









# Occurrence of anti-*Brucella abortus* agglutinins in small ruminants in Sergipe

## Ocorrência de aglutininas anti-*Brucella abortus* em pequenos ruminantes de Sergipe

Huber Rizzo<sup>1\*</sup> , Rachel Livingstone Felizola Soares de Andrade<sup>2</sup> , Helen Carolina de Oliveira Menezes<sup>2</sup> , Urias Fagner Santos Nascimento<sup>2</sup> , Ícaro Wille dos Santos Silva<sup>3</sup> , Taile Katiele Sousa de Jesus<sup>2</sup> , Jeferson Silva Carvalho<sup>1</sup> , Lília Márcia Paulin Silva<sup>4</sup> 

**ABSTRACT:** The objective this study was to evaluate the occurrence of small ruminants seropositive for *Brucella abortus* in Sergipe, Brazil. Samples of sheep (1,200) and goats (675) serum blood was collected from 101 farms of 25 counties of the three mesoregions of the state. The technique of Acidified Buffered Acid stained with Rose Bengal (RB) was used and all samples confirmed a positive result using the 2-Mercaptoethanol technique. Four (0.21%) seropositive animals were diagnosed by technique AAT stained with RB, three sheep's (0.25%) and a goat (0.15%), were confirmed by the 2ME test. In all the positive properties, the intercropping of small ruminants with cattle was adopted, favoring the contact with the agent. The prevalence of anti-*B. abortus* antibodies in herds of small ruminants in Sergipe is described for the first time, and is low, however, it is important to identify seropositive animals to be discarded due to their zoonotic potential.

**KEYWORDS:** buffered acidified antigen; brucellosis; ovine; caprine; serology.

**RESUMO:** Objetivou-se avaliar a ocorrência de pequenos ruminantes soropositivos para *Brucella abortus* em Sergipe, Brasil, a partir de 1.200 amostras de soro sanguíneo de ovinos e 675 de caprinos, oriundos de 101 propriedades de 25 municípios das três mesorregiões do estado, nas quais foi empregada a técnica do Antígeno Acidificado Tamponado (AAT) corado com Rosa Bengala (RB) como triagem e do 2-Mercaptoetanol (2ME) como confirmatória. Foram diagnosticados 4 animais (0,21%) soropositivos pela técnica de AAT corado com RB, 3 ovelhas (0,25%) e 1 cabra (0,15%), que posteriormente foram confirmados pelo teste de 2ME. Em todas as propriedades positivas era adotada a criação consorciada de pequenos ruminantes com bovinos, favorecendo o contato com o agente causador. A ocorrência de anticorpos anti-*B. abortus* em rebanhos sergipanos de pequenos ruminantes é descrita pela primeira vez, e mostra-se baixa; entretanto, é importante a identificação de animais soropositivos para que sejam descartados, devido seu potencial zoonótico.

**PALAVRAS-CHAVE:** antígeno acidificado tamponado; brucelose; ovinos; caprinos; sorologia.

<sup>1</sup>Universidade Federal Rural de Pernambuco – Recife (PE), Brazil

<sup>2</sup>Faculdade Pio Décimo, Câmpus III – Aracaju (SE), Brazil

<sup>3</sup>Universidade Federal de Sergipe – São Cristovam (SE), Brazil

<sup>4</sup>Instituto Biológico – São Paulo (SP), Brazil

\*Corresponding author: hubervet@gmail.com

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Brucellosis is an infecto-contagious and zoonotic disease of importance in the ruminants breeding, caused by *Brucella abortus* in cattle, *B. ovis* in sheep, and *B. melitensis* in goats. It may also occur sporadic infection among these species (MANISH et al., 2013), mainly by *B. abortus* in small ruminants in the intercropped breeding with cattle, where transmission occurs through digestive tract from contact with secretions and miscarriages and/or through the supply of milk from seropositive cows to neonates (CARNEIRO et al., 2005; VARGES et al., 2008). Infected animals may present reproductive disorders such as abortion, placental retention, epididymitis, and orchitis, becoming also potential transmitters of the disease to man (MANISH et al., 2013).

Although microbiological isolation is the most effective method for diagnosing the disease, indirect serological methods are the most routinely used, since they offer high sensitivity and low cost when recommended for cattle and buffaloes. However, it should not be adopted alone mainly in programs of control and eradication of the disease (KALTUNGO et al., 2014), being those recommended by the National Program for the Control and Eradication of Brucellosis Tuberculosis (PNCEBT) the test of the buffered acidified antigen (AAT), 2-mercaptoethanol (2-ME) and the complement fixation reaction (RFC) (MEIRELLES-BARTOLI; MATHIAS, 2010).

Given the scarcity of data about this disease in Sergipe, this study aimed to report the occurrence of *B. abortus* agglutinins in goats and sheep bred in the state.

