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Original Article

Toxoplasma gondii and *Neospora caninum* infections and factors associated in goats in the Parana state, Southern Brazil

Infecções por *Toxoplasma gondii* e *Neospora caninum* e fatores associados em caprinos no estado do Paraná, Sul do Brasil

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Abstract

The aim of this study was to determine the occurrence of anti-*Toxoplasma gondii* and anti-*Neospora caninum* antibodies and their associated factors among goats in farms from the Paraná state, South Brazil. The serological analysis was performed by indirect ELISA on 629 goat serum samples collected from 32 farms distributed in five mesorregions from the state. Seropositivity was observed in 30.7% of the animals for *T. gondii*, 6.3% for *N. caninum*, and 3.0% for both agents. Final multiple regression models showed that the use of assisted reproduction decreased the chance of seropositivity for *T. gondii* (PR=0.70) and the slaughtering site on the farm (PR=2.03) increased it. To *N. caninum*, the supplying concentrated feed to animals (OR=4.80) and animal pre-weaning death (OR=9.96) increased the chance of seropositivity. The variables associated with these seropositivities suggest deficiencies in sanitation and reproductive management. In addition, it recognizes critical points of infection for animal production management.

Keywords: Seroprevalence, *Toxoplasma gondii*, *Neospora caninum*, goats, Brazil.

Resumo

O objetivo desse estudo foi avaliar a ocorrência de anticorpos anti-*Toxoplasma gondii* e anti-*Neospora caninum* e seus fatores associados em caprinos de propriedades do Estado do Paraná, Sul do Brasil. A análise sorológica foi realizada, utilizando-se ELISA indireto em 629 amostras de soro caprino, coletadas em 32 propriedades distribuídas em cinco mesorregiões do Estado. A soropositividade foi observada em 30,7% dos animais para *T. gondii*, 6,3% para *N. caninum* e 3,0% para ambos. Os modelos finais de regressão múltipla mostraram que o uso de reprodução assistida reduziu a chance de soropositividade para *T. gondii* (PR=0,70), enquanto a presença no local de abate na fazenda (PR=2,03) aumentou. Para *N. caninum,* o fornecimento de ração concentrada (OR=4,80) e a morte dos animais no pré-desmame (OR=9,96) aumentaram a chance de soropositividade. O estudo das variáveis associadas às soropositividades sugere deficiências no saneamento e no manejo reprodutivo dos Animais. Além disso, reconhece pontos críticos para a infecção no manejo da produção animal.

Palavras-chave: Soroprevalência, *Toxoplasma gondii*, *Neospora caninum*, caprinos, Brasil.

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Introduction

The national goat herd was estimated at 9,614,722 million heads (IBGE, 2014). According to the register of herds of the Secretariat of Agriculture and Supply, Paraná accounts for 1.6% of the national goat population, ranking eighth in the nation, with approximately 115,718 thousand heads (Paraná, 2019), demonstrating the importance of goat farming in the state. Among the diseases that affect goats is the toxoplasmosis, caused by *Toxoplasma gondii*, which is one of the main agents responsible for reproductive dysfunction. The prevalence of *T. gondii* in Brazil is quite variable, with reported prevalence rates of 14.4% in São Paulo (Mainardi et al., 2003), 17.1% in Rio Grande do Norte (Lima et al., 2008), 49.4% in Piauí (Rêgo et al., 2016), and 39.4% in Paraná (Garcia et al., 2012). In addition, to the direct damage caused by *T. gondii*, such as miscarriages and stillbirths, they can be major sources of infection to humans, through the consumption of raw or under cooked meat, raw milk, and its by-products (Dubey et al., 2014).

Another parasite that can cause reproductive problems is *Neospora caninum*, first described in 2004 by Figliuolo et al. (2004) that reported the prevalence of 6.4% in goat herds in the state of São Paulo. Since then, several studies have been conducted in other states such Paraíba (3.3%; Faria et al., 2007; and 26.11%; Braz et al., 2018), Rio Grande do Norte (1.05%; Lima et al., 2008), and Pernambuco (64,2%; Tembue et al., 2011; and 2.04%; Arraes-Santos et al., 2016). Although the various serologic studies in Brazilian goats' herds, the reproductive impairment caused by *N. caninum* is still scarce evaluated (Braz et al., 2018).

