Brazilian Journal of Veterinary Parasitology
ISSN 1984-2961 (Electronic)
www.cbpv.org.br/rbpv
Braz. J. Vet. Parasitol., Jaboticabal, v. 28, n. 2, p. 245-257, apr.-june 2019
Doi: https://doi.org/10.1590/S1984-29612019032

Original Article

Coxiella burnetii associated with BVDV (Bovine Viral Diarrhea Virus), BoHV (Bovine Herpesvirus), *Leptospira* spp., *Neospora caninum, Toxoplasma gondii* and *Trypanosoma vivax* in reproductive disorders in cattle

Coxiella burnetii associada ao BVDV (Vírus da Diarreia Viral Bovina), BoHV (Vírus do Herpes Bovino), *Leptospira* spp., *Neospora caninum*, *Toxoplasma gondii* e *Trypanosoma vivax* em distúrbios reprodutivos em bovinos

Diego Carlos Souza Zanatto[1,2]; Igor Renan Honorato Gatto[3]; Marcelo Bahia Labruna[4]; Marcia Mariza Gomes Jusi[5]; Samir Issa Samara[3]; Rosangela Zacarias Machado[2]; Marcos Rogério André[2]*

[1] Programa de Pós-graduação em Microbiologia Agropecuária, Universidade Estadual Paulista Júlio de Mesquita Filho – UNESP, Jaboticabal, SP, Brasil

[2] Departamento de Patologia Veterinária, Universidade Estadual Paulista Júlio de Mesquita Filho – UNESP, Jaboticabal, SP, Brasil

[3] Departamento de Medicina Veterinária Preventiva, Universidade Estadual Paulista Júlio de Mesquita Filho – UNESP, Jaboticabal, SP, Brasil

[4] Departamento de Medicina Veterinária Preventiva e Saúde Animal, Universidade de São Paulo – USP, São Paulo, SP, Brasil

[5] Imunodot Diagnósticos, Jaboticabal, SP, Brasil

Received November 29, 2018
Accepted April 22, 2019

Abstract

This is a cross-sectional study to assess the presence of antibodies in ruminants against selected pathogens associated with reproductive disorders in cattle in four Brazilian states, including the zoonotic agent *Coxiella burnetii*. The used tests were Virus Neutralization Assay for IBR and BVD, Microscopic Agglutination Test for *Leptospira* spp., Indirect Fluorescent Antibody Test (IFAT) for *C. burnetii* and *Toxoplasma gondii*, and Enzyme-Linked Immunosorbent Assay for *Neospora caninum* and *Trypanosoma vivax*. Seropositivity for *C. burnetii* was 13.7% with titers from 128 to 131,072; 57.8% for BoHV-1, with titers between 2 and 1,024; 47.1% for BVDV-1a, with titers from 10 to 5,120; 89.2% for *N. caninum*; 50% for *T. vivax*; and 52.0% for *Leptospira* spp., with titers between 100 to 800 (the following serovars were found: Tarassovi, Grippotyphosa, Canicola, Copenhageni, Wolffi, Hardjo, Pomona and Icterohaemorrhagiae); 19.6% for *T. gondii* with titer of 40. This is the first study that has identified *C. burnetii* in cattle associated with BoHV and BVDV, *N. caninum*, *Leptospira* spp., *T. gondii* and *T. vivax*. Thus, future studies should be conducted to investigate how widespread this pathogen is in Brazilian cattle herds.

Keywords: *Coxiella burnetii*, abortion, Q Fever, cattle.

Resumo

Este é um estudo transversal para avaliar a presença de anticorpos em ruminantes contra patógenos selecionados e associados a distúrbios reprodutivos em bovinos de quatro estados brasileiros, incluindo o agente zoonótico *Coxiella burnetii*. Os testes utilizados foram Teste de Vírus-Neutralização para BoHV e BVDV, teste de Aglutinação Microscópica para *Leptospira* spp., Reação de Imunofluorescência Indireta for *C. burnetii* e *Toxoplasma gondii*, e Ensaio de Imunoabsorção Enzimática para *Neospora caninum* e *Trypanosoma vivax*. A soropositividade para *C. burnetii* foi de 13,7% com títulos de 128 a 131.072; 57,8% para BoHV-1, com títulos entre 2 a 1.024; 47,1% para BVDV-1a, com títulos de 10 a 5.120; 89,2% para *N. caninum*; 50% para *T. vivax*; e 52,0% para *Leptospira* spp., com títulos entre 100 a 800 (sorovares encontrados: Tarassovi, Grippotyphosa, Canicola, Copenhageni, Wolffi, Hardjo, Pomona e Icterohaemorrhagiae) 19,6% para *T. gondii*

***Corresponding author:** Marcos Rogério André. Laboratório de Imunoparasitologia, Departamento de Patologia Veterinária, Faculdade de Ciências Agrárias e Veterinárias Júlio de Mesquita Filho – UNESP, Via de Acesso Prof. Paulo Donato Castellane, s/n, Zona Rural, CEP 14884-900, Jaboticabal, SP, Brasil. e-mail: marcosandre.fcav@gmail.com

com título de 40. Este é o primeiro estudo que evidencia a participação de *C. burnetii* em bovinos associada ao Vírus da Rinotraqueíte bovina infecciosa e da diarreia viral bovina, *N. caninum*, *Leptospira* spp., *T. gondii* e *T. vivax* em bovinos. Desta forma, futuros estudos devem ser conduzidos a fim de investigar o quão disseminado se encontra este patógeno em rebanhos bovinos brasileiros.

Palavras-chave: *Coxiella burnetii*, abortamento, Febre Q, gado.

Introduction

Both Brazilian beef and dairy cattle production are prominent in economic scenario worldwide. However, reproductive disorders are a serious obstacle to excellence in production (LUCY, 2001; ROYAL, et al., 2000), and abortion is considered to be one of the biggest causes of economic losses in cattle farming (REICHEL et al., 2013). Considering that infections involving the reproductive system can reduce fertility, the pathogenicity involved infectious agents has been associated with both intrinsic and extrinsic factors (PELLERIN et al., 1994; STENLUND et al., 2003; GROOMS, 2004), including management practices, such as vaccination and introduction of new animals into the herd (CHI et al., 2002).

In the center-west of the United States, it was found that approximately 30% of abortions were caused by infectious agents; half of them involved pathogenic bacteria as etiologic agents (KIRKBRIDE, 1992). Infectious-parasitic agents associated with reproductive disorders in ruminants include *Coxiella burnetii,* Bovine Herpesvirus (BoHV) and bovine viral diarrhea (BVD) virus, *Neospora caninum* (KIRKBRIDE, 1992), *Leptospira* spp., (MURRAY, 1990), *Toxoplasma gondii* (BÁRTOVÁ et al., 2009) and *Trypanosoma vivax* (SILVA et al., 1996, 1998).

Coxiella burnetii, a Gram-negative obligatory intracellular bacterium (VAN SCHAIK et al, 2013; ABDEL-MOEIN & HAMZA, 2017), was reported for the first time in 1930 and has been detected worldwide ever since, except in New Zealand (MAURIN & RAOULT, 1999; ELDIN et al., 2017). This parasite is the zoonotic agent that causes Q fever (MAURIN & RAOULT, 1999). It was also considered as a potential bioterrorism agent by the Center for Disease Control and Prevention (CDC, 2018). Although the main route of infection for humans is via aerosols (MAURIN & RAOULT, 1999; PARKER et al., 2006; TISSOT-DUPONT & RAOULT, 2008), ingestion of contaminated food and infected ticks may also represent alternative routes of transmission of the parasite (ELDIN et al., 2017).

In humans, this agent has been associated with pneumonia, a syndrome similar to influenza and hepatitis; in some cases, pruritus, pericarditis, myocarditis, encephalitis, and osteomyelitis have also been observed (MAURIN & RAOULT, 1999; TISSOT-DUPONT & RAOULT, 2008). The chronic form of the infection can cause endocarditis, which is usually associated with valvular heart disease and immunosuppression. Although less commonly, individuals can develop granulomatous lesions in the bones, joints, liver, lungs, testicles and other tissues (RALPH et al. 2007; TISSOT-DUPONT & RAOULT, 2008).

The first serological evidence of exposure to *C. burnetii* infection in Brazil occurred in the 1950s in humans in the state of São Paulo (BRANDÃO et al., 1953) and in cattle in the state of Rio de Janeiro (TRAVASSOS et al., 1954 apud OLIVEIRA et al., 2018). Ever since, serological and molecular evidence of circulation of the parasite has been found in humans in São Paulo (BRANDÃO et al., 1953; VALLE et al., 1955; SICILIANO et al., 2015), Rio de Janeiro (LAMAS et al., 2009, 2013; LEMOS et al., 2011, 2018; ROZENTAL et al., 2012, 2018; MARES-GUIA et al., 2016), Minas Gerais (RIEMANN et al., 1974; COSTA et al., 2005; 2006) and Bahia (SICILIANO et al., 2008).

The main clinical signs associated with this infection in ruminants are infertility, abortion, stillbirth, endometritis and mastitis (TISSOT-DUPONT & RAOULT, 2008). Although *C. burnetii* is a zoonotic agent associated with the occurrence of abortion in ruminants all over the world, there are few studies about the occurrence of this agent in ruminants in Brazil (TRAVASSOS et al., 1954 apud OLIVEIRA et al., 2018; MARES-GUIA et al., 2014; GUIMARÃES et al., 2017; OLIVEIRA et al., 2018; SOUZA et al., 2018). Therefore, the objective of this study was to investigate the frequency of antibodies against ruminant-selected pathogens, namely *C. burnetii*, BoHV-1, BVDV, *N. caninum*, *Leptospira* spp., *T. gondii* and *T. vivax* in serum samples from cattle with a history of reproductive problems from four Brazilian states.

Materials and Methods

Sample selection

This is a cross-sectional study to assess the presence of antibodies to ruminant-selected pathogens associated to reproductive disorders in cattle from four Brazilian states, including the zoonotic *C. burnetii*. The total of 12 proprieties (Supplementary Material) were conveniently selected from the Reproductive Viruses Center (São Paulo State University, Unesp, Jaboticabal - São Paulo, Brazil). The total of 102 serum samples were obtained from cows showing a history of reproductive disorders. All sample sets were located in the central-western and southeastern regions of Brazil.

