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Degree of technification of the sheep herd may be associated with seropositivity for the maedi-visna virus in South Brasil

Grau de tecnificação do rebanho ovino pode estar associado a soropositividade para o maedi-visna vírus no Sul do Brasil

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Highlights

Serological findings indicates that MVV circulates in Paraná state.

Variables sex, age, and breed aren't statistically associated with MVV seropositivity.

Technification degree may be associated with the occurrence of anti-MVV antibodies.

Abstract

Maedi-Visna (MV) is a chronic progressive multisystem disease that may be asymptomatic for several months or years, but progress rapidly, and may result in death, when signs and symptoms evolve. Viral elimination occurs mainly through direct contact with positive animal secretions. There is no vaccine or treatment, and prophylaxis is necessary for the health of the herd. The present study aimed to verify the seropositivity of MV and evaluate the factors associated with the risk in sheep herds in Paraná. A total of 1549 serum samples were collected from 90 properties. An epidemiological questionnaire was applied to each property, and the variables were analyzed using the Epi-info program and R environment. Of

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the 1549 samples analyzed, 22 were positive (1.4%) for the micro-AGID test in 13.3% of the properties. Our study demonstrated variables associated with the prevention and the risk of seropositivity to MVV. Conducting a breeding season, supplying concentrated feed, and separating the breeding stock before birth were factors associated with protection, whereas the previous occurrence of problems with lice, breeding on pasture, and keeping cats close to the flock were factors associated with risk. The seropositivity observed in the present study suggests the circulation of MVV in sheep herds in Paraná, which reinforces the need to implement prevention and control measures since the level of technification may be associated with the occurrence of anti-MVV antibodies in herds.

Key words: Lentiviruses. Small ruminants. Pneumonia. Retrovirus. Maedi-visna.

Resumo

A Maedi-Visna (MV) é uma doença multissistêmica de caráter crônico-progressivo, os animais infectados podem passar meses e anos sem demonstrarem sinais clínicos e, após desenvolverem sinais, evoluem rapidamente para a morte. A eliminação viral ocorre principalmente por meio do contato direto com secreções de animais positivos. Não existe vacina ou tratamento, sendo necessária a profilaxia para a sanidade do rebanho. O objetivo do presente estudo foi verificar a soropositividade para o MV e avaliar os fatores associados ao risco em rebanhos ovinos do Paraná. Foram colhidas 1549 amostras de soro, oriundas de 90 propriedades. A cada propriedade foi aplicado um questionário epidemiológico, cujas variáveis foram analisadas pelo programa Epi-info e ambiente R. Das 1549 amostras analisadas, 22 foram positivas (1,4%) ao teste de micro-IDGA, em 13,3% das propriedades. Nosso estudo demonstrou variáveis associadas à proteção e ao risco para a ocorrência de anticorpos anti-MVV nas propriedades podem estar relacionadas à tecnificação da mesma. Realização de estação de monta, fornecimento de ração concentrada e separação das matrizes antes do parto foram fatores associados à proteção, enquanto que a ocorrência prévia de problemas com piolhos, criação a pasto e manter gatos junto ao rebanho foram fatores associados ao risco. A soropositividade observada no presente estudo sugere a circulação do MVV nos rebanhos ovinos do Paraná, o que reforça a necessidade de implementação de medidas de prevenção e controle, uma vez que a tecnificação pode interferir na ocorrência de anticorpos anti-MVV nos rebanhos.

Palavras-chave: Lentiviruses. Pequenos ruminantes. Pneumonia. Retrovírus. Maedi-Visna.

Introduction

Lentiviruses of small ruminants (SRLVs) belong to the family Retroviridae and subfamily Lentivirinae (International Committee on Taxonomy Viruses [ICTV], 2020), which are enveloped viruses with tropism by cells of the monocytic-phagocytic system that cause persistent infection and chronic degenerative disease. They

are divided into two phylogenetic groups: the Maedi-Visna virus (MVV), which infects sheep, and the caprine arthritis encephalitis virus (CAEV), which infects goats (Valas et al., 1997). Inter-species transmission may occur (Leroux et al., 2010). Both infections are classified as notifiable diseases by the World Organization for Animal Health [WOAH] (2022).