Toxoplasmosis and neosporosis represent important reproductive and economic losses in the country's goat herds (Dubey, 2010; Dubey et al., 2017). In addition, these diseases have an important role in public health, since this herds can be a source of *T. gondii* infection for men (Dubey & Beattie, 1988; Mesquita et al., 2013). Thus, the identification of factors associated with infection is essential to support programs for the development and promotion of goat farming worldwide (Topazio et al., 2014). The present study aimed to evaluate the seropositivity of *T. gondii* and *N. caninum* and the factors associated with their positivity in Paraná state, Brazil.

Materials and Methods

The study was approved by the Ethics Committee for Animal Use - EMBRAPA Goats and Sheep, Sobral - CE (number 010/2014). These activities were part of the project EMBRAPA (Zoosanitary characterization of goat and sheep farming in Brazil: epidemiology, associated factors and economic impact of diseases, number 02.12.01.032.00). To calculate the sample size (n), an infinite population was used as a reference using EpiInfo 7.2.3.1. The estimated prevalence were based on the study by Fortes et al. (2017), in which *T. gondii* was estimated to have a prevalence of 33.3%, and an error of 4%, resulting in a sample size of 533 animals, and the study by Figliuolo et al. (2004), who estimated a prevalence of 6.4% and an error of 2% for *N. caninum*, resulting in a sample size of 575 animals.

A non-probabilistic sampling was carried out to choose the farms because of a lack of a list of all goat producers in the state of Paraná. Farms were chosen in the municipalities with the greatest representation in goat farming from the mesoregions: Central North, Metropolitan of Curitiba, West, Southwest and Center-South. Within each farm, the animals used were stratified according to age (approximately 60% of adult females, 35% from 6 to 12 months old animals, and all adult males

The blood was sampled by jugular puncture in a tube without anticoagulant and kept refrigerated until the centrifugation for obtaining the serum. The detection of anti-*T gondii* IgG antibodies was performed by an indirect ELISA technique as described by Garcia et al. (2007). The standardized *T. gondii* antigen concentration was 2.5 μg mL^{-1}, the sera were titrated at a ratio of 1:100 and the conjugate (Sigma Aldrich®, USA) was diluted to a ratio of 1:4,000. All the samples were tested in duplicate. The optimal conditions concerning antigen concentration, serum titration and conjugate dilution were established by the highest ratio of mean absorbance of positive samples to mean absorbance of negative samples. The cutoff point for each plate was obtained according to Garcia et al. (2006) using the mean negative sera plus three standard deviations. For the detection of anti-*N. caninum* antibodies, commercial indirect Elisa kits (Imunodot®, Jaboticabal, Brazil) were used. The cutoff index (CFI) was calculated by multiplying the mean optical density of the negative controls by 2.5; the positive samples exhibited a bright yellow color and had an optical density equal to or greater than the CFI, as described by the manufacturer.

A questionnaire was applied to each property with information such as: infrastructure, characteristics of production and commercialization, technological profile, sanitation and presence of other species on the property, domestic and wild. Of each animal whose blood was collected, sex, age and breed information were obtained. The EpiInfo 7.2.3.1 program was used to tabulate the variables from the epidemiological questionnaire along with the serological results and for bivariate analysis. The variables from the epidemiological questionnaire were screened

by Yates-corrected chi-square (χ^2) and Fisher's exact tests, and those with P <0.20 were selected for inclusion in the regression analysis. The existence of associated factors was estimated by multiple logistic regression or multiple log-binomial regression, and the strength of the association was estimated by odds ratio (OR) or prevalence ratio (PR) and their respective 95% confidence intervals using R environment 3.6.2 version, packages epitools (Aragon, 2020) and epiDisplay (Chongsuvivatwong, 2018).

Results and Discussion

This study included 19 municipalities from five of the ten mesoregions in the state of Paraná, and 629 animals from 32 farms were evaluated. The distribution of the agents on the farms indicated that 90.6% (29/32) of the farms were positive for *T. gondii* and 53.1% (17/32) were positive for *N. caninum*, demonstrating an important distribution of agents on farms in Paraná (Table 1). Frequencies of 6.3% for anti-*Neospora caninum* IgG antibodies (39/629; 95% CI = 4.6-8.4) and 30.7% for anti-*Toxoplasma gondii* IgG antibodies (193/629; 95% CI = 27.2 - 34.4) were found in the studied goat population (Table 1). The frequency of antibodies to both agents was 3.0% (19/629). To date, no prevalence studies on *N. caninum* have been performed in goats in the state of Paraná. However, Fortes et al. (2017) described a 33.3% (352/1058) prevalence of *T. gondii* in the state of Paraná, like our study positivity (193/629; 95% CI = 27.2-34.4); both studies used iELISA as the diagnosis method.