Eligibility criteria

Study animals consisted of adult cows (> 24 months) from productions (milk and breeding) (i.e., commercial cattle production). All samples were obtained from the sampled farms between December, 2014 and November, 2015.

Bovine serum samples

Between 2014 and 2015, serum samples collected from 102 cattle presenting reproductive disorders were selected by convenience from four Brazilian states, São Paulo (José Bonifácio, Urânia and Torrinha), Minas Gerais (Espera Feliz, Patos de

Minas, Lagoa Grande and Coromandel), Mato Grosso do Sul (Anastácio) and Goiás (Nova Caixás and Goiatuba). While the proprieties A, B, G and L were composed by beef cattle, the proprieties C, D, E, F, H, I, J and K were composed by dairy cattle (Suplementary Table). Serum samples were stored at -20°C until all serological assays were performed.

Virus Neutralization (VN) assay for detection of BVDV-1 and BoHV-1

The selected bovine serum samples were tested by virus neutralization (VN) assay for the detection of antibodies to BVDV-1 and BoHV-1, as recommended by the "*Manual of Diagnostics Tests and Vaccines for Terrestrial Animals*" (OIE, 2018a,b), with modifications. For this purpose, "Madin-Darby bovine kidney (MDBK)" cell line and cytopathic strains of BVDV-1a (Singer) and BoHV-1 (Nebraska), were used. All seropositive samples were tested twice in order to estimate the geometric mean of titer (GMT) values. In addition to the internal controls, positive and negative controls were also used (GATTO et al., 2018). A sample was considered positive when the total neutralization of 100 $TCID_{50}$ occurred in the serum and no cytopathic effect (CPE) was observed in the cell layer in serum dilutions higher than 1:10 (BVDV) and 1:2 (BoHV-1) (OIE, 2018a,b).

BVDV-1

The serum samples were heat-inactivated for 30 minutes at 56°C. All serum samples were tested in duplicate at the same time and were subjected to successive dilutions ranging from 1:10 to 1:5120, using cell culture medium as diluent. At each dilution of serum, for each sample one well was left without virus, aiming at monitoring for evidence of sample toxicity that could mimic viral cytopathology or interfere with virus replication. An equal volume (50 µL) of a stock of cytopathic strain of BVDV, which contained 100 $TCID_{50}$ (50%) tissue culture infective doses, was added to each well. The plate was incubated for 1 hour at 37°C. Fifty microliters of the cell suspension at 2×10^5 /mL were added to each well. The plate was incubated at 37°C for 4–5 days, either in a 5% CO_2 atmosphere.

BoHV-1

The serum samples were heat-inactivated for 30 minutes at 56°C. All serum samples were tested in duplicate and were subjected to successive dilutions ranging from 1:2 to 1:1024, using cell culture medium as diluent. At each dilution of serum, for each sample, one well was also left without virus in order to monitor the evidence of sample toxicity that could mimic viral cytopathology or interfere with virus replication. An equal volume (50 µL) of a stock of cytopathic strain of BoHV-1, which contained 200 $TCID_{50}$ (50%) tissue culture infective doses, was added to each well. The plate was incubated for 24 hours at 37°C. One hundred microliters of the cell suspension at 3×10^5 cells per well were added to each well. The plate is incubated at 37°C for 3–5 days, either in a 5% CO_2 atmosphere.

Microscopic Agglutination Test (MAT) for detection of IgG antibodies to Leptospira spp.

The *Leptospira* spp. antigens used in serological tests were obtained from bacteria subcultured weekly in liquid EMJH culture medium (Ellighausen, McCullough, Johnson and Harris), with 10% of the medium volume used to seed cultures that were maintained in a bacteriological incubator at 28°C ± 1°C (OIE, 2018c).

The MAT was used to identify the serogroups/serovars. The 24 *Leptospira* serovars used can be found in Table 1. Serum samples were diluted in saline, at an initial dilution of 1:50. Aliquots (25 µL) of serum were placed in polystyrene plates with a flat bottom, with an equal quantity of antigens, resulting in a dilution of 1:100. The serum-antigen mixture was homogenized gently and incubated in an environmental incubator at a temperature of 28 °C for 40 to 120 minutes.

Results were read by dark field microscopy with 10× objective, directly from the plate wells. Samples shoeing 50% agglutination were considered to be reactive. Samples reactive at the initial dilution were assayed with serial twofold dilutions from the original 1:100 dilution. Positive tests were defined as MAT results ≥1:100 for at least one of the 24 serovars (Table 1).

Indirect Fluorescent Antibody Test (IFAT) for detection of IgG anti-C. burnetii and anti-T. gondii antibodies

The detection of anti-*C. burnetii* IgG antibodies was performed with crude antigen of strain At12 of *C. burnetii*, phase-1 reactive (PACHECO et al., 2013). The serum samples

Table 1. *Leptospira* antigens used in MAT.

No	Serogroup	Serovar
1	Australis	Australis
2	Australis	Bratislava
3	Autumnalis	Autumnalis
4	Autumnalis	Butembo
5	Ballum	Castellonis
6	Bataviae	Bataviae
7	Canicola	Canicola
8	Celledoni	Whitcombi
9	Cynopteri	Cynopteri
10	Grippotyphosa	Grippotyphosa
11	Hebdomadis	Hebdomadis
12	Icterohaemorrhagiae	Copenhageni
13	Icterohaemorrhagiae	Icterohaemorrhagiae
14	Javanica	Javanica
15	Panama	Panama
16	Pomona	Pomona
17	Pyrogenes	Pyrogenes
18	Sejroe	Hardjo
19	Sejroe	Wolffi
20	Shermani	Shermani
21	Tarassovi	Tarassovi
22	Andamana	Andamana
23	Seramanga	Patoc
24	Djasiman	Sentot

were first diluted at 1:64 in phosphate-buffered saline solution, PBS pH 7.4 (130 mM NaCl; 2.7 mM KCl; 5.6 mM Na_2HPO_4; 1.0 mM KH_2PO_4; 0.8 mM NaH_2PO_4). After that, 20 µL of each diluted serum sample was deposited in wells of slides containing the antigen of *C. burnetii*. The slides were incubated at 37°C for 30 minutes, in a moist chamber. Afterwards, they were washed with Washing Buffer solution (phosphate-buffered saline solution, PBS pH 7.4 + 1% Triton) and then dried. Each slide received 20 µL conjugate (bovine anti-IgG diluted at 1:200) marked by fluorescein isothiocyanate (Sigma-Aldrich®, St. Louis - Missouri, USA) for test samples as well as positive and negative controls. The slides were then incubated for a further 30 minutes at 37°C in a moist chamber. After being washed (this time was added 1.5 ml of Evans Blue on each wash in the washing buffer solution) and dried again, the slides were evaluated through ultraviolet light microscopy. The reaction was considered to be positive when cells were fluorescent at the dilution of 1:64 (PACHECO et al., 2013), according to the protocol previously described by Reeves et al. (2006). Bovine serum samples previously tested for *C. burnetii* and considered non-reactive and reactive were used as negative and positive controls, respectively (GUIMARÃES et al., 2017).

For *T. gondii*, the tachyzoites of the RH strain were used as an antigen, according to a protocol previously described (OLIVEIRA et al., 2008; ANDRÉ et al., 2010). Samples were considered positive when titration was above 40. The used protocol was similar to that one used for detection of IgG antibodies for *C. burnetii*, with some modifications, such as following: i.) PBS pH 7.4 was used in all washing procedures; ii.) Evans Blue at 10% to the conjugate solution was used instead of the washing solution. IFAT used a serial dilution of the test sera to the log base 2 for titration of antibodies. The positive and negative controls used in this serological assay were serum samples from cattle known to be positive and negative for *T. gondii*, provided by IMUNODOT Diagnósticos (Jaboticabal - São Paulo, Brazil).

Indirect Immunoassay (iELISA) for detection of IgG antibodies anti-Neospora caninum and anti-Trypanosoma vivax

The detection of IgG antibodies anti-*T. vivax* was performed with the indirect ELISA (iELISA), according to the protocol established by Machado et al. (1997) and Aquino et al. (1999), with minor adaptations. After purification of the trypomastigote forms, as described by González et al. (2005), and sonication in a 750w Ultrasonic Processor (Coleparmer – Montréal – Quebec, Canada), the protein concentration of the soluble antigen was determined by the bicinchoninic acid method (Pierce BCA Protein Assay Kit, Thermo Fisher Scientific, San Diego – California, USA). The total antigen of *T. vivax* was diluted in carbonate-bicarbonate buffer 0.5M and pH 9.6 and its optimal concentration was adjusted to 0.1 µg/mL. After incubation for 12 hours at 8 °C, blocking was performed with PBS Tween 20 (pH 7.2), and 6% normal rabbit serum was added. The plates (Nunc Maxisorp®, Thermo Fisher Scientific, São Paulo - São Paulo, Brazil) sensitized with *T. vivax* antigen were incubated for 90 minutes at 37°C, in a moist chamber. After three washes with PBS-Tween 20 buffer, reference positive and negative sera (FIDELIS et al., 2016) and test sera were added to the ELISA plate. All serum samples were diluted 1:400 solution in PBS-Tween 20 plus 5% of skimmed milk powder (Molico®, Nestlé, São Paulo - São Paulo, Brazil). The plates were incubated again at 37°C for 90 minutes. After three washes with PBS-Tween 20 buffer, the ELISA plate received the anti-bovine IgG antibody conjugate linked to alkaline phosphatase (Sigma®, St. Louis - Missouri, USA) at a dilution of 1:30000 in PBS-Tween 20 plus 5% of skimmed milk powder (Molico®, Nestlé, São Paulo - São Paulo Brazil), with subsequent incubation and washing. Finally, the substrate of the enzyme alkaline phosphatase, p-nitrophenyl phosphate (Sigma®, St. Louis - Missouri, USA), diluted to 1 mg/mL at pH 9.8 buffer diethanolamine (Sigma®, St. Louis - Missouri USA) was added.