In 1997, a sheep herd in Paraná was serologically positive for MVV, the virus was isolated from a sheep with mastitis and arthritis and phylogenetically identified by Ravazzolo et al. (2001).

MV is a multisystemic disease with a chronic-progressive character (Minguijón et al., 2015), which is extremely important and extensively discussed today (Czopowicz et al., 2018), as it is present in the herds of several countries and is one of the reasons for restricting international trade (Giangaspero et al., 2011). Infected animals may be asymptomatic for several months or years; however, rapid progression after the evolution of clinical signs may culminate in serious illness, possibly resulting in death (Blacklaws, 2012). Immunosuppressor factors such as aging, co-infections or stressful environments accelerates virus replication and clinical manifestations of the disease (Gomez-Lucia et al., 2018). Clinical signs include arthritis, interstitial pneumonia, mastitis, progressive weight loss, encephalitis, and paralytic syndromes, which in some cases may cause death (Ramírez et al., 2012; Mcvey et al., 2016). Viral spread mainly occurs through secretions, which contain cells of the monocyte-phagocytic system rich in viral particles (Blacklaws, 2012).

Transmission occurs mainly through the horizontal route through contact with mucous membranes, aspiration of aerosols, and ingestion of colostrum (Cortez-Romero et al., 2013; Junkuszew et al., 2016). Gregory et al. (2011) and Cortez-Romero et al. (2013) demonstrated viral transmission through infected breeding semen, and Souza et al. (2013) detected MV in frozen semen samples.

As recently proved by Araújo et al. (2020), the intrauterine route is a potentially source of infection in newborn goats. There is no vaccine or treatment for any form of MV, and this necessitates prophylaxis for the health of the herd (Tu et al., 2017). No studies have accurately estimated the economic impact of herd infection; however, the losses are related to the restriction of international trade, death of animals, early discard, expenses with treatments and diagnosis, decrease in productivity, delay in growth, and devaluation of the herd (Perez et al., 2010; Giangaspero et al., 2011; Michiels et al., 2018).

Due to the chronic and multisystemic nature of the disease (Minguijón et al., 2015), serological tests are indicated for diagnosis (Pinheiro et al., 2018). The agar gel immunodiffusion test (AGID) and ELISA are between the tests indicated by the WOA for MV routine diagnosis, the first is practical, low cost and simple to perform, the second requires greater qualification of human resources and equipment, but allows to carry out large-scale tests quickly. The sensitivity and specificity of both, depend on the antigen strain and preparation technique, Western Blot (WB) is indicated as gold standard to access the accuracy parameters (WOAH, 2017). For direct diagnosis, PCR is the most used, it is mainly employed as supplemental test to determine the infection status of cases that cannot be solved by serology, it is very important to confirm the specific virus and strain involved in the infection, but not feasible for surveillance routine diagnosis (WOAH, 2017).

In Brazil, the Ministry of Agriculture, Livestock, and Supply (MAPA) has developed a national caprine and ovine health program

(PNSCO). It lists some important diseases for the country, including the SRLVs, as it requires effective measures against the risk of spreading the agent (Ministério da Agricultura Pecuária e Abastecimento [MAPA], 2017). In 2006, the Secretariat of Agriculture and Supply of the State of Paraná (SEAB) implemented the state program to support the structuring of sheep and goat productive chains (PAECPOC). The program aimed to strengthen the production structure: providing a greater source of income for producers; guaranteeing product quality, continuity of supply, and genetic improvement of the herd; and facilitating competitiveness in the market (Secretaria de Agricultura e do Abastecimento do Paraná [SEAB], 2008).

The objective of the present study was to verify the seropositivity of MVV and evaluate its risk factors in sheep herds in Paraná.

Material and Methods

Ethical aspects

The study was approved by the Animal Use Ethics Committee of EMBRAPA (Brazilian Farming and Agricultural Research Company) Goats and Sheep, Sobral, CE under number 010/2014. These activities were part of the EMBRAPA Zoosanitary Characterization of goat and sheep farming in Brazil: epidemiology, associated factors, and economic impact of diseases (number 02.12.01.032.00).