The bivariate data analysis revealed an association between the *T. gondii* positivity and the following variables: use of assisted reproduction methods on the farm, the presence of cats on the farm, the presence of wild felines on the farm, the presence of animals who drink water directly from the source, the existence of a slaughtering site on the farm, supplying placental remains to cats, supplying concentrated feed to animals, and the presence of animals over eight months (Table 2). In the final multiple regression model, the following variables were adjusted for: the use of assisted reproduction methods on the farm (P = 0.0042; PR = 0.70, 95% CI = 0.55-0.89) and the existence of a slaughtering site on the farm (P <0.0001; PR = 2.03, 95% CI = 1.59-2.60) (Table 3).

The assisted reproduction variable (controlled breeding and artificial insemination), compared to uncontrolled natural breeding, was a protective factor for the positivity of *T. gondii* in the present study. Pereira et al. (2012) found a significant association for natural breeding, which increased the chance of toxoplasmosis occurrence by 6.09, in the multivariate analysis. Farms in which assisted reproductive techniques are used may represent the adoption of good production practices and management and improved selection of breeders, indicating proper sanitary care as well as beneficial management techniques. In addition, it was not a significant association between reproductive impairment and toxoplasmosis and/or neosporosis. Similar results are observed with *N. caninum* prevalence in goats of Paraíba state (Braz et al., 2018); however, it was contrary with other studies about *T. gondii*.

The slaughter of animals on rural farms, even if transported to a specific site for this purpose, does not always occur under satisfactory hygienic-sanitary conditions. In addition, blood and viscera may collect on the floor, which is highly attractive to cats. The presence of rodents in supplement stores is another condition that is attractive to cats that end up defecating and eliminating oocysts that are later ingested by the livestock via food (Romanelli et al., 2007). Weigel et al. (1999) suggested that the presence of cats on rural farms is the main risk factor for environmental contamination and human infection by *T. gondii*. Cavalcante et al. (2008) reported that the risk of *T. gondii* infection

Table 1. Frequency of anti-*Toxoplasma gondii* and anti-*Neospora caninum* antibodies by mesoregion in goat farms of Paraná state, Brazil.

Mesoregion	*T. gondii* Farms Positive/Total (%)	*T. gondii* Animals Positive/Total (%)	*N. caninum* Farms Positive/Total (%)	*N. caninum* Animals Positive/Total (%)
West	12/14 (85.7)	64/277 (23.1)	8/14 (57.1)	14/277 (5.1)
South-west	9/9 (100.0)	76/177 (42.9)	4/9 (44.4)	8/177 (4.5)
Central-south	4/4 (100.0)	30/80 (37.5)	2/4 (50.0)	13/80 (16.3)
Central-north	1/2 (50.0)	6/42 (14.3)	2/2 (100.0)	4/42 (9.5)
Metropolitan	2/2 (100.0)	13/33 (39.4)	0/2 (00.0)	0/33 (00.0)
Total	**29/32 (90.6)**	**193/629 (30.7)**	**17/32 (53.1)**	**39/629 (6.2)**

Table 2. Frequencies and bivariate analysis (*P*<0.20) from variables statistically associated with the positivity of anti-*Toxoplasma gondii* antibodies in goats from Paraná state, Brazil.

	Toxoplasma gondii		
Variables	**Positive/Total (%)**	***P***	**PR (95% CI)**
Assisted reproduction			
Controlled natural mounts, artificial insemination	67/264[a] (25.4)	0.0179	0.74 (0.57-0.94)
Natural mounts without control	126/365 (34.5)		
Provides concentrated feed			
Yes	159/487 (32.6)	0.0227	1.52 (1.06-2.19)
No	26/121 (21.5)		
Drink water straight from the source			
Yes	59/149 (39.6)	0.0068	2.18 (1.20-3.95)
No	10/55 (18.2)		
Adult over 8 months			
Yes	86/230 (37.4)	0.0001	1.83 (1.34-2.51)
No	43/211 (20.4)		
Suitable place for slaughter on the farm			
Yes	122/278 (43.9)	<0.0001	1.97 (1.54-2.52)
No	69/309 (22.3)		
Presence of cats			
Yes	135/421 (32.1)	0.0052	1.81 (1.17-2.78)
No	19/107 (17.8)		
Presence of wild felids			
Yes	52/89 (58.4)	0.0072	1.43 (1.13-1.81)
No	92/225 (41.0)		
Cats feed on placental remains			
Yes	44/81 (54.3)	<0.0001	1.78 (1,39-2.29)
No	119/390 (30.5)		