The total antigen of *N. caninum* (IMUNODOT®, Jaboticabal - São Paulo, Brazil), at optimal concentration of 0.1 µg/mL, was diluted in carbonate-bicarbonate buffer 0.5M and pH 9.6. After incubation for 12 hours at 8 °C, blocking was performed with PBS Tween 20 (pH 7.2), and 6% of skimmed milk powder (Molico®, Nestlé, São Paulo - São Paulo, Brazil). The plates (Nunc Maxisorp®, Thermo Fisher Scientific, São Paulo - São Paulo, Brazil) were sensitized with the antigen of this parasite and then were incubated for 60 minutes at 37°C in a moist chamber. After three washes with PBS-Tween 20 buffer, the ELISA plate received the reference positive and negative sera, supplied with the kit, and tested sera, which were diluted at 1:200 in PBS-Tween 20 solution. The plates were incubated again at 37°C for 60 minutes. After three washes with PBS-Tween 20, the ELISA plate received the anti-bovine IgG antibody conjugate linked to alkaline phosphatase (Sigma®, St. Louis - Missouri, USA) at a dilution of 1:30000 in PBS-Tween 20, with subsequent incubation and washing. Finally, the substrate of the enzyme alkaline phosphatase, p-nitrophenyl phosphate (Sigma®, St. Louis - Missouri, USA), diluted at 1 mg/mL at pH 9.8 diethanolamine buffer (Sigma®, St. Louis - Missouri, USA) was added.

The plates were sealed with aluminum foil and incubated for 45 minutes at ambient temperature. Reading was performed in ELISA plate reader (B.S.-100; Embrabio, São Paulo - São Paulo, Brazil), with a 405nm filter. The cut-off point was calculated as 2.5 times the average absorbance of negative control sera (MACHADO et al., 1997).

Results

The seropositivity for *C. burnetii* was 13.7% (14/102), with titers ranging from 128 to 131072; 57.8% (59/102) for BoHV-1, with titers ranging from 2 to 1024; 47.1% (48/102) for BVDV-1a, with titers ranging from 10 to 5120; 19.6% (20/102) for *T. gondii*, with titers of 40; 89.2% (91/102) for *N. caninum,* with optical densities ranging from 0.309 to 1.646 (cutoff point: 0.309); 50% (51/102) for *T. vivax,* with optical densities ranging from 0.406 to 2.597 (cutoff point: 0.393); and 52.0% (53/102) for *Leptospira* spp., with titers ranging from 100 to 800 (serovars found: Tarassovi, Grippotyphosa, Canicola, Copenhageni, Wolffi, Hardjo, Pomona and Icterohaemorrhagiae).

Antibodies for *C. burnetii* were found in two animals in the state of Goiás (10%, 2/20), four animals in the state of São Paulo (12.5% 4/32), seven animals in the state of Minas Gerais (17.9%, 7/40) and only one animal in Mato Grosso do Sul (10%, 1/10). All seropositive animals for *C. burnetii* also showed antibodies to at least one more agents. There were significant differences for the presence of antibodies against *Leptospira* spp., *T. gondii* and BVDV-1 between the animals sampled in the four selected states (Table 2 and Table 3).

Among the selected animals presenting reproductive disorders in the present study, a higher seropositivity for *N. caninum* was found. In this sense, antibodies to *N. caninum* were found in 100% (10/10) of the animals sampled in the state of Mato Grosso do Sul, 85% (17/20) of the animals sampled in the state of Goiás, 93.7% (30/32) of the animals sampled in the state of São Paulo, and 85% (34/40) of the animals sampled in the state of Minas Gerais. Six animals showed to be seropositive only for *N. caninum* (Table 2 and Figure 1).

Table 2. Seropositive samples for selected bovine pathogens associated to abortion in cattle in Brazil.

PATHOGENS	% OF SOROPOSITIVITY IN EACH STATE				Total (102)
	Goiás (20)	São Paulo (32)	Minas Gerais (40)	Mato Grosso do Sul (10)	
Coxiella burnetii	10% (2)	12.5% (4)	17.5% (7)	10% (1)	13.7% (14)
Leptospira spp.	65% (13) ab	37.5% (12) b	47.5% (19) b	90% (9) a	52.0% (53)
BVDV-1a (Singer)	50% (10) ab	25% (8) b	62.5% (25) a	50% (5) ab	47.1% (48)
BoHV-1 (Nebraska)	60% (12)	68.7% (22)	50% (20)	50% (5)	57.8% (59)
Neospora caninum	85% (17)	93.7% (30)	85% (34)	100% (10)	89.2% (91)
Toxoplasma gondii	10% (2) b	40.6% (13) a	12.5% (5) b	0% b	19.6% (20)
Trypanosoma vivax	55% (11)	34.4% (11)	55% (22)	70% (7)	50.0% (51)

Different lowercase letters on the same row represent a significant difference (P <0.05), a>b.

Table 3. Seropositivity for parasites associated with abortion in cattle in Brazil according to place of origin.

	Goiás	São Paulo	Mato Grosso do Sul	Minas Gerais	Total
	20	32	10	40	
N.c	1 (0.98%)	1 (0.98%)	-	-	2 (1.96%)
BoHV-1	-	-	-	1 (0.98%)	1 (0.98%)
C.b. + T.g.	-	-	-	1 (0.98%)	1 (0.98%)
N.c. + T.g	-	1 (0.98%)	-	-	1 (0.98%)
N.c. + T.v.	-	-	-	3 (2.94%)	3 (2.94%)
N.c. + BVDV-1	1 (0.98%)	-	-	1 (0.98%)	2 (1.96%)
N.c. + BoHV-1	-	2 (1.96%)	-	1 (0.98%)	3 (2.94%)
***N.c. + L.* sp.**	2 (1.96%)	2 (1.96%)	2 (1.96%)	1 (0.98%)	7 (6.86%)
T.v. + BVDV-1	-	-	-	1 (0.98%)	1 (0.98%)
BVDV-1 + BoHV-1	-	-	-	1 (0.98%)	1 (0.98%)
C.b. + N.c. + T.v.	1 (0.98%)	-	-	-	1 (0.98%)
C.b. + T.g. + BoHV-1	-	1 (0.98%)	-	-	1 (0.98%)
C.b. + N.c. + BVDV-1	-	-	1 (0.98%)	-	1 (0.98%)
***C.b. + N.c. + L.* sp.**	-	-	-	1 (0.98%)	1 (0.98%)
N.c. + T.v. + BVDV-1	-	-	-	2 (1.96%)	2 (1.96%)
N.c. + T.g. + T.v.	-	1 (0.98%)	-	-	1 (0.98%)
N.c. + T.g. + BoHV-1	-	2 (1.96%)	-	-	2 (1.96%)
***N.c. + T.v. + L.* sp.**	1 (0.98%)	-	1 (0.98%)	1 (0.98%)	3 (2.94%)
N.c. + T.v. + BoHV-1	1 (0.98%)	5 (4.90%)	-	1 (0.98%)	7 (6.86%)
N.c. + BVDV-1 + BoHV-1	-	-	-	2 (1.96%)	2 (1.96%)
***N.c. + T.g. + L.* sp.**	1 (0.98%)	-	-	1 (0.98%)	2 (1.96%)
***N.c. + BVDV-1 + L.* sp.**	-	1 (0.98%)	-	1 (0.98%)	2 (1.96%)
T.v. + BVDV-1 + BoHV-1	-	-	-	1 (0.98%)	1 (0.98%)
***BVDB-1 + BoHV-1 + L.* sp.**	1 (0.98%)	-	-	-	1 (0.98%)
***C.b. + T.v. + BVDV-1 + L.* sp.**	-	-	-	1 (0.98%)	1 (0.98%)
***C.b. + N.c. + BoHV-1 + L.* sp.**	-	1 (0.98%)	-	-	1 (0.98%)
***C.b. + N.c. + BVDV-1 + L.* sp.**	-	1 (0.98%)	-	1 (0.98%)	2 (1.96%)
C.b. + N.c. + T.g. + T.v.	-	1 (0.98%)	-	-	1 (0.98%)

C.b.: *Coxiella burnetii; N.c.: Neospora caninum; T.g.: Toxoplasma gondii; T.v.: Trypanosoma vivax; L.* sp.: *Leptospira* sp.

Table 3. Continued...

	Goiás	São Paulo	Mato Grosso do Sul	Minas Gerais	Total
	20	32	10	40	
C.b.* + *N.c.* + *BVDV-1* + *BoHV-1	-	-	-	1 (0.98%)	1 (0.98%)
C.b.* + *N.c.* + *T.v.* + *BoHV-1	-	-	-	1 (0.98%)	1 (0.98%)
***N.c.* + *T.g.* + *BoHV-1* + *L.* sp.**	-	2 (1.96%)	-	-	2 (1.96%)
***N.c.* + *T.g.* + *BVDV-1* + *L.* sp.**	-	-	-	1 (0.98%)	1 (0.98%)
***N.c.* + *T.g.* + *T.v.* + *L.* sp.**	-	-	-	2 (1.96%)	2 (1.96%)
***N.c.* + *T.v.* + *BoHV-1* + *L.* sp.**	-	-	1 (0.98%)	2 (1.96%)	3 (2.94%)
N.c.* + *T.g.* + *T.v.* + *BoHV-1	-	1 (0.98%)	-	-	1 (0.98%)
N.c.* + *T.v.* + *BVDV-1* + *BoHV-1	2 (1.96%)	1 (0.98%)	-	4 (3.92%)	7 (6.86%)
***N.c.* + *T.v.* + *BoHV-1* + *L.* sp.**	2 (1.96%)	1 (0.98%)	2 (1.96%)	1 (0.98%)	6 (5.88%)
***N.c* + *BVDV-1* + *BoHV-1* + *L.* sp.**	3 (2.94%)	2 (1.96%)	-	3 (2.94%)	8 (7.84%)
N.c.* + *T.g.* + *BVDV-1* + *BoHV-1	-	3 (2.94%)	-	-	3 (2.94%)
***T.v.* + *BVDV-1* + *BoHV-1* + *L.* sp.**	1 (0.98%)	-	-	-	1 (0.98%)
***N.c.* + *T.g.* + *T.v.* + *BoHV-1* + *L.* sp.**	-	1 (0.98%)	-	-	1 (0.98%)
***N.c.* + *T.v.* + *BVDV-1* + *BoHV-1* + *L.* sp.**	2 (1.96%)	-	3 (2.94%)	2 (1.96%)	7 (6.86%)
***C.b.* + *N.c.* + *BVDV-1* + *BoHV-1* + *L.* sp.**	-	-	-	1 (0.98%)	1 (0.98%)
***C.b.* + *N.c.* + *T.g.* + *T.v.* + *L.* sp.**	1 (0.98%)	-	-	-	1 (0.98%)

C.b.: *Coxiella burnetii; N.c.: Neospora caninum; T.g.: Toxoplasma gondii; T.v.: Trypanosoma vivax; L.* sp.: *Leptospira* sp.