Study area

In 2015, sheep serum samples were collected (Figure 1) from 90 properties from 25 municipalities in six mesoregions in the state of Paraná located at latitudes from 23°S to 26°S and classified in the CFA (Humid Subtropical Climate) and CFB (Temperate Oceanic Climate) (Instituto de Terras, Cartografia e Geociências [ITCG], 2006). According to the Brazilian Institute of Geography and Statistics (IBGE), Paraná had an effective herd of 556,512 sheep in 2018, which placed it at the 8th position in the national classification of sheep production, accounting for 2.94% of the national production (Instituto Brasileiro de Geografia e Estatística [IBGE], 2019).

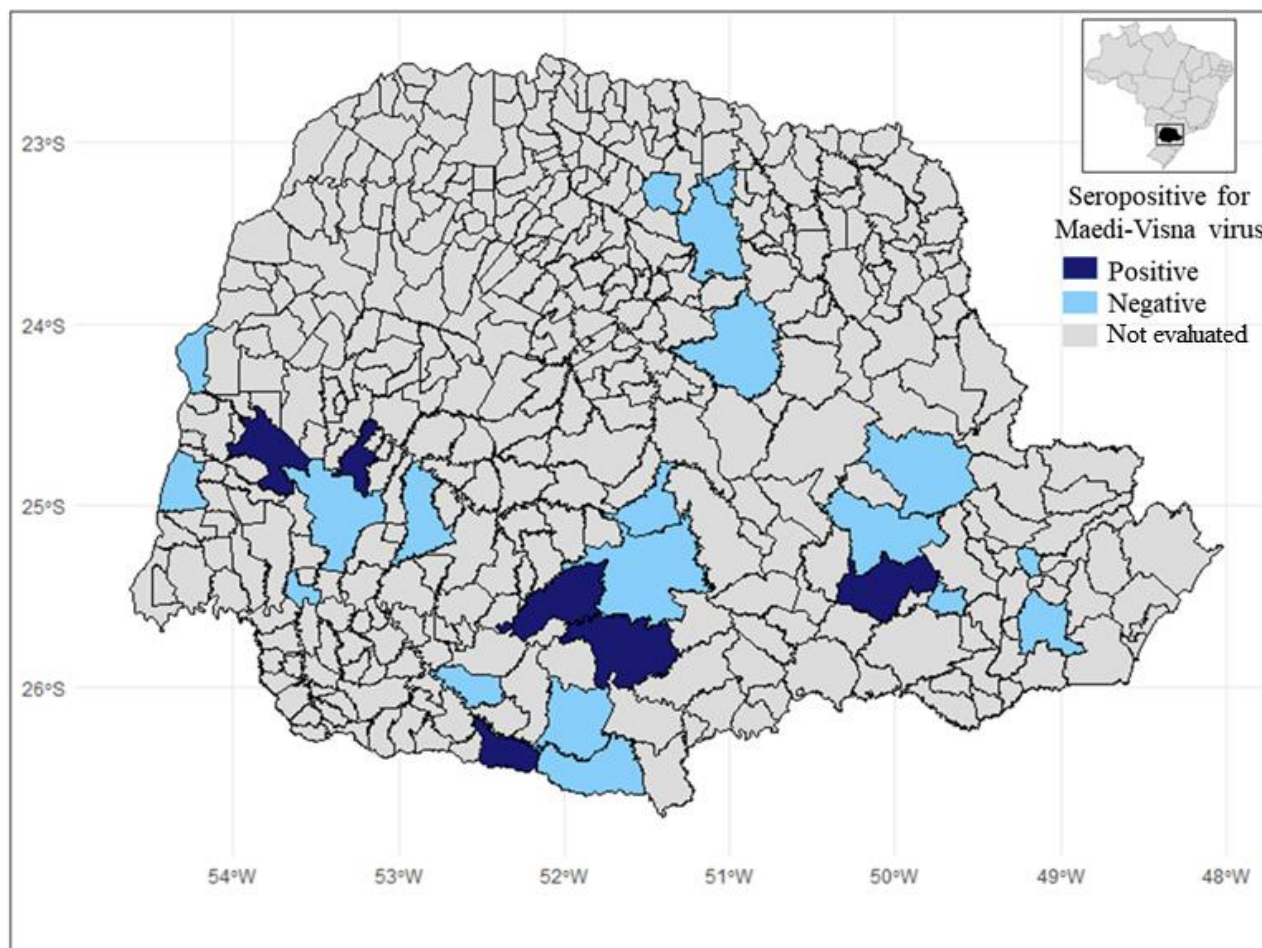


Figure 1. Map of the state of Paraná, highlighted in dark blue to the municipalities where sheep serum samples collected were seropositive for Maedi-Visna Virus, in light blue to the negative ones and in gray to the municipalities not evaluated in the present study.

Sample and sampling

An infinite population was used as a reference to calculate the sample size (n). The estimated prevalence was 50% based on the absence of studies in the state of Paraná. The admitted error was 2.5%, resulting in a sample size of 1,537 animals with a 95% confidence level.

Due to the absence of a list of all sheep producers in the state, a non-probabilistic sampling of the properties was carried out, considering the municipalities with the greatest representation in sheep farming in Paraná. In each property, 20 animals were sampled and stratified as follows: 60.0% (12) were breeding females, 35.0% (7) were young females of 6-12 months of age, and 5% (1) was a male breeder.

Epidemiological survey

A survey, with 23 pages and 126 questions, was conducted on each farm by EMBRAPA technicians with information such as infrastructure, characteristics of production and commercialization, technological profile, sanitation and presence of other animal species on the property. Regarding sheep, information about sex, age, and breed were collected.

Sample collection and serological detection of anti-MVV antibodies

Blood was collected by jugular puncture each animal sampled in a tube without anticoagulant and refrigerated until centrifugation to obtain the serum. A commercial kit (Biovotech®) was employed for the detection of antibody reactivity to p28 protein using micro-AGID technique (MAPA license nº 9050). The tests were performed using disposable Petri dishes with a diameter of 90mm. Fifteen milliliters of 1% agarose was added to each plate, which was remelted