[a]The totals of the frequencies varied according to the number of questionnaires answered by the owners of the animals. P: p value; PR: prevalence ratio; CI: confidence interval.

in goat was increased by 1.73 times in farms from Ceará state with more than 10 cats This finding is explained by the free breeding of these animals which had access to various zootechnical facilities, pastures and natural water supply and storage sources. Although wild felids do not inhabit the human peridomiciliary, they have free access to pastures, where they often feed on animal carcasses and placental remains, become infected and subsequently spread *T. gondii* oocysts throughout the environment through their feces.

Regarding age, a significant association was observed for animals older than eight months, which increased the chance of toxoplasmosis by 2.53 times. Anderlini et al. (2011) found a positive association for the prevalence of *T. gondii* in adult animals compared to younger goats in herds from Alagoas state. Jittapalapong et al. (2005) observed that older animals (older than two years) were 2.70 times more likely to be *T. gondii*-seropositive. This

is explained by the fact that there is an increased chance of animals becoming infected over time due to oocyst ingestion from the contaminated environment.

In relation to water source, Cavalcante et al. (2008) found that contamination was associated with the poor sanitation of drinking fountains and facilities used for capturing and storing large volumes of water, which are practically impossible to sanitize, thus becoming sources of infection to animals. This result was corroborated by Villari et al. (2009), who described a correlation between untreated water supplied to animals and the presence of anti-*T. gondii* antibodies. The epidemiological analysis results of water contamination by *T. gondii* oocysts can be extrapolated to water contamination by *N. caninum* oocysts from dog feces. The implementation of good hygiene practices on the farms is essential to avoid environmental contamination of food and water by *T. gondii* and *N. caninum* oocysts (Machacova et al., 2015).

The variables supplying concentrated feed to animals and animal pre-weaning death were factors associated with the positivity of neosporosis in goats in Paraná state according to the bivariate analysis (Table 4). In the final multiple logistic regression model, two variables were associated factors, supplying concentrated feed to animals, which increased the odds of *N. caninum* seropositivity by 4.80 times ($P = 0.0356$; 95% CI = 1.32-74.70), and animal pre-weaning death, which increased the odds by 9.96 times ($P = 0.0253$; 95% CI = 1.11-20.75) (Table 5). Table 5 shows that the odds ratio confidence intervals calculated by the logistic regression method are wide, and this maybe is due to the small number of seropositive animals for *N. caninum*. Feed supplementation with concentrates is a necessary practice in ruminant animal breeding during grazing shortages. However, the storage of this food product requires appropriate conditions. Although the presence of dogs was not a significant variable in this study, dogs are commonly found sheltering in these places, where they end up defecating, eliminating *N. caninum* oocysts in the feces and contaminating stored food, as described by Dubey & Schares (2011). In the present study, a mortality rate of 8.6% pre-weaning period was observed, but no etiological diagnosis was made. The observation of neonatal deaths or miscarriages may not be a direct result of neosporosis; however, there is a likelihood of spread among herds when dogs eventually feed on placental or miscarriage material or the remains of neonates who were infected

Table 3. Final models of multiple regression analysis from variables statistically associated with the positivity of anti-*Toxoplasma gondii* antibodies in goats from Paraná state, Brazil, ($P<0.05$).

Toxoplasma gondii	**Multiple log-binomial regression**					
Variables	**Coefficient**	**Standard Error**	***P*-Wald**	**Degrees of freedom**	***P***	**Adjusted PR (95% CI)**
Controlled natural mounts and/or artificial insemination	0.3596	0.1226	0.004	1	0.0038	0.70 (0.55-0.89)
Suitable place for slaughter on the farm	0.7095	0.1249	<0.001	1	<0.0001	2.03 (1.59-2.60)
Intercept	-1.3838	0.1108		1	<0.0001	

P-Wald: P Wald value; P: P value; PR: prevalence ratio; CI: confidence interval.

Table 4. Frequencies and bivariate analysis ($P<0.20$) from variables statistically associated with the positivity of anti-*Neospora caninum* antibodies in goats from Paraná state, Brazil.