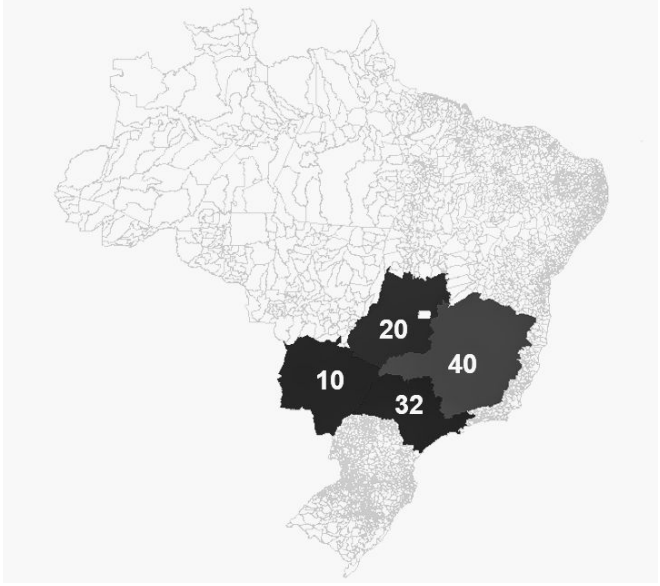

Figure 1. Selected sampling States: Goiás, Minas Gerais, Mato Grosso do Sul, São Paulo. This map was generated using MapInfo software.

Discussion

Although several pathogens interfere with the reproductive performance of cattle, e.g., the bovine viral diarrhea virus (BVDV), bovine herpesvirus (BoHV), *Brucella abortus* (occasionally *B. melitensis*), *N. caninum, C. burnetii, Campylobacter fetus venerealis* or *Campylobacter fetus fetus*, *Leptospira* spp., *Tritrichomonas foetus* and *Chlamydia abortus* (AGERHOLM, 2013; BUTZLER, 2004; DAMMAN et al., 2015; DÍAZ APARICIO, 2013; CERQUEIRA-CÉZAR et al., 2017), there is still not enough information on the occurrence and the epidemiological impact of some pathogens on reproductive disorders in ruminants.

Despite the low rate of seropositivity found for *C. burnetii* (13.7%, 14/102) among the sampled bovine population with reproductive disorders, it should be noted that most (71.4%, 10/14) of the seropositive animals showed high antibody titers, ranging from 512 to 131072. Even though high antibody titers are suggestive of recent infection (GUIMARÃES et al., 2017; OLIVEIRA et al., 2018), the antigen used in the present study is only reactive to phase-1 antibodies, found only in the chronic phase of the disease (PEACOCK et al., 1983). Therefore, it is possible that the seropositive animals were in the chronic phase of the disease at the time of sampling. Additionally, despite the high titers found in the present study, we can't rule out the occurrence of serological cross-reaction with other agents, such as *Legionella* sp. and *Bartonella* sp. (MUSSO & RAOULT, 1997; SCOLA & RAOULT, 1996). Although a previous study reported the occurrence of antibodies to *C. burnetii* in cattle in Brazil (BRANDÃO et al., 1953), the present work showed high antibody titers against this agent in this country.

Although serological evidence of exposure to *C. burnetii* has already been reported in cattle in Europe (CZAPLICKI et al., 2009; AGGER et al., 2010; BOTTCHER et al., 2011; GACHE et al., 2017; RYAN et al., 2018; VARELA-CASTRO et al., 2018; VIDAL et al., 2017; SOFTIC et al., 2018), Asia (CETINKAYA et al., 2000; NOKHODIAN et al., 2016), North America (MCQUISTON et al., 2005) and Africa (KAMGA-WALADJO et al., 2010; SCOLAMACCHIA et al., 2010), there are few data on seroprevalence to this agent in ruminants in Brazil. In this regard, while Guimarães et al. (2017) found a seropositivity rate of 2% for *C. burnetii* among sheep sampled in the state of Piauí, northeast of Brazil, Oliveira et al. (2018) found a seroprevalence of 55.1% in a herd of goats with history of reproductive disorders in the state of Alagoas, Northeast of Brazil. Recently, Souza et al. (2018) found 2.2% of the goats and 2.1% of the sheep seropositive to *C. burnetii* in the state of Pernambuco, also in northeastern Brazil.

In southeastern Brazil, Mares-Guia et al. (2014) found 66.6% seropositivity in sheep and 50% seropositivity in goats in the state of Rio de Janeiro.

According to Mori & Roest (2018), vaginal swabs samples from cows that aborted within 8 days should preferably be collected aiming at the direct detection of *C. burnetii* by PCR. However, if the abortion occurred a longer time ago or if there is any history of reproductive problem in the herd, serologic tests can be used to verify if there was any contact with *C. burnetii* in the herd. This approach justifies the use of IFAT on cattle serum samples for investigating the occurrence of *C. burnetii* in the cattle showing reproductive disorders in the present study. Unfortunately, additional information regarding the abortion in each sampled property was not available.

In the present study, the seropositive samples were originated from both beef and dairy cattle herds. The ELISA test in milk storage tanks has proved very effective in sorting lactating animals seropositive for *C. burnetii* (VAN DEN BROM et al., 2012; GUATTEO et al., 2007) as well as have produced results concordant with those found from individual serum samples because of the transfer of immunoglobulins from blood to milk (NIELSEN et al., 2011). Thus, future large-scale studies should be conducted in Brazil using this approach aiming at investigating the exposure of dairy cows to the agent under study. The detection of DNA of *C. burnetii*, but not of viable bacteria in dairy products (cheese, yoghurt, butter and creams) in France (ELDIN et al., 2013) emphasizes the need for additional studies in Brazil to investigate the prevalence of this agent in dairy cattle.

Although the main clinical signs associated with Q fever in ruminants are linked to reproduction (infertility, abortion, stillbirth, endometritis and mastitis) (TISSOT-DUPONT & RAOULT, 2008; ELDIN et al., 2017), seronegative cows that do not have medical abortion are also capable of spreading the parasite through vaginal discharges (RODOLAKIS et al., 2007). Considering that *C. burnetii* can be dispersed via aerosols at distances greater than 30 Km (ELDIN et al., 2016) and survives for long periods in the environment as a form of resistance (*Small Cell Variant*) (ELDIN et al., 2017), this agent is very likely to be widely distributed in regions close to the areas sampled in this study. Because *C. burnetii* is an agent that can infect a wide range of hosts, from unicellular beings (such as amoeba) to invertebrates, reptiles, birds and mammals (CUTLER et al., 2007; NASPHV, 2013; ELDIN et al., 2017), the dispersion of this pathogen in extensive areas seems to be easily achieved. In this sense, the data presented in this study were relevant, both from an economic point of view and for public health, considering that cattle are one of the main reservoirs for human infection (GEORGIEV et al., 2013; NOKHODIAN et al., 2016). An outbreak of Q fever has been recently described among cadets in the state of Rio de Janeiro, Southeastern Brazil, whose diagnosis was confirmed by serologic tests (LEMOS et al., 2018). Additionally, future studies should be conducted about the participation of ticks in the transmission of *C. burnetii* between animals and humans in Brazil.

In the present study, we have also reported the occurrence of antibodies against BoHV-1, BVDV and *Leptospira* spp. In Brazil, previous seroepidemiological studies have indicated a significant dissemination of BVDV, BoHV-1 (AFFONSO et al., 2010; FINO et al., 2012; FLORES et al., 2013), and *Leptospira* sp. in herds (FÁVERO et al., 2017). While prevalence rates ranging from 22.2% to 85.4% (FLORES et al., 2013; QUINCOZES et al., 2007; ALMEIDA et al., 2013) have been reported for BVDV, seropositivity rates between 18% and 90% were found for BoHV-1 in non-vaccinated herds in different geographic regions of Brazil (LOVATO et al., 1995; TAKIUCHI et al., 2001; DIAS et al., 2013). The *Leptospira* spp. serovars Hardjo, Wolffi and Grippotyphosa, are the most frequently associated with the occurrence of abortions in cattle (MINEIRO et al., 2007), which corroborates the results of the present study. The serological results of this study for BVDV, BoHV-1 and *Leptospira* sp. support the evidence that these three etiological agents are disseminated in Brazil and may directly favor the occurrence of reproductive disorders in cattle, thus participating as primary infectious agents or worsening clinical co-infection conditions.

The highest rate of seropositivity (89.2%, 91/102) in the present study was found for the apicomplexan *N. caninum*. Our data corroborate those found previously in other Brazilian states, in which *N. caninum* has been reported as the main causative agent of abortion in cattle. In this sense, seropositivity rates of 23-91.7% to this agent were described in cattle presenting abortions in the states of Rio Grande do Sul, Minas Gerais, Paraná, Rondônia, Mato Grosso and Amazonas (CERQUEIRA-CÉZAR et al., 2017). In Brazil, seroprevalence to *N. caninum* in cattle ranges from 6.7 to 91.7%, depending on the geographic region and cattle breed. Cattle can be infected by ingesting oocysts of the parasite eliminated through feces of Canidae (horizontal transmission) or via the placenta (vertical transmission) (CERQUEIRA-CÉZAR et al., 2017). Transplacental transmission was incriminated the main *N. caninum*'s route of transmission in cattle in the state of Pernambuco, northeastern Brazil, allowing the maintenance of this agent in production systems in the study region (RAMOS et al., 2016). In this sense, farms that use embryo recipients should take preventive measures to avoid economic losses and perpetuation of the parasite on farms.

While *N. caninum* stands out as one of the most important parasites associated with reproductive problems in cattle, such animals are considered to be resistant to *T. gondii* (CERQUEIRA-CÉZAR et al., 2017), in spite of its worldwide distribution and its association with abortions in small ruminants (WYROSDICK & SCHAEFER, 2015). Toxoplasmosis in animal production is serious because it poses potential risks to public health, as a result of the consumption of non-pasteurized milk and raw or underdone beef, associated with economic losses caused by abortion in small ruminants (TENTER et al., 2000; WYROSDICK & SCHAEFER, 2015). Herein, a seroprevalence of 19.2% (20/102) was found to *T. gondii*, which is lower than that others reported in previous studies (29.1-32.9%) in Brazil (TILAHUN et al., 2018). This fact corroborates results of previous studies that indicate low pathogenicity of *T. gondii* in cattle, with little or no influence on the occurrence of abortion in herds (CERQUEIRA-CÉZAR et al., 2017).

