly selected for a single coprological examination for giardiasis by Taminelli and Eckert (11). On average,

26.6% of the calves excreted *Giardia* cysts. They postulated that *Giardia* infections were frequent and geographically widely distributed in calves and lambs in Switzerland.

In Canada, there was only one study reported in the literature on the prevalence of giardiasis. Calves were sampled from 505 dairy farms by Ruest et al. (12) who reported that 45.7% of the farms were found to be positive for *Giardia* spp.

The high *Giardia* infection rates and intensities in calves of a wide age range and the clinical response to metronidazole suggest that *Giardia* infection has contributed to outbreaks of diarrhoea (13).

We did not encounter any information about giardiasis in the gall bladder of animals. For this reason, we believe that the localization of the parasite and the fact that it was observed in an adult cow makes this a very valuable case.

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