	Neospora caninum		
Variables	**Positive/Total (%)**	***P***	**OR (95% CI)**
Provides concentrated animal feed			
Yes	37/487[a] (7.6)	0.0291	4.89 (1.16-20.59)
No	2/121 (1.6)		
Pre-weaning death			
Yes	27/308 (8.8)	0.0126	9.80 (1.31-73.05)
No	1/103 (0.9)		

[a]The totals of the frequencies varied according to the number of questionnaires answered by the owners of the animals.

P: P value; OR: odds ratio; CI: confidence interval.

Table 5. Final models of multiple regression analysis from variables statistically associated with the positivity of anti-*Neospora caninum* antibodies in goats from Paraná state, Brazil, (*P*<0.05).

Neospora caninum	Multiple logistic regression					
Variables	**Coefficient**	**Standard Error**	***P*-Wald**	**Degrees of freedom**	***P***	**Adjusted OR (95% CI)**
Provides concentrated animal feed to animals	1.5692	0.7468	0.032	1	0.0356	4.80 (1.11-20.75)
Pre-weaning death	2.2992	1.0278	0.028	1	0.0253	9.96 (1.32-74.70)
Intercept	-5.9627	1.2527		1	<0.0001	

P: P value; OR: odds ratio; CI: confidence interval.

with *N. caninum* during pregnancy. However, Buxton et al. (2002) reported that the first infection of *N. caninum* in sheep and goats can be fatal to the fetus; however, in subsequent pregnancies, fetal infection may be relatively uncommon, a fact attributed to maternal immunity in these species.

Conclusion

This research highlights the positivity for anti-*T. gondii* and anti-*N. caninum* antibodies in goats in the state of Paraná. The variables associated with these seropositivity suggest deficiencies in sanitation and reproductive management; such as presence of cats, that feed on placental remains, the lack of a suitable place for slaughter on the farm, to drink water straight from the source, the occurrence of pre-weaning death and do not to use assisted reproduction methods. In addition, recognizes critical points of infection for the animal production management, like to provide concentrated feed, do not avoid the presence of wild felids on the farm and to have animals with age over eight months.

References

Anderlini GA, Mota RA, Faria EB, Cavalcanti EFTSF, Valença RMB, Pinheiro Júnior JW, et al. Occurrence and risk factors associated with infection by *Toxoplasma gondii* in goats in the State of Alagoas, Brazil. *Rev Soc Bras Med Trop* 2011; 44(2): 157-162. http://dx.doi.org/10.1590/S0037-86822011005000017. PMid:21503550.

Aragon TJ. *Epitools: Epidemiology Tools. R package version 0.5-10.1* [online]. 2020 [cited 2020 May 5]. Available from: https://CRAN.R-project.org/package=epitools

Arraes-Santos AI, Araújo AC, Guimarães MF, Santos JR, Pena HFJ, Gennari SM, et al. Seroprevalence of anti-*Toxoplasma gondii* and anti-*Neospora caninum* antibodies in domestic mammals from two distinct regions in the semi-arid region of Northeastern Brazil. *Vet Parasitol Reg Stud Rep* 2016; 5: 14-18. http://dx.doi.org/10.1016/j.vprsr.2016.08.007. PMid:31014531.

Braz BMA, Valente JDM, Villalobos EMC, Lara MCCSH, Machado CAL, Barbosa IC, et al. Seroepidemiology of *Neospora caninum* among goats (*Capra hircus*) in the state of Paraíba, northeastern Brazil. *Arq Bras Med Vet Zootec* 2018; 70(1): 147-152. http://dx.doi.org/10.1590/1678-4162-9453.

Buxton D, McAllister MM, Dubey JP. The comparative pathogenesis of neosporosis. *Trends Parasitol* 2002; 18(12): 546-552. http://dx.doi.org/10.1016/S1471-4922(02)02414-5. PMid:12482540.

Cavalcante ACR, Carneiro M, Gouveia AMG, Pinheiro RR, Vitor RWA. Risk factors for infection by *Toxoplasma gondii* in herds of goats in Ceará, Brazil. *Arq Bras Med Vet Zootec* 2008; 60(1): 36-41. http://dx.doi.org/10.1590/S0102-09352008000100006.