Trypanosoma vivax infects a large number of species of domestic and wild ungulates and, in South America, it is the main etiologic agent of trypanosomiasis in cattle, hence it causes great economic losses (JONES & DÁVILA, 2001). In Brazil, the transmission

occurs mechanically by Tabanidae (horseflies) and *Stomoxys calcitrans* (stable fly) (PAIVA et al., 2000; RODRIGUEZ-BATISTA et al., 2005; CADIOLI et al., 2012), or iatrogenically by fomites (CADIOLI et al., 2012). The present study reported a seroprevalence of 50% (51/102) for this agent among the cattle population with reproductive disorders. This protozoan is currently associated with the occurrence of abortions in different Brazilian regions (DÁVILA & SILVA, 2000; LINHARES et al., 2006; CADIOLI et al., 2012). Previously considered as an endemic agent in cattle herds in the Pantanal region in the Brazilian states of Mato Grosso (SILVA et al., 1996) and Mato Grosso do Sul (PAIVA et al., 1997, 2000), *T. vivax* has been recently causing outbreaks of the disease in several Brazilian states, e.g., Maranhão (FEITOSA et al., 2004), Tocantins (LINHARES et al., 2006), Paraíba (BATISTA et al., 2007), Minas Gerais (CARVALHO et al., 2008), Rio Grande do Sul (SILVA et al., 2009), Pernambuco (PIMENTEL et al., 2012), São Paulo (CADIOLI et al., 2012), Alagoas (ANDRADE et al., 2015), Santa Catarina (FÁVERO et al., 2016), Goiás (BASTOS et al., 2017), Sergipe (VIEIRA et al., 2017) and Piauí (LOPES et al., 2018). Our study did not find significant differences regarding the presence of antibodies against *T. vivax* across the cows sampled in the four states, which suggests that the parasite is being scattered across the country. Such expansion of its geographical distribution in Brazil is mainly due to the movement of cattle from endemic regions to those where there is a favorable epidemiological situation (climatic conditions favoring the presence of hematophagous dipterans, animals without prior immunity to the parasite in question and long-standing practice of sharing needles contaminated with blood [fomites] during vaccination and application of oxytocin) (CADIOLI et al., 2012; BASTOS et al., 2017).

Although the found results presented information about serological evidence of exposure of cattle to the selected pathogens, it is not possible to determine which one was the real causative agent of the abortion, or if there was an association of agents that could possibly have potentialized the final outcome of a reproductive disorder. The present study was the first to show *C. burnetii* as a pathogen associated with bovine rhinotracheitis and bovine viral diarrhea viruses, *N. caninum*, *Leptospira* spp., *T. gondii* and *T. vivax* in cattle with abortion in Brazil. Future studies should be conducted to investigate how widespread *C. burnetii* is in Brazilian cattle herds and its real role in the etiology of reproductive problems in cattle in South America, as the single agent causing abortion or in co-infections with other agents.

Acknowledgements

This research was supported by Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq). Zanatto has received scholarship under the master's program in Agricultural Microbiology from de Faculdade de Ciências Agrárias e Veterinárias (FCAV), Universidade Estadual Paulista Júlio de Mesquita Filho (UNESP). We appreciate Dr. Samara for kindly supplied the cattle serum samples, Dr. Labruna for the *Coxiella burnetii* antigen and IMUNODOT Diagnósticos Ltda. (Jaboticabal, SP) for *N. caninum, T. gondii, T. vivax* antigens and serological kits. MRA is a fellowship researcher of "Conselho Nacional de Desenvolvimento Científico e Tecnólogico" (CNPq Process number #302420/2017-7).

References

Abdel-Moein KA, Hamza DA. The burden of *Coxiella burnetii* among aborted dairy animals in Egypt and its public health implications. *Acta Trop* 2017; 166: 92-95. http://dx.doi.org/10.1016/j.actatropica.2016.11.011. PMid:27845064.

Affonso IB, Amoril JG, Alexandrino B, Buzinaro MDG, Medeiros ASR, Samara SI. Anticorpos contra o Herpesvírus Bovino Tipo 1 (BoHV-1) nas dez regiões de planejamento do estado de Goiás, Brasil. *Cienc Anim Bras* 2010; 11(4): 892-898. http://dx.doi.org/10.5216/cab.v11i4.6318.

Agerholm JS. *Coxiella burnetii* associated reproductive disorders in domestic animals-a critical review. *Acta Vet Scand* 2013; 55(1): 13. http://dx.doi.org/10.1186/1751-0147-55-13. PMid:23419216.

Agger JF, Christoffersen AB, Rattenborg E, Nielsen J, Agerholm JS. Prevalence of *Coxiella burnetii* antibodies in Danish dairy herds. *Acta Vet Scand* 2010; 52(1): 5. http://dx.doi.org/10.1186/1751-0147-52-5. PMid:20092653.

Almeida LL, Miranda ICS, Hein HE, Santiago WS No, Costa EF, Marks FS, et al. Herd-level risk factors for bovine viral diarrhea virus infection in dairy herds from southern Brazil. *Res Vet Sci* 2013; 95(3): 901-907. https://doi.org/10.1016/j.rvsc.2013.08.009. PMid:24079841.

Andrade AQA No, Afonso JAB, Mendonça CL, Souto RJC, André MR, Machado RZ. Surtos de tripanossomíase em bovinos leiteiros no agreste dos estados de Pernambuco e Alagoas. *Biologico* 2015; 77(S2): 143.

André MR, Adania CH, Teixeira RH, Silva KF, Jusi MM, Machado ST, et al. Antibodies to *Toxoplasma gondii* and *Neospora caninum* in captive neotropical and exotic wild canids and felids. *J Parasitol* 2010; 96(5): 1007-1009. http://dx.doi.org/10.1645/GE-2502.1. PMid:20950109.

Aquino LP, Machado RZ, Alessi AC, Marques LC, Castro MB, Malheiros EB. Clinical, parasitological and immunological aspects of experimental infection with *Trypanosoma evansi* in dogs. *Mem Inst Oswaldo Cruz* 1999; 94(2): 255-260. http://dx.doi.org/10.1590/S0074-02761999000200025. PMid:10224539.

Bártová E, Sedlák K, Literák I. *Toxoplasma gondii* and *Neospora caninum* antibodies in sheep in the Czech Republic. *Vet Parasitol* 2009; 161(1-2): 131-132. http://dx.doi.org/10.1016/j.vetpar.2008.12.022. PMid:19181450.

Bastos TSA, Faria AM, Madrid DMC, Bessa LC, Linhares GFC, Fidelis OL Jr, et al. First outbreak and subsequent cases of *Trypanosoma vivax* in the state of Goiás, Brazil. *Rev Bras Parasitol Vet* 2017; 26(3): 366-371. http://dx.doi.org/10.1590/s1984-29612017019. PMid:28678894.

Batista JS, Riet-Correa F, Teixeira MMG, Madruga CR, Simões SDV, Maia TF. Trypanosomiasis by *Trypanosoma vivax* in cattle in the Brazilian semiarid: Description of an outbreak and lesions in the nervous system. *Vet Parasitol* 2007; 143(2): 174-181. http://dx.doi.org/10.1016/j.vetpar.2006.08.017. PMid:16965857.

Böttcher J, Vossen A, Janowetz B, Alex M, Gangl A, Randt A, et al. Insights into the dynamics of endemic *Coxiella burnetii* infection in cattle by application of phase-specific ELISAs in an infected dairy herd. *Vet Microbiol* 2011; 151(3-4): 291-300. http://dx.doi.org/10.1016/j.vetmic.2011.03.007. PMid:21482042.

Brandão H, Vale LAR, Christovão DA. Investigações sôobre a febre "Q" em São Paulo. I - Estudo sorológico em operários de um frigorífico. *Arq Fac Hig Saude Publica Univ Sao Paulo* 1953; 7(1): 127-131. http://dx.doi.org/10.11606/issn.2358-792X.v7i1p127-131.

Butzler JP. *Campylobacter*, from obscurity to celebrity. *Clin Microbiol Infect* 2004; 10(10): 868-876. http://dx.doi.org/10.1111/j.1469-0691.2004.00983.x. PMid:15373879.

Cadioli FA, Barnabé PA, Machado RZ, Teixeira MCA, André MR, Sampaio PH, et al. First report of *Trypanosoma vivax* outbreak in dairy cattle in São Paulo state, Brazil. *Rev Bras Parasitol Vet* 2012; 21(2): 118-124. http://dx.doi.org/10.1590/S1984-29612012000200009. PMid:22832751.

Carvalho AU, Abrão DC, Facury EJ Fo, Paes PRO, Ribeiro MFB. Occurrence of *Trypanosoma vivax* in Minas Gerais state, Brazil. *Arq Bras Med Vet Zootec* 2008; 60(3): 769-771. http://dx.doi.org/10.1590/S0102-09352008000300037.

Centers for Disease Control and Prevention – CDC. *Bioterrorism agents/diseases* [online]. 2018 [cited 2018 Dec 22]. Available from https://emergency.cdc.gov/agent/agentlist.asp

Cerqueira-Cézar CK, Calero-Bernal R, Dubey JP, Gennari SM. All about neosporosis in Brazil. *Rev Bras Parasitol Vet* 2017; 26(3): 253-279. http://dx.doi.org/10.1590/s1984-29612017045. PMid:28876360.

Cetinkaya B, Kalender H, Ertas HB, Muz A, Arslan N, Ongor H, et al. Seroprevalence of coxiellosis in cattle, sheep and people in the east of Turkey. *Vet Rec* 2000; 146(5): 131-136. http://dx.doi.org/10.1136/vr.146.5.131. PMid:10706331.

Chi J, VanLeeuwen JA, Weersink A, Keefe GP. Management factors related to seroprevalences to bovine viral-diarrhoea virus, bovine-leukosis virus, *Mycobacterium avium* subspecies *paratuberculosis*, and *Neospora caninum* in dairy herds in the Canadian Maritimes. *Prev Vet Med* 2002; 55(1): 57-68. http://dx.doi.org/10.1016/S0167-5877(02)00067-3. PMid:12324207.