following the manufacturer's instructions. After gelling, the gels were perforated using a perforator with seven molds, six peripheries, and one central. In the central well, the antigen was applied to the peripherals of the standard sera and the sera to be tested. Each of the wells for the sample had a capacity of 30 µL, already in the standards and antigen, with a capacity of 10 µL each. After pipetting the sera, the plates were incubated in a humid chamber at 25°C. The reading was performed after 36 h of incubation. Only samples that showed a precipitation line between the sample and the antigen and confluence between

the precipitation lines of the standard sera were considered positive, guaranteeing the specificity of the test. The examination, as well as the reading and interpretation of results, were performed following the manufacturer's recommendations.

Statistical analysis

The EpiInfo 7.1.5.2 program was used to tabulate the variables in the epidemiological questionnaire and the serological results. The variables of the epidemiological survey were analyzed using the chi-squared test corrected by Yates and Fisher's exact tests; those with $p < 0.20$ were selected for inclusion in the multiple logistic regression analysis. The strength of the association was estimated by reason of prevalence (PR) and its respective 95% confidence intervals with Environment R version 3.6.2. Only the variables with data from more than 85% of completed questionnaires were analyzed.

Results and Discussion

Of the total of 1549 sheep samples, 1.4% (22/1549) were positive for the micro-AGID test. A total of 13.3% (12/90) of the farms had at least one seropositive animal. Of the municipalities studied, 24.0% (6/25) were positive for Candói, Clevelândia and Pinhão, which belong to the Mid-South mesoregion; Corbélia and Toledo, to Western mesoregion; Palmeira, to Eastern Center mesoregion. Palmeira and Candi had the highest prevalence, with 5.1% (7/137) of seropositive individuals, while Corbélia had the lowest, with 1.7%. Of the different sheep breeds analyzed, the Santa Inês breed had

the highest occurrence of antibodies (2.7%, 2/73), while no anti-MVV antibodies were detected in the Suffolk breed. The results stratified by the municipality are shown in Table 1, and those stratified by race are shown in Table 2. The gender and age variables

were informed in 56.48% (875/1549) of the questionnaires, 52.23% (457/875) of the sheep were breeding females, 36.23% (317/875) were young females, 3.31% (29/875) was a male breeder and 8.23% (72/875) were young males.