Chongsuvivatwong V. *epiDisplay: Epidemiological Data Display Package. R package version 3.5.0.1* [online]. 2018 [cited 2020 May 5]. Available from: https://CRAN.R-project.org/package=epiDisplay

Dubey JP, Beattie CP. *Toxoplasmosis of animals and man*. Florida: CRC-Press; 1988.

Dubey JP, Hemphill A, Calero-Bernal R, Schares G. *Neosporosis in animals*. Florida: CRC-Press; 2017. http://dx.doi.org/10.1201/9781315152561.

Dubey JP, Schares G. Neosporosis in animals – the last five years. *Vet Parasitol* 2011; 180(1-2): 90-108. http://dx.doi.org/10.1016/j.vetpar.2011.05.031. PMid:21704458.

Dubey JP, Verma SK, Ferreira LR, Oliveira S, Cassinelli AB, Ying Y, et al. Detection and survival of *Toxoplasma gondii* in milk and cheese from experimentally infected goats. *J Food Prot* 2014; 77(10): 1747-1753. http://dx.doi.org/10.4315/0362-028X.JFP-14-167. PMid:25285492.

Dubey JP. *Toxoplasmosis of animals and humans*. 2nd ed. Florida: CRC-Press; 2010.

Faria EB, Gennari SM, Pena HFJ, Athayde ACR, Silva MLCR, Azevedo SS. Prevalence of anti-*Toxoplasma gondii* and anti-*Neospora caninum* antibodies in goats slaughtered in the public slaughterhouse of Patos city, Paraíba State, Northeast region of Brazil. *Vet Parasitol* 2007; 149(1–2): 126-129. http://dx.doi.org/10.1016/j.vetpar.2007.07.009. PMid:17706359.

Figliuolo LPC, Rodrigues AAR, Viana RB, Aguiar DM, Kasai N, Gennari SM. Prevalence of anti-*Toxoplasma gondii* and anti-*Neospora caninum* antibodies in goat from São Paulo State, Brazil. *Small Rumin Res* 2004; 55(1–3): 29-32. http://dx.doi.org/10.1016/j.smallrumres.2003.12.013.

Fortes MS, Lopes-Mori FMR, Caldart ET, Constantino C, Evers F, Pagliari S, et al. Caprine toxoplasmosis in Southern Brazil: a comparative seroepidemiological study between the indirect immunofluorescence assay, enzyme-linked immunosorbent assay, and the modified agglutination test. *Trop Anim Health Prod* 2017; 50(2): 413-419. http://dx.doi.org/10.1007/s11250-017-1450-1. PMid:29079943.

Garcia G, Sotomaior C, Nascimento AJ, Navarro IT, Soccol VT. *Toxoplasma gondii* in goats from Curitiba, Paraná, Brazil: risks factors and epidemiology. *Rev Bras Parasitol Vet* 2012; 21(1): 42-47. http://dx.doi.org/10.1590/S1984-29612012000100009. PMid:22534944.

Garcia JL, Navarro IT, Biazzono L, Freire RL, Guimarães JS Jr, Cryssafidis AL, et al. Protective activity against oocyst shedding in cats vaccinated with crude rhoptry proteins of the *Toxoplasma gondii* by the intranasal route. *Vet Parasitol* 2007; 145(3-4): 197-206. http://dx.doi.org/10.1016/j.vetpar.2007.01.007. PMid:17296268.

Garcia JL, Navarro IT, Vidotto O, Gennari SM, Machado RZ, Luz Pereira AB, et al. *Toxoplasma gondii*: comparison of a rhoptry-ELISA with IFAT and MAT for antibody detection in sera of experimentally infected pigs. *Exp Parasitol* 2006; 113(2): 100-105. http://dx.doi.org/10.1016/j.exppara.2005.12.011. PMid:16458299.

Instituto Brasileiro de Geografia e Estatística – IBGE. *Estatísticas do registro civil* [online]. Rio de Janeiro: IBGE; 2014 [cited 2019 Nov 14]. Available from: https://biblioteca.ibge.gov.br/visualizacao/periodicos/135/rc_2014_v41.pdf

Jittapalapong S, Sangvaranond A, Pinyopanuwat N, Chimnoi W, Khachaeram W, Koizumi S, et al. Seroprevalence of *Toxoplasma gondii* infection in domestic goats in Satun Province, Thailand. *Vet Parasitol* 2005; 127(1): 17-22. http://dx.doi.org/10.1016/j.vetpar.2004.08.019. PMid:15619370.