Costa PSG, Brigatte ME, Greco DB. Antibodies to *Rickettsia rickettsii, Rickettsia typhi, Coxiella burnetii, Bartonella henselae, Bartonella quintana*, and *Ehrlichia chaffeensis* among healthy population in Minas Gerais, Brazil. *Mem Inst Oswaldo Cruz* 2005; 100(8): 853-859. http://dx.doi.org/10.1590/S0074-02762005000800006. PMid:16444416.

Costa PSG, Brigatte ME, Greco DB. Questing one Brazilian query: reporting 16 cases of Q fever from Minas Gerais, Brazil. *Rev Inst Med Trop São Paulo* 2006; 48(1): 5-9. http://dx.doi.org/10.1590/S0036-46652006000100002. PMid:16547572.

Cutler SJ, Bouzid M, Cutler RR. Q fever. *J Infect* 2007; 54(4): 313-318. http://dx.doi.org/10.1016/j.jinf.2006.10.048. PMid:17147957.

Czaplicki G, Houtain JY, Mullender C, Manteca C, Saegerman C. Bulk tank milk, reliable tool for diagnosing Q Fever in dairy herds? *Epidémiol Santé Anim* 2009; 56: 117-127.

Damman A, Viet AF, Arnoux S, Guerrier-Chatellet MC, Petit E, Ezanno P. Modelling the spread of bovine viral diarrhea virus (BVDV) in a beef cattle herd and its impact on herd productivity. *Vet Res* 2015; 46(1): 12. http://dx.doi.org/10.1186/s13567-015-0145-8. PMid:25828555.

Dávila AM, Silva RAM. Animal trypanosomiasis in South America: current status, partnership, and information technology. *Ann N Y Acad Sci* 2000; 916(1): 199-212. http://dx.doi.org/10.1111/j.1749-6632.2000.tb05291.x. PMid:11193622.

Dias JA, Alfieri AA, Ferreira-Neto JS, Gonçalves VS, Muller EE. Seroprevalence and Risk factors of bovine herpesvirus 1 infection in cattle herds in the state of Parana. *Transbound Emerg Dis* 2013; 60(1): 39-47. http://dx.doi.org/10.1111/j.1865-1682.2012.01316.x. PMid:22364224.

Díaz Aparicio E. Epidemiology of brucellosis in domestic animals caused by *Brucella melitensis, Brucella suis* and *Brucella abortus. Rev Sci Tech* 2013; 32(1): 43-60. http://dx.doi.org/10.20506/rst.32.1.2187. PMid:23837364.

Eldin C, Mélenotte C, Mediannikov O, Ghigo E, Million M, Edouard S, et al. From Q fever to *Coxiella burnetii* infection: a paradigm change. *Clin Microbiol Rev* 2017; 30(1): 115-190. http://dx.doi.org/10.1128/CMR.00045-16. PMid:27856520.

Eldin C, Mélenotte C, Million M, Cammilleri S, Sotto A, Elsendoorn A, et al. 18F-FDG PET/CT as a central tool in the shift from chronic Q fever to *Coxiella burnetii* persistent focalized infection: A consecutive case series. *Medicine* 2016; 95(34): e4287. http://dx.doi.org/10.1097/MD.0000000000004287. PMid:27559944.

Eldin C, Renvoisé A, Raoult D, Angelakis E. *Coxiella burnetii* DNA, but not viable bacteria, in dairy products in France. *Am J Trop Med Hyg* 2013; 88(4): 765-769. http://dx.doi.org/10.4269/ajtmh.12-0212. PMid:23382158.

Fávero JF, Araújo HL, Lilenbaum W, Machado G, Tonin AA, Baldissera MD, et al. Bovine leptospirosis: Prevalence, associated risk factors for infection and their cause-effect relation. *Microb Pathog* 2017; 107: 149-154. http://dx.doi.org/10.1016/j.micpath.2017.03.032. PMid:28351712.

Fávero JF, Silva AS, Biazus AH, Volpato A. *Trypanosoma vivax* infection in goat in west of Santa Catarina state, Brazil. *Comp Clin Pathol* 2016; 25(2): 497-499. http://dx.doi.org/10.1007/s00580-015-2216-7.

Feitosa AB Jr, Guerra RMSNC, Santos HP, Abreu-Silva AL. Registro e morfometria de *Trypanosoma vivax* em esfregaço sanguíneo de bovino no município de Itapecuru-Mirim, Maranhão. *Rev Bras Parasitol Vet* 2004; 13(Supl. 1): 232.

Fidelis OL Jr, Sampaio PH, Machado RZ, André MR, Marques LC, Cadioli FA. Evaluation of clinical signs, parasitemia, hematologic and biochemical changes in cattle experimentally infected with *Trypanosoma vivax. Rev Bras Parasitol Vet* 2016; 25(1): 69-81. http://dx.doi.org/10.1590/S1984-29612016013. PMid:27007249.

Fino TCM, Melo CB, Ramos AF, Leite RC. Diarréia bovina a vírus (BVD) - uma breve revisão. *Rev Bras Med Vet* 2012; 34(2): 131-140.

Flores EF, Weiblen R, Cargnelutti JF, Bauermann V, Spilki FR, Mori E, et al. Emerging animal viruses: real threats or simple bystanders? *Pesq Vet Bras* 2013; 33(10): 1161-1173. http://dx.doi.org/10.1590/S0100-736X2013001000001.

Gache K, Rousset E, Perrin JB, De Cremoux R, Hosteing S, Jourdain E, et al. Estimation of the frequency of Q fever in sheep, goat and cattle herds in France: results of a 3-year study of the seroprevalence of Q fever and excretion level of *Coxiella burnetii* in abortive episodes. *Epidemiol Infect* 2017; 145(15): 3131-3142. http://dx.doi.org/10.1017/S0950268817002308. PMid:29039279.

Gatto IRH, Linhares DCL, Souza Almeida HM, Mathias LA, Medeiros ASR, Poljak Z, et al. Description of risk factors associated with the detection of BVDV antibodies in Brazilian pig herds. *Trop Anim Health Prod* 2018; 50(4): 773-778. http://dx.doi.org/10.1007/s11250-017-1493-3. PMid:29264821.

Georgiev M, Afonso A, Neubauer H, Needham H, Thiery R, Rodolakis A, et al. Q fever in humans and farm animals in four European countries, 1982 to 2010. *Euro Surveill* 2013; 18(8): 20407. PMid:23449232.

González A, Gómez BL, Diez S, Hernández O, Restrepo A, Hamilton AJ, et al. Purification and partial characterization of a *Paracoccidioides brasiliensis* protein with capacity to bind to extracellular matrix proteins. *Infect Immun* 2005; 73(4): 2486-2495. http://dx.doi.org/10.1128/IAI.73.4.2486-2495.2005. PMid:15784595.

Grooms DL. Reproductive consequences of infection with bovine viral diarrhea virus. *Vet Clin North Am Food Anim Pract* 2004; 20(1): 5-19. http://dx.doi.org/10.1016/j.cvfa.2003.11.006. PMid:15062471.

Guatteo R, Beaudeau F, Joly A, Seegers H. *Coxiella burnetii* shedding by dairy cows. *Vet Res* 2007; 38(6): 849-860. http://dx.doi.org/10.1051/vetres:2007038. PMid:17903418.

Guimarães MF, Araujo AC, Freire DP, Machado DM, Martins NN, Moraes-Filho J, et al. Infection survey of *Rickettsia rickettsii* and *Coxiella burnetii* in sheep and goats from National Park of Serra das Confusões, Piauí. *Pesq Vet Bras* 2017; 37(6): 555-560.

Jones TW, Dávila AM. *Trypanosoma vivax*–out of Africa. *Trends Parasitol* 2001; 17(2): 99-101. http://dx.doi.org/10.1016/S1471-4922(00)01777-3. PMid:11228017.

Kamga-Waladjo AR, Gbati OB, Kone P, Lapo RA, Chatagnon G, Bakou SN, et al. Seroprevalence of *Neospora caninum* antibodies and its consequences for reproductive parameters in dairy cows from Dakar–Senegal, West Africa. *Trop Anim Health Prod* 2010; 42(5): 953-959. http://dx.doi.org/10.1007/s11250-009-9513-6. PMid:19997972.

Kirkbride CA. Etiologic agents detected in a 10-year study of bovine abortions and stillbirths. *J Vet Diagn Invest* 1992; 4(2): 175-180. http://dx.doi.org/10.1177/104063879200400210. PMid:1616982.

Lamas CC, Ramos RG, Lopes GQ, Santos MS, Golebiovski WF, Weksler C, et al. *Bartonella* and *Coxiella* infective endocarditis in Brazil: molecular evidence from excised valves from a cardiac surgery referral center in Rio de Janeiro, Brazil, 1998 to 2009. *Int J Infect Dis* 2013; 17(1): e65-e66. http://dx.doi.org/10.1016/j.ijid.2012.10.009. PMid:23219032.

Lamas CC, Rozental T, Bóia MN, Favacho ARM, Kirsten AH, Silva APM, et al. Seroprevalence of *Coxiella burnetii* antibodies in human immunodeficiency virus-positive patients in Jacarepaguá, Rio de Janeiro, Brazil. *Clin Microbiol Infect* 2009; 15(Supl. 2): 140-141. http://dx.doi.org/10.1111/j.1469-0691.2008.02144.x. PMid:19298403.

Lemos ERS, Rozental T, Mares-Guia MAM, Almeida DN, Moreira N, Silva RG, et al. Q fever as a cause of fever of unknown origin and thrombocytosis: first molecular evidence of *Coxiella burnetii* in Brazil. *Vector Borne Zoonotic Dis* 2011; 11(1): 85-87. http://dx.doi.org/10.1089/vbz.2009.0261. PMid:20569012.

Lemos ERS, Rozental T, Siqueira BN, Pessoa AA Jr, Joaquim TE, Silva RG, et al. Q Fever in Military Firefighters during Cadet Training in Brazil. *Am J Trop Med Hyg* 2018; 99(2): 303-305. http://dx.doi.org/10.4269/ajtmh.17-0979. PMid:29943714.

Linhares GFC, Dias FC Fo, Fernandes PR, Duarte SC. Tripanossomíase em bovinos no município de Formoso do Araguaia, Tocantins. *Cienc Anim Bras* 2006; 7(4): 455-460.

