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First report of *Cryptosporidium* spp. and *Giardia duodenalis* in calves from northeastern Brazil

Thais FERREIRA FEITOSA, Vinícius LONGO RIBEIRO VILELA*, Ana Célia RODRIGUES ATHAYDE
Post-Graduating Program in Veterinary Medicine, Federal University of Campina Grande, Patos, Paraíba State, Brazil

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Abstract: This study aimed to investigate the occurrence of *Cryptosporidium* spp. and *Giardia* spp. in cattle calves from the Sertão region of Paraíba State in northeastern Brazil. The experiment was conducted using 100 calves from 10 farms. To evaluate the presence of oocysts, the technique of flotation in a sucrose solution was used, and for detection of *C. parvum* and *G. duodenalis*, the immunochromatographic test RIDA®QUICK *Cryptosporidium*/*Giardia* strips were used. It was noted that animals of 1 to 30 days of age were statistically more affected ($P < 0.05$) by *Cryptosporidium* spp. Only 4 calves (25% of the positive animals) had diarrhea ($P < 0.05$). Nine animals were positive for *G. duodenalis*; diarrhea was present in 6 (66.67%) of those animals, which was considered a risk factor ($P < 0.05$). It was concluded that *Cryptosporidium* spp. and *G. duodenalis* were present in calves from northeastern Brazil.

Key words: *Cryptosporidium* spp., *Giardia* spp., protozoan, semiarid, zoonosis

Cryptosporidium spp. is an opportunistic protozoan that affects several species of birds and mammals, including humans (1). It is a pathogen of great concern for cattle farmers, especially affecting young animals, for which the cryptosporidiosis can cause severe enteritis, resulting in significant morbidity, which can reach 100% and can result in several economic losses to livestock (2). Cattle are the main host of this protozoan, and the eliminated oocysts have been implicated as a major source of contamination for human cryptosporidiosis outbreaks (3).

Cryptosporidiosis in cattle is largely caused by *C. parvum* (4). However, other species can be involved, such as *C. andersoni*, *C. bovis*, and *C. ryanae* (*C. parvum* deer-like genotype) (5).

Giardia duodenalis (syn. *G. lamblia*, *G. intestinalis*) affects humans and animals worldwide (6). This parasite can cause serious damage to livestock production due to diarrhea and consequent weight loss, but some infections are asymptomatic and occur only in environmental infestation (7).

Due to the incipient knowledge about these protozoa in semiarid environments and the impact that they have on cattle farming and public health, this study aimed to verify the occurrence of *Cryptosporidium* spp. and *Giardia* spp. in calves from the Sertão region of Paraíba State in northeastern Brazil.

The study was conducted in the cities of Patos, Quixabá, Malta, São José do Bonfim, and São José dos Espinharas in the Sertão region of Paraíba State in northeastern Brazil. This region has a semiarid climate, with a dry season and a rainy season from January to May when over 90% of the rainfall occurs. The annual mean temperature is 30.6 °C (minimum: 28.7 °C, maximum: 35.5 °C), with little variation during the year.

The animals were sourced from 10 cattle production farms (Figure), chosen regardless of the health status of the herd when calf diarrhea was present. A total of 100 calves were used, 56 males and 44 females, with ages ranging from 6 days to 3 months old. Fecal samples were collected from 10 calves per farm. The samples were individually collected and sent for fecal analyses to the Laboratory of Parasitic Diseases of Domestic Animals at the Federal University of Campina Grande, Patos, Paraíba State, Brazil.

All the fecal samples were tested for the presence of oocysts of *Cryptosporidium* spp. and *Giardia* spp. using the technique of flotation in a sucrose solution, with a specific gravity of 1.205 g/cm^3. The sample was considered positive for this protozoan if at least 1 oocyst was detected upon direct microscopic (400×) examination, according to the criteria of oocyst optical properties, internal structure, size, and shape as described by Fayer and Xiao (8).

To detect *C. parvum* and *G. duodenalis*, the commercial immunochromatographic test RIDA®QUICK *Cryptosporidium*/*Giardia* strips were used, with a sensitivity of 100% and a specificity of 98.1% (9).

