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Study of infection by *Babesia bovis* and *Anaplasma marginale* in dairy water buffaloes in the state of São Paulo - Oliveira M.C.S.<sup>1\*</sup>, Néo T.A.<sup>2</sup>, Giglioti R.<sup>3</sup>, Rabelo M.D.<sup>1</sup>, Copriva A.K.<sup>4</sup>, Brito L.G.<sup>5</sup>

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The aim of this study was to evaluate the prevalence of infection by *Babesia bovis* and *Anaplasma marginale* in dairy water buffaloes infested by ticks on farms, either in exclusive buffalo herds or in in mixed herds along with cattle. From each type of farm, blood samples were collected from 50 buffaloes (25 cows and 25 calves), for a total of 100 samples. The samples were drawn from the tail vein and were submitted to DNA extraction with the Easy Kit (Invitrogen). To date, PCR and nested-PCR reactions have been prepared for amplifications of gene parts of *Anaplasma marginale* and *Babesia bovis*. The Chi-square test was applied to compare the frequency of infections of the animals in the two stock-raising systems. No significant differences were observed in the frequency of infection by the species studied between the two systems. The PCR and nested-PCR reactions showed a high infection rate of 98% by *A. marginale* in the animals raised in both exclusive and mixed herds. The *B. bovis* infection rate was 14.6% for animals from mixed herds and 18.3% for those from exclusive buffalo herds. These preliminary results suggest that the presence of cattle along with buffaloes in herds does not influence the infection rate by either *A. marginale* or *B. bovis*.

Key-words: tick fever, infection, PCR

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# Study of infection by *Babesia bovis* and *Anaplasma marginale* in dairy water buffaloes in the state of São Paulo

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#### INTRODUCTION

The Babesiosis protozoa in conjunction with the rickettsia Anaplasma marginale are intraerythrocytic pathogens that are responsible for the most prevalent and costly tick borne diseases (TBD's) of cattle worldwide (Suarez et al., 2011). The aim of this study was to evaluate the prevalence of infection by Babesia bovis and Anaplasma marginale in dairy water buffaloes infested by ticks on farms, either in exclusive buffalo herds or in in mixed herds along with cattle.

#### METHODS

- ✓ Buffalo: from each type of farm, blood samples were collected from 50 buffaloes (25 cows and 25 calves) by the tail vein using tubes containing EDTA anticoagulant, for a total of 100 samples.
- Extraction DNA: The samples were submitted to DNA extraction with the Easy Kit (Invitrogen) and were quantified using NanoDrop ND 1000 spectrophotometer (Thermo Scientific).
- Analysis: PCR and nested-PCR reactions have been prepared for amplifications of gene parts of Anaplasma marginale and Babesia bovis.
- Statistics: Statistical analysis was performed in order to compare the frequency of infection by B. bovis and A. marginale in animals of two age groups using the Fisher test (F) and chi-square. Analyses were performed by SAS statistical package (2002/2003).



Figure 1. Property with buffaloes and cattle



Figure 2. Property with exclusive creation of buffaloes







Figure 4. Anaplasma marginale

## RESULTS

No significant differences were observed in the frequency of infection by the species studied between the two systems. The PCR and nested-PCR reactions showed a high infection rate of 98% by A. marginale in the animals raised in both exclusive and mixed herds. For the categories, the results were similar, with 100% of cows were infected in both locations (Table 1).

Table 1. Values of the frequencies of thenNested-PCR analysis for categories cows and calves and in conditions with and without the presence of bovine cattle.

	Nestet PCR by category			
	Calves		Cows	
di .	With bovine	Without bovine	With bovine	Without bovine
Positive	96% (24)	96% (24)	100%(25)	100%(25)
Negative	4% (1)	4% (1)		

The *B. bovis* infection rate was 14.6% for animals from mixed herds and 18.3% for those from exclusive buffalo herds. The frequencies of infection by *B. bovis* separated by categories and conditions are shown in Figure 5.

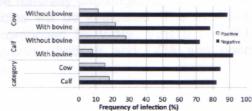


Figure 5. Frequencies of infection by *B. bovis* separated by categories and

POTENCIAL IMPACTS ON SOLVING ANIMAL HEALTH ISSUES These preliminary results suggest that the presence of cattle along with buffaloes in herds does not influence the infection rate by either A. marginale or B. bovis.

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