Lopes STP, Silva Prado B, Martins GHC, Esmeraldo H, Beserra A, Sousa MAC Fo, et al. *Trypanosoma vivax* em bovino leiteiro. *Acta Sci Vet* 2018; 46(Supl. 1): 287.

Lovato LT, Weiblen R, Tobias FL, Moraes MP. Herpesvírus bovino tipo 1 (HVB 1): inquérito soro-epidemiológico no rebanho leiteiro do Estado do Rio Grande do Sul, Brasil. *Cienc Rural* 1995; 25(3): 425-430. http://dx.doi.org/10.1590/S0103-84781995000300017.

Lucy MC. Reproductive loss in high-producing dairy cattle: where will it end? *J Dairy Sci* 2001; 84(6): 1277-1293. http://dx.doi.org/10.3168/jds.S0022-0302(01)70158-0. PMid:11417685.

Machado RZ, Montassier HJ, Pinto AA, Lemos EG, Machado MRF, Valadão IFF, et al. An enzyme-linked immunosorbent assay (ELISA) for the detection of antibodies against *Babesia bovis* in cattle. *Vet Parasitol* 1997; 71(1): 17-26. http://dx.doi.org/10.1016/S0304-4017(97)00003-4. PMid:9231985.

Mares-Guia MA, Rozental T, Guterres A, Ferreira MS, Botticini RG, Terra AK, et al. Molecular identification of Q fever in patients with a suspected diagnosis of dengue in Brazil in 2013–2014. *Am J Trop Med Hyg* 2016; 94(5): 1090-1094. http://dx.doi.org/10.4269/ajtmh.15-0575. PMid:26928831.

Mares-Guia MAM, Rozental T, Guterres A, Gomes R, Almeida DN, Moreira NS, et al. Molecular identification of the agent of Q fever – *Coxiella burnetii* – in domestic animals in State of Rio de Janeiro, Brazil. *Rev Soc Bras Med Trop* 2014; 47(2): 231-234. http://dx.doi.org/10.1590/0037-8682-0076-2013. PMid:24861300.

Maurin M, Raoult DF. Q fever. *Clin Microbiol Rev* 1999; 12(4): 518-553. http://dx.doi.org/10.1128/CMR.12.4.518. PMid:10515901.

McQuiston JH, Nargund VN, Miller JD, Priestley R, Shaw EI, Thompson HA. Prevalence of antibodies to *Coxiella burnetii* among veterinary school dairy herds in the United States, 2003. *Vector Borne Zoonotic Dis* 2005; 5(1): 90-91. http://dx.doi.org/10.1089/vbz.2005.5.90. PMid:15815154.

Mineiro ALBB, Bezerra EEA, Vasconcellos SA, Costa FAL, Macedo NA. Infecção por leptospira em bovinos e sua associação com transtornos reprodutivos e condições climáticas. *Arq Bras Med Vet Zootec* 2007; 59(5): 1103-1109. http://dx.doi.org/10.1590/S0102-09352007000500003.

Mori M, Roest HJ. Farming, Q fever and public health: agricultural practices and beyond. *Arch Public Health* 2018; 76(1): 2. http://dx.doi.org/10.1186/s13690-017-0248-y. PMid:29321921.

Murray RD. A field investigation of causes of abortion in dairy cattle. *Vet Rec* 1990; 127(22): 543-547. PMid:2281595.

Musso D, Raoult D. Serological cross-reactions between *Coxiella burnetii* and *Legionella micdadei. Clin Diagn Lab Immunol* 1997; 4(2): 208-212. PMid:9067657.

National Association of State Public Health Veterinarians – NASPHV. Prevention and control of *Coxiella burnetii* infection among humans and animals: guidance for a coordinated public health and animal health response [online]. Des Moines: National Assembly of State Animal Health Officials. 2013 [cited 2019 Apr 24]. Available from: http://www.nasphv.org/Documents/Q_Fever_2013.pdf

Nielsen KT, Nielsen SS, Agger JF, Christoffersen AB, Agerholm JS. Association between antibodies to *Coxiella burnetii* in bulk tank milk and perinatal mortality of Danish dairy calves. *Acta Vet Scand* 2011; 53(1): 64. http://dx.doi.org/10.1186/1751-0147-53-64. PMid:22136406.

Nokhodian Z, Feizi A, Moradi A, Yaran M, Hoseini SG, Ataei B, et al. Detection and risk factors of *Coxiella burnetii* infection in dairy cattle based on bulk tank milk samples in center of Iran. *Prev Vet Med* 2016; 134: 139-144. http://dx.doi.org/10.1016/j.prevetmed.2016.10.003. PMid:27836035.

Office International des Epizooties – OIE. *Manual of diagnostics tests and vaccines for terrestrial animals* [online]. Paris: OIE; 2018a [cited 2019 Apr 23]. Chapter 3.4.7, Bovine Viral Diarrhoea; p. 1075-1096. Available from: http://www.oie.int/fileadmin/Home/eng/Health_standards/tahm/3.04.07_BVD.pdf

Office International des Epizooties – OIE. *Manual of diagnostics tests and vaccines for terrestrial animals* [online]. Paris: OIE; 2018b [cited 2019 Apr 23]. Chapter 3.4.11, Infectious bovine rhinotracheitis/infectious pustular vulvovaginitis; p. 1139-1157. Available from: http://www.oie.int/fileadmin/Home/eng/Health_standards/tahm/3.04.11_IBR_IPV.pdf

Office International des Epizooties – OIE. *Manual of diagnostics tests and vaccines for terrestrial animals* [online]. Paris: OIE; 2018c [cited 2019 Apr 23]. Chapter 3.4.12, Leptospirosis; p. 503-516. Available from: http://www.oie.int/fileadmin/Home/eng/Health_standards/tahm/3.01.12_LEPTO.pdf

Oliveira JMB, Rozental T, Lemos ERS, Forneas D, Ortega-Mora LM, Porto WJN, et al. *Coxiella burnetii* in dairy goats with a history of reproductive disorders in Brazil. *Acta Trop* 2018; 183: 19-22. http://dx.doi.org/10.1016/j.actatropica.2018.04.010. PMid:29621535.

Oliveira TMFS, Furuta PI, Carvalho D, Machado RZ. A study of cross-reactivity in serum samples from dogs positive for *Leishmania* sp., *Babesia canis* and *Ehrlichia canis* in enzyme-linked immunosorbent assay and indirect fluorescent antibody test. *Rev Bras Parasitol Vet* 2008; 17(1): 7-11. http://dx.doi.org/10.1590/S1984-29612008000100002. PMid:18554433.

Pacheco RC, Echaide IE, Alves RN, Beletti ME, Nava S, Labruna MB. *Coxiella burnetii* in ticks, Argentina. *Emerg Infect Dis* 2013; 19(2): 344-346. http://dx.doi.org/10.3201/eid1902.120362. PMid:23343647.

Paiva F, Lemos RAA, Nakazato L, Mori AE, Brum KB, Bernardo KC. *Trypanosoma vivax* em bovinos no Pantanal do Estado do Mato Grosso do Sul, Brasil: I.-Acompanhamento clínico, laboratorial e anatomopatológico de rebanhos infectados. *Rev Bras Parasitol Vet* 2000; 9(2): 135-141.

Paiva F, Lemos RAA, Oshiro AE, Salvador SC, Nakazato L. Ocorrência de *Trypanosoma vivax* em bovinos do Estado de Mato Grosso do Sul. *Rev Bras Parasitol Vet* 1997; 6(Supl. 1): 349.

Parker NR, Barralet JH, Bell AM. Q fever. *Lancet* 2006; 367(9511): 679-688. http://dx.doi.org/10.1016/S0140-6736(06)68266-4. PMid:16503466.

Peacock MG, Philip RN, Williams JC, Faulkner RS. Serological evaluation of O fever in humans: enhanced phase I titers of immunoglobulins G and A are diagnostic for Q fever endocarditis. *Infect Immun* 1983; 41(3): 1089-1098. PMid:6885155.

Pellerin C, Van Den Hurk J, Lecomte J, Tijssen P. Identification of a new group of bovine viral diarrhea virus strains associated with severe outbreaks and high mortalities. *Virology* 1994; 203(2): 260-268. http://dx.doi.org/10.1006/viro.1994.1483. PMid:8053150.

Pimentel DS, Ramos CA, Ramos RA, Araújo FR, Borba ML, Faustino MA, et al. First report and molecular characterization of *Trypanosoma vivax* in cattle from state of Pernambuco, Brazil. *Vet Parasitol* 2012; 185(2-4): 286-289. http://dx.doi.org/10.1016/j.vetpar.2011.10.019. PMid:22054681.

Quincozes CG, Fisher G, Hübner SO, Vargas GDA, Vidor T, Brod CS. Prevalence and factors associated with bovine viral diarrhea virus infection in south of Rio Grande do Sul. *Semina: Ciênc Agrár* 2007; 28(2): 269-276. http://dx.doi.org/10.5433/1679-0359.2007v28n2p269.

Ralph A, Markey P, Schultz R. Q fever cases in the Northern Territory of Australia from 1991 to 2006. *Commun Dis Intell Q Rep* 2007; 31(2): 222-227. PMid:17724999.

Ramos IAS, Silva RJ, Maciel TA, Silva JABA, Fidelis OL Jr, Soares PC, et al. Assessment of transplacental transmission of *Neospora caninum* in dairy cattle in the Agreste region of Pernambuco. *Rev Bras Parasitol Vet* 2016; 25(4): 516-522. http://dx.doi.org/10.1590/s1984-29612016055. PMid:27737368.

Reeves WK, Loftis AD, Sanders F, Spinks MD, Wills W, Denison AM, et al. *Borrelia, Coxiella* and *Rickettsia* in *Carios capensis* (Acari: Argasidae) from a brown pelican (*Pelecanus occidentalis*) rookery in South Carolina, USA. *Exp Appl Acarol* 2006; 39(3-4): 321-329. http://dx.doi.org/10.1007/s10493-006-9012-7. PMid:16821092.

Reichel MP, Ayanegui-Alcérreca MA, Gondim LF, Ellis JT. What is the global economic impact of *Neospora caninum* in cattle – the billion dollar question. *Int J Parasitol* 2013; 43(2): 133-142. http://dx.doi.org/10.1016/j.ijpara.2012.10.022. PMid:23246675.