* Correspondence: vilelavlr@yahoo.com.br

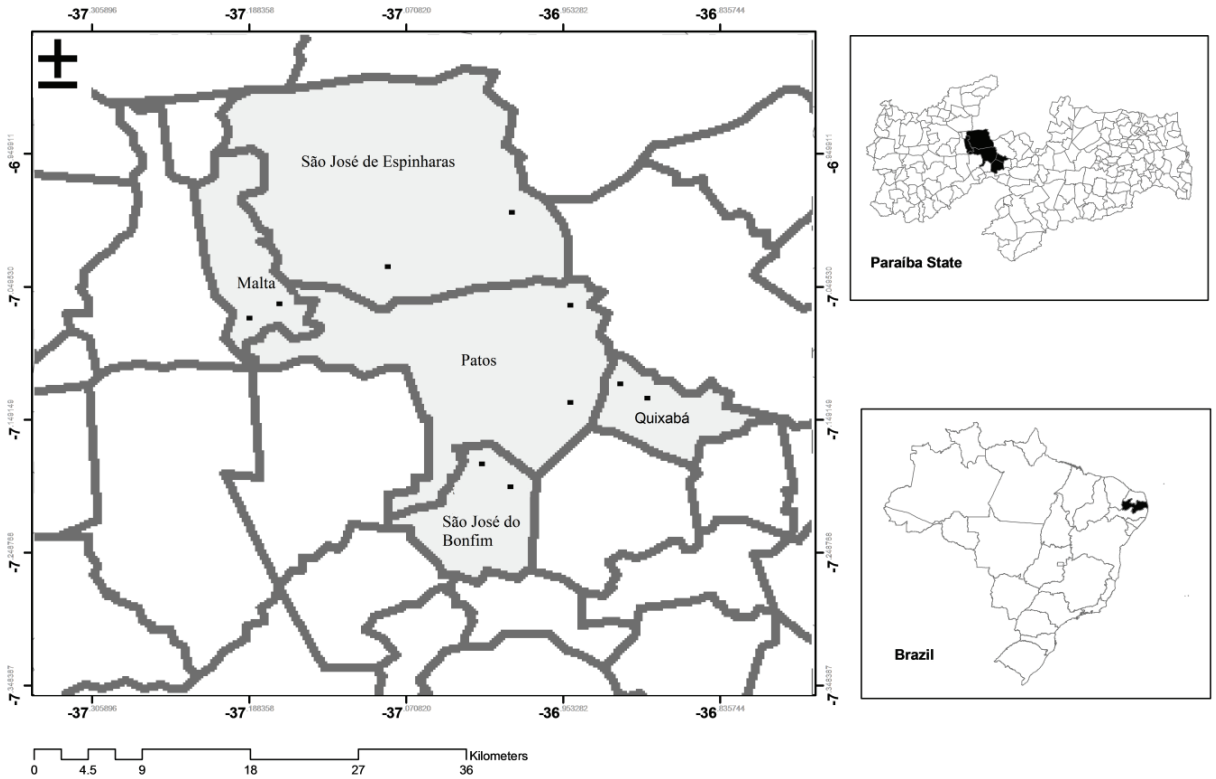

Figure. Geographic location of the calf farms in the Sertão region of Paraíba State in northeastern Brazil.

To evaluate possible associations between the different age groups and sex or presence of diarrhea, chi-square and Fisher's exact tests were used, respectively. Statistical tests were considered significant when $P < 0.05$. Data were analyzed by BioEstat 5.0.

Seven farms were detected (70%) with at least 1 animal positive for *Cryptosporidium* spp., and 3 (30%) farms were positive for *Giardia* spp., determined using flotation in a sucrose solution. This is the first report of *Cryptosporidium* spp. and *Giardia* spp. in calves from northeastern Brazil.

It was noted that 16% of the fecal samples were positive for *Cryptosporidium* spp. through the flotation test. Among the positive animals, 68.75% (11/16) were of 1 to 30 days of age and were statistically more affected ($P < 0.05$) by this parasite than the other age group (Table 1).

Only 4 calves (25% of the positive animals) had major clinical signs of diarrhea ($P > 0.05$), while 12 (75%) were eliminating oocysts asymptomatically.