Table 1

Distribution of the results of the micro-AGID test for the detection of anti-MVV antibodies in reactive sheep stratified by the municipalities in the state of Paraná with at least one seropositive animal

Municipality	Nº Samples	Nº Positive	% Positive
Corbélia	60	1	1.7
Candói	78	4	5.1
Toledo	78	3	3.8
Clevelândia	83	4	4.8
Pinhão	100	3	3.0
Palmeira	137	7	5.1

Table 2

Distribution of the results of the micro-AGID test for the detection of anti-MVV antibodies stratified by breed in the state of Paraná

Breed	Nº Samples	Positive	% Positive
Suffolk	15	0	0.0
Santa inês	74	2	2.7
Ille de france	101	1	1.0
Dorper	116	3	2.6
Texel	346	3	0.9
NDB*	897	13	1.4

* Non defined breed.

Table 3
Frequencies and bivariate analysis ($P < 0.20$) of the variables statistically associated with positivity for the anti-MVV antibodies in sheep in the state of Paraná, Brazil

	Positive/Total (%)	P	PR (CI 95%)
Farm performs breeding season			
Yes	3/553 (37.5)	0.0128	0.23 (0.06-0.81)
No	18/802 (64.4)		
Farm provides concentrated feed			
Yes	9/912 (1.0)	0.0457	0.41 (0.17-0.97)
No	13/552 (2.4)		
Domestic cats that remain with the flock on the farm			
Yes	20/913 (2.2)	0.0068	5.57 (1.29-23.95)
No	2/500 (0.4)		
Production system adopted on the farm			
Intensive	2/415 (0.5)	0.0031	5.15 (1.16-22.79)
Semi-intensive	1/379 (0.3)		
On pasture	14/575 (2.4)		
Having an exclusive stall or paddock for births on the farm			
Yes	4/712 (0.56)	0.0040	0.22 (0.07-0.65)
No	18/720 (2.5)		
Farm already had problems with lice			
Yes	11/313 (3.5)	0.0076	3.46 (1.48-8.07)
No	11/1058 (1.1)		

Table 4
Final model of the multiple logistic regression analysis of variables statistically associated with positivity for anti-MVV antibodies in sheep from the state of Paraná, Brazil ($P < 0.05$)

	Coefficient	Standard error	Degrees of freedom	P	Adjusted PR (95% CI)
Production system adopted on the farm on pasture	16.386	0.7647	2	0.0321	5.14 (1.15-23.04)
Domestic cats that remain with the flock on the farm	19.230	0.7611	1	0.0115	6.84 (1.53-30.40)
Farm already had problems with lice	14.389	0.5020	1	0.0042	4.21 (1.57-11.27)
Constant	-70.591	1.0054		<0.0001	

The positivity of anti-MVV antibodies observed in the present study (1.4%) can be classified as very low (1% to 9%) (Reina et al., 2009) as in other Brazilian regions investigated using AGID. Mazzinghy et al. (2016) found 1.62% (6/369) of seropositive individuals in Tocantins. In Bahia, Martinez et al. (2011) observed 0.3-0.5% and Pinheiro et al. (2018) 1.56% (13/831) of seropositive individuals. Costa et al. (2007) found a seroprevalence of 1.0-8.2% in different areas of Pernambuco. Lombardi et al. (2009) observed a 2.7% seropositivity in São Paulo. Because it is an incurable disease (Tu et al., 2017), which has already been phylogenetically identified in Paraná (Ravazzolo et al., 2001), the serological findings prove that the virus remains in circulation in the state.

