

Occurrence of antibodies to *Toxoplasma gondii* in water buffaloes and meat cattle in Rio Grande do Sul State, southern Brazil

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Abstract

Serum samples from 169 water buffaloes and 121 beef cattle were analyzed for antibodies to *T. gondii* by an indirect fluorescent antibody test (IFAT). Positive results were obtained in 27.2% of water buffaloes and 17.4% of cattle. Statistical analysis indicated significant differences between the prevalence in cattle and buffalo ($p \leq 0.05$). The highest titres found in positive animals were 1:256 (buffaloes) and 1:64 (cattle). In both bovine species, toxoplasmosis frequency in young animals (less than 2 years old) was lower compared to older individuals, although the differences seen in cattle were not statistically significant.

Keywords

Bubalus bubalis, water buffaloes, toxoplasmosis, cattle, felids

Introduction

Toxoplasmosis is a disease caused by *Toxoplasma gondii*, an obligatory intracellular, protozoan parasite which is a member of the phylum Apicomplexa (Rorman *et al.* 2006; Dubey 2008). Cats, including both domestic and wild felids, are the definitive hosts of this parasite (Dubey 2008; Jones and Dubey 2010). *T. gondii* is prevalent in most areas of the world and is of veterinary and medical importance, because it may cause abortion or congenital disease in its intermediate hosts, including humans. In Brazil, previous studies have shown that *T. gondii* infections in animals and man are relatively frequent. The infection in cattle does not usually cause clinical symptoms as these hosts show high natural resistance to the parasite (Dubey and Thulliez 1994). However, little is known about the prevalence of *T. gondii* antibodies in water buffaloes (*Bubalus bubalis*), but the indications are that prevalence is generally lower than in cattle (Dubey *et al.* 1998; Huong *et al.* 1998), albeit one study in Brazil showed the opposite tendency. Water buffaloes and cattle are a main source of animal

proteins and products such as bones, skins and goods made from their hides are of great importance for man (Kakar and Kakarsulemankhel, 2008). Since there is a trend to increase the number of water buffaloes farmed in Brazil, we considered it necessary to investigate the importance of *T. gondii* in bovines.

Materials and Methods

Blood samples

A total of 290 blood samples (169 water buffaloes and 121 cattle) were obtained from a single farm. The samples were collected during a three month period (May – July 2011). The group of water buffaloes consisted of Murrah females of different ages (1–18 years old), whereas the cattle were Charolais of both sexes and of different ages (1–12 years old). None of these animals had a history of recent abortions or showed any disease signs. The farm under study was located in the

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Table I. Comparison of positivity of anti-*T. gondii* antibodies obtained (IFAT) according to the age in water buffaloes and beef cattle in southern, Brazil

Species	Age in years				
	1–2	2,1–4	4,1–6	6,1–8	>8
Cattle	9 (14,8%) ^a	0 (0%) ^a	8 (22,2%) ^a	4 (20%) ^a	0 (0%) ^a
Buffalo	18 (19,8%) ^a	09 (47,4%) ^b	08 (40%) ^{a,b}	04 (44,4%) ^{a,b}	07 (23,3%) ^{a,b}

*Value followed by different letters in the same line differ statistically. Regarding age, the prevalence was 19.8% for 1–2 age and significantly higher for adults: 47.4% ($\chi^2 = 9.66$, $P < 0.05$).

State of Rio Grande do Sul in southern Brazil (31°42'S, 52°24'W). Blood samples (5–10 ml from cephalic veins) were collected by qualified health professionals from the University Federal of Pelotas (the present study was approved by the University Federal of Pelotas Animal Ethics Committee with number 2744). Blood was recovered in tubes without anticoagulant and serum was separated by centrifugation at 1800 x g for 10 min at room temperature. Samples were subsequently stored at –20°C until use.

Serological tests

The sera from experimental animals were analyzed for anti-*T. gondii* antibodies by employing a commercial indirect fluorescent antibody test (Camargo 1974). The sera were screened at dilutions of 1:64 to 1:256 and an IFAT titer of 1:64 was regarded as a cut-off level for *T. gondii* antibodies. Sera from non-infected and experimentally infected bovine calves were included as negative and positive controls in both tests.

Statistical analyses

Seroprevalence of anti-*T. gondii* antibodies was calculated separately for cattle and buffaloes. For the statistical analysis of the possible effects of the attributes of species range on the occurrence of anti-*T. gondii* antibodies the chi-squared test was used, at a significance level of 95% ($p < 0.05$) using 2 x 2 contingency tables with one degree of freedom, and different age groups of buffalo and cattle using 2 x 5 contingency tables with four degree of freedom. Statistical analyses were performed by using EpiInfo, version 6.04.

Results and Discussion

Analysis of 169 water buffaloes and 121 meat cattle serum samples revealed relatively high prevalence of *T. gondii* infection (27.2% and 17.4%, respectively). With regard to the bovine species, significant differences in toxoplasmosis level were readily seen ($\chi^2 = 3.86$, $p < 0.05$). The highest titer in cattle was 1 : 64 and in buffaloes was 1 : 256. Age group had a significant effect on seroprevalence in the water buffaloes ($\chi^2 = 9.66$, $p \leq 0.05$) (Table I). However, no significant effect on seroprevalence existed between age group in cattle (Table I).

Previous results in Brazil and other countries have indicated lower prevalence of *T. gondii* in bovines (no more than 4%, as reported by Huong *et al.* 1998; Gondim *et al.* 1999; Fujii *et al.* 2001 and Silva *et al.* 2010, which is in contrast to the results reported here. The reason for such a discrepancy may be due to the fact that changes in feline populations are occurring in the area under study. First, relocation of endangered felines is underway in the state of Rio Grande do Sul, as a result of the activities endorsed by the Wildlife Rehabilitation Centre Program. These activities may have increased the dissemination of *Toxoplasma* oocysts in the area, as observed in other geographic locations (Silva *et al.* 2007; Dubey 2009; Jones and Dubey 2010). Second, a rural settlement location near to the grazing area has been closed since 2008, which has led to the abandonment of domestic cats. These stray animals now feed on birds or small wild mammals, including rodents in that area, and this is likely to increase their chances of contracting toxoplasmosis (Dubey and Beattie 1988; Albuquerque *et al.* 2011). It is evident that bovines, such as herbivorous mammals, may have been infected only by widespread oocyst contamination occurring in their habitat, especially on the pastures that they graze (Frenkel 1970; Miller *et al.* 1972; Dubey and Beattie 1988; Dubey and Jones 2008).

The finding that older water buffaloes showed a tendency to higher levels of toxoplasmosis is not surprising since this fact has been observed previously both in humans (Speare 1985) and animals (Persad 2011). The fact that the same effect was not observed in cattle may have been due to the smaller sample size for this group. Alternatively, age may not be an important factor influencing susceptibility to toxoplasmosis in that species.

The present results are very interesting, especially our finding of a moderate prevalence of toxoplasmosis in bovines. Products derived from these animals may be infective to man. Preliminary studies conducted by us in the sampled farm indicate that toxoplasmosis in humans has a low prevalence, although the group studied was too small to draw any definitive conclusions on this matter. From 1975 to 2000, the water buffalo population in the Brazilian Amazon increased at nearly 13% per year, making it one of the fastest growing herds in the world (Sheikh *et al.* 2006). This fact underlines the need for a better understanding of zoonosis such as toxoplasmosis that may be transferred from domestic animals to man.

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