During the years 2011 and 2014, in 25 municipalities of Sergipe, according to the availability of producers, 101 properties of small ruminants distributed in the three mesoregions of the state were visited: East (n = 44), Agreste (n = 30), and Sertão (n = 27). Of these, 1,875 samples (1,556 females and 319 males) were collected by venopunction of the external jugular, being 1,200 of sheep, between the years of 2011 and 2012, and 675 of goats, between 2013 and 2014, of 60 and 41 properties respectively, where randomly selected and without repetition, from seven to twenty animals of different zootechnical patterns, apparently healthy, aged over six months and with no history of vaccination for brucellosis. The biological material was processed in the serology laboratory of the Faculdade Pio Décimo, Aracaju, Sergipe, and the blood serum was subjected to research of anti-*B. abortus* agglutinins by the technique of AAT stained with RB, prepared and commercialized by the Biological Institute of São Paulo. Samples that contained formation of clumps of agglutination were considered positive. Subsequently, the positive sera by the technique of AAT stained with RB were submitted to the 2-ME method for confirmation (MADRUGA et al., 2001). Descriptive statistical analysis by absolute and relative distribution was used to calculate the occurrence. The research project was approved by the Ethics committee of the Faculdade Pio Décimo, Aracaju, Sergipe, under the number 06/2011 of April 16, 2011.

Of the 1,875 serum from small ruminants created in Sergipe, submitted to anti-*B. abortus* agglutinin research by the

technique of AAT stained with RB, 4 (0.21%) females were seropositive, 3 ewes of Santa Inês breed, 0.25% of sheep, and 1 goat without definite breed, 0.15% of goats (Table 1). The serum of these four animals when tested by the 2-ME test confirmed the seropositivity, thus reducing the chances of false positive animals, since it is more specific than the AAT stained with RB, however less sensitive, besides identifying vaccinal antibodies. There is the possibility of performing the RFC, as a confirmatory serological test, especially in cases of positive animals for the AAT stained with RB and inconclusive for the 2ME, due to its high specificity (MEIRELLES-BARTOLI; MATHIAS, 2010; KALTUNGO et al., 2014), a situation that did not occur with sera from the positive sheep and goats of Sergipe.

Even with low occurrence, small infected ruminants were identified, which may serve as disseminators of the disease in herds, not observed in surveys carried out in Minas Gerais (SALABERRY et al., 2011), Paraná (PASQUALI et al., 2017) and in northeastern herds of the semiarid of Paraíba (ALVES et al., 1997), Sertão de Itaparica, Pernambuco (NASCIMENTO et al., 2015), Teresina, Piauí (DA SILVA et al., 2017), Petrolina in Pernambuco, and Juazeiro and Valente (PEIXOTO et al., 2016) and region of Senhor do Bonfim, Bahia (ALBUQUERQUE et al., 2014), with no seropositive animals among those surveyed. Studies that identified small seropositive ruminants found rates between 0.2 and 9% in the states of Rio de Janeiro (FRAGUAS et al., 2004; LILENBAUM et al., 2007; VARGES et al., 2008), Mato Grosso (SANTOS et al., 2016), Pernambuco (PINHEIRO JUNIOR et al., 2008), and Tocantins (MARTINS et al., 2013).

In 3.33% (2/60) and 2.44% (1/41) of the properties of sheep and goats, at least one seropositive animal was diagnosed, where they all adopted the breeding intercropped with cattle, which was observed in 85% (51/60) and 41.5% (17/41) of the breeding of sheep and goats studied. This association between dairy goats and cattle was reported in studies that presented high seropositive rates in relation to the other national studies in Bahia (CARNEIRO et al., 2005) and Rio de Janeiro where the cow that provided milk for the goats was diagnosed infected by *B. abortus* (VARGES et al., 2008) and the herds that had seropositive caprine animals had a history of lactation with bovine milk (LILENBAUM et al., 2007). In sheep, the presence of bovine animals was also reported by SANTOS et al. (2016), in Mato Grosso, which attempted to compromise the PNCEBT in the state due to the presence of infected sheep in cattle herds, in addition to the risk of public health (CARNEIRO et al., 2005). The 3 properties with seropositive animals presented occurrence of 10% (1/20) and 20% (2/20), and were located in different municipalities (Simão Dias, Estancia, and Salgado), representing 12% (3/25) of the surveyed. In relation to the Mesoregions, in the Sertão (n = 504) all animals were seronegative, while in the Agreste and East were diagnosed two animals each, representing 0.39% (2/513) and 0.23% (2/858) of the small ruminants collected in the respective mesoregions. None of the seropositives was created in the extensive system, predominantly observed in

the Sertão mesoregion, which has a drier climate in relation to the others, reducing the availability of *Brucellas* in the environment (MANISH et al., 2013). According to CARNEIRO et al. (2005), there is a relationship between the breeding system and the seropositivity, being the animals under semi-intensive regime more predisposed to contract the disease in relation to the intensive. Sanitary conditions, climate, and breeding system can contribute to the differences observed in the various regions surveyed (CARNEIRO et al., 2005; PINHEIRO JUNIOR et al., 2008).