SRLV studies have compared the sensitivity and specificity of serological tests and have shown greater sensitivity of ELISA and WB when compared to AGID. Nascimento et al. (2014), demonstrated greater sensitivity of the indirect ELISA in relation to AGID and a kappa of 50%. The authors concluded that both can be recommended and used as individual diagnostic methods and in epidemiological surveys. de Azevedo et al. (2019), observed greater positivity in WB when compared to AGID and attributed the low sensitivity to the antigen-antibody interaction mechanism, considering that AGID requires multiple interactions. AGID and ELISA are techniques recommended by the WOAHA to determine prevalence of MVV infection, to verify infection prior to movement and for surveillance. On the other hand, due to its high sensitivity, the ELISA is the most suitable for use in eradication programs and for certifying herds as free of the virus. AGID, due to its high specificity, is

the most adequate to confirm clinical cases. (WOAH, 2017).

According to the Instituto Paranaense de Desenvolvimento Social [IPARDES] (2016), the mesoregions of Centro Oriental Paranaense, Centro Sul Paranaense, and Oeste Paranaense had the highest number of sheep in Paraná in 2014, with growths of 34.04%, 1.31%, and 110.87%, respectively, compared with 2004. Sheep breeding is getting increasingly technified and intensive, which improves production and standardizes the herd. This is an important measure for the success of the activity and one of the recommendations of the Support Program for the Structuring of Sheep and Goat Productive Chain (PAECPOC).

One of the main routes of transmission of the agent is contact with aerosols which can contain the virus. The intensification of production system tends to favor transmission, as it increases contact between animals (Minguijón et al., 2015). On the other hand, our study demonstrated that the variables that indicated some level of technification of the property, such as carrying out a breeding season (PR = 0.23), supplying concentrated feed (PR = 0.41), and separating the matrices before parturition (PR = 0.22), are protective factors.

Viral transmission through semen has also been reported (Gregory et al., 2011; Cortez-Romero et al., 2013), although the rate of transmission through semen is not well known, the disease causes orchitis, epididymitis and postitis (Peterson et al., 2008). In our study, the breeding season was considered a protective factor, possibly because the study area was the same as that of PAECPOC, which has as an

objective distribute breeders to producers and cooperatives, aiming at a genetic gain in the herd and the control of infectious diseases. MVV control may occur indirectly, since neither the females nor the semen are evaluated to its etiological agent.

In this study, we observed that farms that present or have already presented problems with lice have increased risks. There are no reports in the literature about such a correlation; however, lice cause itching in the animal, which in turn encourages the practice of licking among animals (Durden, 2019), exchanging salivary secretions among themselves, and promoting viral transmission.

Keeping cats close to the herd and the production system on pasture were also identified as risk factor.

Although there are no reports in the literature that associate these factors with MV, all risk factors in this study are probably associated with less technified properties, which is contrary to what has been described by other authors (Illius et al., 2020). However, this type of breeding is mainly associated with poor nutritional and sanitary management, which are predisposing factors for viral infection (Alves, 2017)

In the present study, no statistically significant differences were observed in sex, age, and breed, which was also reported by other authors (Mazzinghy et al., 2016; Larruskain & Jugo, 2013).

Conclusion

The seropositivity observed in the present study suggests the circulation

of MVV in sheep flocks in Paraná, which reinforces the need to implement the measures proposed in PNSCO, since the technification interferes with the occurrence of anti-MVV antibodies in herds.

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Authors' Contributions

All authors contributed equally for the conception and writing of the manuscript. All authors critically revised the manuscript and approved of the final version.

References

- Alves, J. R. A., Limeira, C. H., Lima, G. M. de S., Pinheiro, R. R., Alves, F. S. F., Santos, V. W. S. dos, Azevedo, S. S. de, & Alves, C. J. (2017). Caracterização epidemiológica e fatores de risco associados às lentiviroses em pequenos ruminantes comercializados em feira de animais no Sertão de Pernambuco, semiárido brasileiro. *Semina: Ciências Agrárias*, 38(4), 1875-1886. doi: 10.5433/1679-0359.2017v38n4 p1875
- Araújo, F. J., Andrioli, A., Pinheiro, R. R., Sider, L. H., Sousa, A. L. M. de, Azevedo, D. A. A., Peixoto, R. M., Lima, A. M. C., Damasceno, E. M., Souza, S. C. R., & Teixeira, M. F. S.