All animals were negative for *C. parvum* based on the immunochromatography test.

A total of 9 animals tested positive (9%) for *G. duodenalis* (4 were 1 to 30 days old and 5 were 31 to 90 days old); the elimination of oocysts did not differ ($P > 0.05$) between the age groups. All animals with *G. duodenalis* were positive for both tests.

Diarrhea was also present in 6 of the 9 animals positive for *G. duodenalis* (66.67%), and this clinical sign was considered a risk factor ($P < 0.05$) (Table 2).

No animal was positive for both *Cryptosporidium* spp. and *G. duodenalis*.

Table 1. Age range of cattle calves evaluated for the presence of *Cryptosporidium* spp. in the Sertão region of Paraíba State, northeastern Brazil.

Age range (days)	Negative animals	Positive animals	Total	Chi-square	P-value
1–30	30	11	41	4.775	0.0289[a]
31–90	54	5	59		
Total	84	16	100		

[a]: Statistically significant ($P < 0.05$).

Table 2. Fecal state of the cattle calves evaluated for the presence of *G. duodenalis* in the semiarid region of northeastern Brazil.

Fecal state	Negative animals	Positive animals	Total	Chi-square	P-value
Diarrheic	10	6	16		
Nondiarrheic	81	3	84	18.891	<0.0001[a]
Total	91	9	100		

[a]: Statistically significant (P < 0.05).

The presence of *Cryptosporidium* spp. at a rate of 16% in this study is similar to the findings of Tiranti et al. (10) when analyzing *Cryptosporidium* spp. in feces of 620 calves in the city of Cordoba, Argentina, where they found a prevalence of 19.35% of this protozoan. However, Sevá et al. (11) noted a very low rate, 3% of 197 animals, in São Paulo State, Brazil.

It was noted that the younger animals are more likely to eliminate oocysts, which corroborates the findings of Feitosa et al. (12), who reported that the easiest age at which to find fecal oocysts in calves is 7 days old. Tiranti et al. (10) observed that the probability of finding oocysts in the feces of calves of less than 2 weeks old is 4 times greater in comparison to older ages.

Asymptomatic elimination of oocysts is common, according to Feitosa et al. (12), who found 73.7% (15/57) of animals eliminating oocysts with no apparent clinical signs. However, Souza and Lopes (13), in a survey conducted in Rio de Janeiro State, Brazil, found 72.13% (39/54) of diarrheic animals positive for *Cryptosporidium* spp.

The presence of *C. parvum* species in the animals studied was not observed, which corroborates the findings of Almeida et al. (14), who concluded that the higher rate of animals positive for *Cryptosporidium* spp. relative to *C. parvum* indicates the existence of other species of *Cryptosporidium* spp. infecting calves. Cardoso et al. (15) observed a very low prevalence of *C. parvum* (0.6%) in 849 cattle. However, numerous studies have shown *C. parvum* as the predominant species in bovine calves. Almeida et al. (14) found a 45% prevalence, and Santín et al. (3) observed the presence of this parasite in 97% of calves before weaning in the state of Maryland, USA.

The current study shows the prevalence of calves infected by *Giardia* spp. to be 9%. Researchers worldwide have described a prevalence for this parasite in cattle that ranges from 3% to 64% (16). Ng et al. (17) obtained a prevalence of 26.9% through PCR in preweaned calves from Western Australia and New South Wales, although Geurden et al. (16) observed 50% of animals positive for this protozoan in northern Vietnam.

Some authors describe parasitism by *G. duodenalis* as the cause of diarrhea in cattle (16–18). Besides the presence of diarrhea in the majority of infected animals, more research is necessary to prove the real damage caused by this parasite in animals (19). In humans, the occurrence of diarrhea is associated with nutritional factors; malnourished children are more likely to present with this symptomatology (20), suggesting that in animals this disturbance manifests in the same way.

Cryptosporidium spp. and *G. duodenalis* infect calves from the semiarid region of Paraíba State in northeastern Brazil, and due to lack of an accurate diagnosis, the occurrence of this parasite may be underreported.

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