Revista Brasileira de Parasitologia Veterinária – RBPV [online]. 2019 [cited 2019 Apr 23]. Available from: http://www.scielo.br/rbpv

Riemann HP, Brant PC, Franti CE, Reis R, Buchanan AM, Stormont C, et al. Antibodies to *Toxoplasma gondii* and *Coxiella burnetii* among students and other personnel in veterinary colleges in California and Brazil. *Am J Epidemiol* 1974; 100(3): 197-208. http://dx.doi.org/10.1093/oxfordjournals.aje.a112028. PMid:4606228.

Rodolakis A, Berri M, Héchard C, Caudron C, Souriau A, Bodier CC, et al. Comparison of *Coxiella burnetii* shedding in milk of dairy bovine, caprine, and ovine herds. *J Dairy Sci* 2007; 90(12): 5352-5360. http://dx.doi.org/10.3168/jds.2006-815. PMid:18024725.

Rodríguez-Batista Z, Leite RC, Oliveira PR, Lopes CML, Borges LMF. Populational dynamics of *Stomoxys calcitrans* (Linneaus) (Diptera: Muscidae) in three biocenosis, Minas Gerais, Brazil. *Vet Parasitol* 2005; 130(3-4): 343-346. http://dx.doi.org/10.1016/j.vetpar.2005.03.006. PMid:15908125.

Royal MD, Darwash AO, Flint APF, Webb R, Woolliams JA, Lamming GE. Declining fertility in dairy cattle: changes in traditional and endocrine parameters of fertility. *Anim Sci* 2000; 70(3): 487-501. http://dx.doi.org/10.1017/S1357729800051845.

Rozental T, Mascarenhas LF, Rozenbaum R, Gomes R, Mattos GS, Magno CC, et al. *Coxiella burnetii*, the agent of Q fever in Brazil: its hidden role in seronegative arthritis and the importance of molecular diagnosis based on the repetitive element IS*1111* associated with the transposase gene. *Mem Inst Oswaldo Cruz* 2012; 107(5): 695-697. http://dx.doi.org/10.1590/S0074-02762012000500021. PMid:22850965.

Rozental T, Silva ASV, Oliveira RC, Favacho ARM, Oliveira MLA, Bastos FI, et al. Seroprevalence of *Bartonella* spp., *Coxiella burnetii*, and *Hantavirus* among people who inject drugs in Rio de Janeiro, Brazil: a retrospective assessment of a biobank. *Rev Inst Med Trop São Paulo* 2018; 60(0): e31. http://dx.doi.org/10.1590/s1678-9946201860031. PMid:30043935.

Ryan ED, Wrigley K, Hallinan A, McGrath G, Clegg TA. Antibodies to *Coxiella burnetii* in Irish bulk tank milk samples. *Vet Rec* 2018; 182(19): 550. http://dx.doi.org/10.1136/vr.104663. PMid:29445016.

Scola B, Raoult D. Serological cross-reactions between *Bartonella quintana, Bartonella henselae*, and *Coxiella burnetii. J Clin Microbiol* 1996; 34(9): 2270-2274. PMid:8862597.

Scolamacchia F, Handel IG, Fèvre EM, Morgan KL, Tanya VN, Bronsvoort BMC. Serological patterns of Brucellosis, Leptospirosis and Q Fever in *Bos indicus* Cattle in Cameroon. *PLoS One* 2010; 5(1): e8623. http://dx.doi.org/10.1371/journal.pone.0008623. PMid:20098670.

Siciliano RF, Castelli JB, Mansur AJ, Santos FP, Colombo S, Nascimento EM, et al. *Bartonella* spp. and *Coxiella burnetii* associated with community-acquired, culture-negative endocarditis, Brazil. *Emerg Infect Dis* 2015; 21(8): 1429-1432. http://dx.doi.org/10.3201/eid2108.140343. PMid:26197233.

Siciliano RF, Ribeiro HB, Furtado RHM, Castelli JB, Sampaio RO, Santos FCP, et al. Endocardite por *Coxiella burnetii* (febre Q): doença rara ou pouco diagnosticada? Relato de caso. *Rev Soc Bras Med Trop* 2008; 41(4): 409-412. http://dx.doi.org/10.1590/S0037-86822008000400017. PMid:18853017.

Silva AS, Costa MM, Polenz MF, Polenz CH, Teixeira MMG, Lopes STA, et al. Primeiro registro de *Trypanosoma vivax* em bovinos no Estado do Rio Grande do Sul, Brasil. *Cienc Rural* 2009; 39(8): 2550-2554. http://dx.doi.org/10.1590/S0103-84782009005000189.

Silva RAMS, Eguez A, Morales G, Eulert E, Montenegro A, Ybañez R, et al. Bovine Trypanosomiasis in Bolivian and Brazilian Lowlands. *Mem Inst Oswaldo Cruz* 1998; 93(1): 29-32. http://dx.doi.org/10.1590/S0074-02761998000100006. PMid:9698839.

Silva RAMS, Silva JA, Schneider RC, Freitas J, Mesquita D, Mesquita T, et al. Outbreak of trypanosomiasis due to *Trypanosoma vivax* (Ziemann,

1905) in bovines of the Pantanal, Brazil. *Mem Inst Oswaldo Cruz* 1996; 91(5): 561-562. http://dx.doi.org/10.1590/S0074-02761996000500005. PMid:9137742.

Softic A, Asmare K, Granquist EG, Godfroid J, Fejzic N, Skjerve E. The serostatus of *Brucella* spp., *Chlamydia abortus, Coxiella burnetii* and *Neospora caninum* in cattle in three cantons in Bosnia and Herzegovina. *BMC Vet Res* 2018; 14(1): 40. http://dx.doi.org/10.1186/s12917-018-1361-z. PMid:29394895.

Souza EAR, Castro EMS, Oliveira GMB, Azevedo SS, Peixoto RDM, Labruna MB, et al. Serological diagnosis and risk factors for *Coxiella burnetii* in goats and sheep in a semi-arid region of Northeastern Brazil. *Rev Bras Parasitol Vet* 2018; 27(4): 514-520. http://dx.doi.org/10.1590/s1984-296120180086. PMid:30517422.

Stenlund S, Kindahl H, Uggla A, Björkman C. A long-term study of *Neospora caninum* infection in a Swedish dairy herd. *Acta Vet Scand* 2003; 44(1-2): 63-71. http://dx.doi.org/10.1186/1751-0147-44-63. PMid:14650545.

Takiuchi E, Alfieri AF, Alfieri AA. Bovine herpervirus type 1: infection and diagnosis methods. *Semina: Ciênc Agrár* 2001; 22(2): 203-209. http://dx.doi.org/10.5433/1679-0359.2001v22n2p203.

Tenter AM, Heckeroth AR, Weiss LM. *Toxoplasma gondii*: from animals to humans. *Int J Parasitol* 2000; 30(12-13): 1217-1258. http://dx.doi.org/10.1016/S0020-7519(00)00124-7. PMid:11113252.

Tilahun B, Tolossa YH, Tilahun G, Ashenafi H, Shimelis S. Seroprevalence and risk factors of *Toxoplasma gondii* infection among domestic ruminants in East Hararghe zone of Oromia Region, Ethiopia. *Vet Med Int* 2018; 2018: 4263470. http://dx.doi.org/10.1155/2018/4263470. PMid:29887984.

Tissot-Dupont H, Raoult D. Q fever. *Infect Dis Clin North Am* 2008; 22(3): 505-514, ix. http://dx.doi.org/10.1016/j.idc.2008.03.002. PMid:18755387.

Travassos J, Ubatuba A, Silva NP, Mello MT. Febre Q no Rio de Janeiro. *Cienc Cult* 1954; 6: 199-200.

Valle LAR, Brandão H, Christovão DDA, D´Apice M. Investigações sôbre a Febre Q em São Paulo. *Arq Fac Hig Saude Publica Univ Sao Paulo* 1955; 9(1-2): 167-180. http://dx.doi.org/10.11606/issn.2358-792X.v9i1-2p167-180.

Van den Brom R, Van Engelen E, Luttikholt S, Moll L, Van Maanen K, Vellema P. *Coxiella burnetii* in bulk tank milk samples from dairy goat and dairy sheep farms in The Netherlands in 2008. *Vet Rec* 2012; 170(12): 310. http://dx.doi.org/10.1136/vr.100304. PMid:22351793.

Van Schaik EJ, Chen C, Mertens K, Weber MM, Samuel JE. Molecular pathogenesis of the obligate intracellular bacterium *Coxiella burnetii. Nat Rev Microbiol* 2013; 11(8): 561-573. http://dx.doi.org/10.1038/nrmicro3049. PMid:23797173.

Varela-Castro L, Zuddas C, Ortega N, Serrano E, Salinas J, Castellà J, et al. On the possible role of ticks in the eco-epidemiology of *Coxiella burnetii* in a Mediterranean ecosystem. *Ticks Tick Borne Dis* 2018; 9(3): 687-694. http://dx.doi.org/10.1016/j.ttbdis.2018.02.014. PMid:29478883.

Vidal S, Kegler K, Greub G, Aeby S, Borel N, Dagleish MP, et al. Neglected zoonotic agents in cattle abortion: tackling the difficult to grow bacteria. *BMC Vet Res* 2017; 13(1): 373. http://dx.doi.org/10.1186/s12917-017-1294-y. PMid:29197401.

Vieira OLE, Macedo LOD, Santos MAB, Silva JABA, Mendonça CLD, Faustino MAG, et al. Detection and molecular characterization of *Trypanosoma (Duttonella) vivax* in dairy cattle in the state of Sergipe, northeastern Brazil. *Rev Bras Parasitol Vet* 2017; 26(4): 516-520. http://dx.doi.org/10.1590/s1984-29612017048. PMid:29091120.

Wyrosdick HM, Schaefer JJ. *Toxoplasma gondii*: history and diagnostic test development. *Anim Health Res Rev* 2015; 16(2): 150-162. http://dx.doi.org/10.1017/S1466252315000183. PMid:26568360.

Supplementary Material

Supplementary material accompanies this paper.

Supplementary Table. Detailed serological results for selected bovine pathogens associated to abortion in cattle in Brazil.

This material is available as part of the online article from Revista Brasileira de Parasitologia Veterinária (RBPV, 2019) - Home Page - SciELO.