- (2020). Vertical transmissibility of small ruminant lentivirus. *PLOS ONE*, 15(11), e0239916. doi: 10.1371/journal.pone.0239916
- Azevedo, D. A. A. de, Pinheiro, R. R., Santos, V. W. S. dos, Damasceno, E. M., Sousa, A. L. M. de, Araújo, J. F., Andrioli, A., Sider, L. H., Peixoto, R. M., & Teixeira, M. D. S. (2019). Comparação de testes sorológicos e molecular para diagnóstico da Artrite Encefalite Caprina e avaliação clínica da glândula mamária de caprinos leiteiros infectados. *Acta Scientiae Veterinariae*, 47(1), 1668e. doi: 10.22456/1679-9216.92281
- Blacklaws, B. A. (2012). Small ruminant lentiviruses: Immunopathogenesis of visna-maedi and caprine arthritis and encephalitis virus. *Comparative Immunology, Microbiology and Infectious Diseases*, 35(3), 259-269. doi: 10.1016/j.cimid.2011.12.003
- Cortez-Romero, C., Pellerin, J. L., Ali-Al-Ahmad, M. Z., Chebloune, Y., Gallegos-Sánchez, J., Lamara, A., Pépin, M., & Fieni, F. (2013). The risk of small ruminant lentivirus (SRLV) transmission with reproductive biotechnologies: State-of-the-art review. *Theriogenology*, 79(1), 1-9. doi: 10.1016/j.theriogenology.2012.09.021
- Costa, L. S. P., Lima, P. P., Callado, A. K. C., Nascimento, S. A. do, & Castro, R. S. de. (2007). Lentivírus de pequenos ruminantes em ovinos Santa Inês: isolamento, identificação pela PCR e inquérito sorológico no estado de Pernambuco. *Arquivos do Instituto Biológico*, 74(1), 11-16. doi: 10.1590/1808-1657v74p0112007
- Czopowicz, M., Szalus-Jordanow, O., Mickiewicz, M., Moroz, A., Witkowski, L., Markowska-Daniel, I., Reczynska, D., Bagnicka, E., & Kaba, J. (2018). Decline of maternal antibodies to small ruminant lentivirus in goat kids. *Animal Science Journal*, 89(9), 1364-1370. doi: 10.1111/asj.13038
- Durden, L. A. (2019). *Medical and veterinary entomology*. In G. R. Mullen, & L. A. Durden (Eds.), *Lice (Phthiraptera)*. (3rd ed., Chap. 7, pp. 79-106). London.
- Giangaspero, M., Osawa, T., Orusa, R., Frossard, J., Naidu, B., Robetto, S., Tatami, S., Takagi, E., Moriya, H., Okura, N., Kato, K., Kimura, A., & Harasawa, R. (2011). Epidemiological survey for visna-maedi among sheep in northern prefectures of Japan. *Veterinaria Italiana*, 47(4), 437-451.
- Gomez-Lucia, E., Barquero, N., & Domenech, A. (2018). Maedi-Visna virus: Current perspectives. *Veterinary Medicine: Research and Reports*, 9(1), 11-21. doi: 10.2147/VMRR.S136705
- Gregory, L., Lara, M. C. C. S., Hasegawa, M. Y., Castro, R. S., Rodrigues, J. N. M., Araújo, J., Keller, L. W., Silva, L. K. F., & Durigon, E. L. (2011). Detecção do vírus da artrite encefalite caprina no sêmen através das técnicas de PCR e Nested-PCR. *Arquivo do Instituto Biológico*, 78(4), 599-603. doi: 10.1590/1808-1657v78p5992011
- Guilherme, R. F., Azevedo, S. S., Higino, S. S., Alves, F. S. F., Santiago, L. B., Lima, A. M. C., Pinheiro, R. R., & Alves, C. J. (2017). Caracterização epidemiológica e fatores de risco associados à infecção por lentivírus de pequenos ruminantes na