The presence of antibodies anti-*B. abortus* in Sergipe herds of small ruminants was very low. However, it is important to

identify seropositive animals to be discarded, especially in properties with breeding with cattle due to the predilection of *B. abortus* to this species of ruminants and their zoonotic potential.

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**Table 1.** Occurrence of antibodies anti-*B. abortus* in serum from small ruminants determined by AAT and 2-ME, according to properties with seropositive animals, municipalities, and mesoregion of the state of Sergipe. 2011–2014.

Municipality	Goats (%)		Sheep		Total	
	Prop. (+)/ Prop.	Positive/Total	Prop. (+)/ Prop.	Positive/Total	Prop. (+)/ Prop.	Positive/Total
Aracaju	0/3 (0)	0/50 (0)	0/1 (0)	0/20 (0)	0/4 (0)	0/70 (0)
Araúá	-	-	0/3 (0)	0/60 (0)	0/3 (0)	0/60 (0)
Boquim	-	-	0/1 (0)	0/20 (0)	0/1 (0)	0/20 (0)
Divina Pastora	-	-	0/3 (0)	0/60 (0)	0/3 (0)	0/60 (0)
Estância	0/1 (0)	0/20 (0)	1/4 (25)	1/80 (1,25)	1/5 (20)	1/100 (1)
Itabaianinha	-	-	0/9 (0)	0/180 (0)	0/9 (0)	0/180 (0)
Itaporanga D'Ajuda	0/1 (0)	0/20 (0)	0/5 (0)	0/100 (0)	0/6 (0)	0/120 (0)
Maruim	-	-	0/2 (0)	0/40 (0)	0/2 (0)	0/40 (0)
Nossa Sra. do Socorro	0/1 (0)	0/18 (0)	0/1 (0)	0/20 (0)	0/2 (0)	0/38 (0)
Salgado	1/2 (50)	0/30 (0)	0/3 (0)	0/60 (0)	1/5 (20)	1/90 (1,1)
São Cristóvão	0/1 (0)	0/20 (0)	0/2 (0)	0/40 (0)	0/3 (0)	0/60 (0)
Neópolis	0/1 (0)	0/20 (0)	-	-	0/1 (0)	0/20 (0)
Mesorregião Leste	1/10 (10)	1/178 (0,56)	1/34 (2,94)	1/680 (0,15)	2/44 (4,55)	2/858 (0,23)
Cumbe	-	-	0/1 (0)	0/20 (0)	0/1 (0)	0/20 (0)
Itabaiana	0/1 (0)	0/20 (0)	-	-	0/1 (0)	0/20 (0)
Lagarto	0/1 (0)	0/19 (0)	0/6 (0)	0/120 (0)	0/7 (0)	0/139 (0)
Macambira	0/3(0)	0/42 (0)	-	-	0/3(0)	0/42 (0)
Nossa Sra. das Dores	-	-	0/4 (0)	0/80 (0)	0/4 (0)	0/80 (0)
Poço Verde	0/11 (0)	0/154 (0)	-	-	0/11 (0)	0/154 (0)
Simão Dias	-	-	2/3 (66,7)	2/60 (3,33)	2/3 (66,7)	2/60 (3,33)
Mesorregião Agreste	0/16 (0)	0/235 (0)	2/14 (14,29)	2/280 (0,71)	2/30 (6,67)	2/515 (0,39)
Canindé São Francisco	0/6 (0)	0/119 (0)	0/4 (0)	0/80 (0)	0/10 (0)	0/199 (0)
Feira Nova	-	-	0/2 (0)	0/40 (0)	0/2 (0)	0/40 (0)
Itabi	-	-	0/2 (0)	0/80 (0)	0/2 (0)	0/80 (0)
Nossa Sra. da Glória	0/6 (0)	0/93 (0)	0/4 (0)	0/80 (0)	0/10 (0)	0/173 (0)
Pedra Mole	0/1 (0)	0/11 (0)	-	-	0/1 (0)	0/11 (0)
Pinhão	0/2 (0)	0/39 (0)	-	-	0/2 (0)	0/39 (0)
Mesorregião Sertão	0/15 (0)	80/262 (0)	0/12 (0)	0/280 (0)	0/27 (0)	0/542 (0)
Total	1/41 (2,44)	1/675 (0,15)	3/60 (5)	3/1.200 (0,25)	4/101 (3,96)	4/1.875 (0,21)

Prop. (+): properties with at least one seropositive animal for *B. abortus*.

Prop.: total properties that were performed to collect serum for diagnosis of *B. abortus*

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