Lima JTR, Ahid SMM, Barrêto RA Jr, Pena HFJ, Dias RA, Gennari SM. Prevalence to *Toxoplasma gondii* and *Neospora caninum* antibodies in goats from Mossoró, Rio Grande do Norte. *Braz J Vet Res Anim Sci* 2008; 45(2): 81-86. http://dx.doi.org/10.11606/issn.1678-4456.bjvras.2008.26703.

Machacova T, Bartova E, Sedlak K, Budikova M, Piccirillo A. Risk factors involved in transmission of *Toxoplasma gondii* and *Neospora caninum* infection in rabbit farms in Northern Italy. *Ann Agric Environ Med* 2015; 22(4): 677-679. http://dx.doi.org/10.5604/12321966.1185774. PMid:26706976.

Mainardi RS, Modolo JR, Stachissini AVM, Padovani CR, Langoni H. Soroprevalência de *Toxoplasma gondii* em rebanhos caprinos no Estado de São Paulo. *Rev Soc Bras Med Trop* 2003; 36(6): 759-761. http://dx.doi.org/10.1590/S0037-86822003000600021. PMid:15049121.

Mesquita LP, Nogueira CI, Costa RC, Orlando DR, Bruhn FR, Lopes PF, et al. Antibody kinetics in goats and conceptuses naturally infected with *Neospora caninum. Vet Parasitol* 2013; 196(3-4): 327-333. http://dx.doi.org/10.1016/j.vetpar.2013.03.002. PMid:23537945.

Paraná. Governo do Estado. Secretaria de Estado da Agricultura e do Abastecimento – SEAB. Departamento de Economia Rural – DERAL. *Números da Pecuária Paranaense - Ano 2019* [online]. Curitiba: SEAB; 2019 [cited 2020 July 28]. Available from: http://www.agricultura.pr.gov.br/system/files/publico/Conjuntura/nppr.pdf

Pereira MF, Peixoto RM, Langoni H, Greca H Jr, Azevedo SS, Porto WJN, et al. Fatores de risco associados à infecção por *Toxoplasma gondii* em ovinos e caprinos no estado de Pernambuco. *Pesq Vet Bras* 2012; 32(2): 140-146. http://dx.doi.org/10.1590/S0100-736X2012000200009.

Rêgo WMF, Paula NRO, Vitor RWA, Silva RAB, Diniz BLM, Sousa M, et al. Risk factors for *Toxoplasma gondii* infection in goats and sheep raised in the State of Piauí in northeast Brazil. *Small Rumin Res* 2016; 141: 17-23. http://dx.doi.org/10.1016/j.smallrumres.2016.04.010.

Romanelli PR, Freire RL, Vidotto O, Marana ER, Ogawa L, De Paula VS, et al. Prevalence of *Neospora caninum* and *Toxoplasma gondii* in sheep and dogs from Guarapuava farms, Paraná State, Brazil. *Res Vet Sci* 2007; 82(2): 202-207. http://dx.doi.org/10.1016/j.rvsc.2006.04.001. PMid:17266999.

Tembue AA, Ramos RA, Sousa TR, Albuquerque AR, Costa AJ, Meunier IM, et al. Serological survey of *Neospora caninum* in small ruminants from Pernambuco State, Brazil. *Rev Bras Parasitol Vet* 2011; 20(3): 246-248. http://dx.doi.org/10.1590/S1984-29612011000300013. PMid:21961757.

Topazio JP, Weber A, Camillo G, Vogel FF, Machado G, Ribeiro A, et al. Seroprevalence and risk factors for *Neospora caninum* in goats in Santa Catarina state, Brazil. *Rev Bras Parasitol Vet* 2014; 23(3): 360-366. http://dx.doi.org/10.1590/S1984-29612014062. PMid:25271457.

Villari S, Vesco G, Petersen E, Crispo A, Buffolano W. Risk factors for toxoplasmosis in pigs bred in Sicily, Southern Italy. *Vet Parasitol* 2009; 161(1-2): 1-8. http://dx.doi.org/10.1016/j.vetpar.2009.01.019. PMid:19246158.

Weigel RM, Siegel AM, Dyer D, Dubey JP. Risk factors for infection with *Toxoplasma gondii* for residents and workers on swine farms in Illinois. *Am J Trop Med Hyg* 1999; 60(5): 793-798. http://dx.doi.org/10.4269/ajtmh.1999.60.793. PMid:10344655.