- região do semiárido paraibano, Nordeste do Brasil. *Pesquisa Veterinária Brasileira*, 37(6), 544-548. doi: 10.1590/S0100-736X2017000600002
- Illius, A. W, Lievaart-Peterson, K., McNeilly, T. N., & Savill, N. J. (2020). Epidemiology and control of maedi-visna virus: curing the flock. *PLOS ONE*, 15(9), e0238781. doi: 10.1371/journal.pone.0238781
- Instituto Brasileiro de Geografia e Estatística. (2019). *Pesquisa da pecuária municipal*. IBGE. <https://www.ibge.gov.br/estatisticas/economicas/9107-producao-da-pecuaria-municipal.html?=&t=o-que-e>
- Instituto de Terras, Cartografia e Geociências. (2006). *Clima*. ITCG. <http://www.geo.pr.gov.br/ms4/itcg/geo.html>
- Instituto Paranaense de Desenvolvimento Econômico e Social. (2016). *Base de dados do estado*. IPARDES. <http://www.ipardes.pr.gov.br/imp/index.php>
- International Committee on Taxonomy Viruses (2020). *Virus Taxonomy: 2022 Release*. <https://talk.ictvonline.org/taxonomy/>
- Junkuszew, A., Dudko, P., Bojar, W., Olech, M., Osinski, Z., Gruszecki, T. M., Kania, G. M., Kuzmak, J., & Czernski, G. (2016). Risk factors associated with small ruminant lentivirus infection in eastern Poland sheep flocks. *Preventive Veterinary Medicine*, 127(1), 44-49. doi: 10.1016/j.prevetmed.2016.03.011
- Larruskain, A., & Jugo, B. M. (2013). Retroviral infections in sheep and goats: small ruminant lentiviruses and host interaction. *Viruses*, 5(8), 2043-2061. doi: 10.3390/v5082043
- Leroux, C., Cruz, J., & Mornex, J. (2010). SRLVs: a genetic continuum of lentiviral species in sheep and goats with cumulative evidence of cross species transmission. *Current HIV Research*, 8(1), 94-100. doi: 10.2174/157016210790416415
- Lombardi, A. L., Nogueira, A. H. C., Feres, F. C., Paulo, H. P., Castro, R. S., Feitosa, F. L. F., Cadioli, F. A., Peiró, J. R., Perri, S. H. V, Lima, V. F. M., & Mendes, L. C. N. (2009). Soroprevalência de Maedi-Visna em ovinos na região de Araçatuba, SP. *Arquivo Brasileiro de Medicina Veterinária e Zootecnia*, 61(6), 1434-1437. doi: 10.1590/S0102-09352009000600025
- Martinez, P. M., Costa, J. N., Souza, T. S., Lima, C. C. V., Costa, A. de O., Neto, & Pinheiro, R. R. (2011). Prevalência sorológica da Maedi-Visna em rebanhos ovinos da microrregião de Juazeiro. *Ciência Animal Brasileira*, 12(2), 322-329. doi: 10.5216/cab.v12i2.4454
- Mazzinghy, C. L., Almeida, K. S., Veschi, J. L. A., Castro, R. S., Martins, N. É. X., & Sousa, M. G. (2016). Frequência de ovinos soropositivos para lentivírus de pequenos ruminantes no município de Colinas do Tocantins, estado do Tocantins, Brasil. *Arquivos do Instituto Biológico*, 83(1), 1-5. doi: 10.1590/1808-1657000542014
- Mcvey, D. S., Kennedy, M., & Chengappa, M. M. (2016). *Microbiologia veterinária*. Guanabara Koogan.
- Michiels, R., Mael, E. V., Quinet, C., Welby, S., Cay, A. N., & Regge, N. D (2018). Seroprevalence and risk factors related to small ruminant lentivirus infections in Belgian sheep and goats. *Preventive Veterinary Medicine*, 151(1), 13-20. doi: 10.1016/j.prevetmed.2017.12.014

- Minguijón, E., Reina, R., Pérez, M., Polledo, L., Villoria, M., Ramírez, H., Leginagoikoa, I., Badiola, J. J., García-Marín, J. F., de Andrés, D., Luján, L., Amorena, B., & Juste, R. A. (2015). Small ruminant lentivirus infections and diseases. *Veterinary Microbiology*, 181(1-2), 75-89. doi: 10.1016/j.vetmic.2015.08.007
- Ministério da Agricultura Pecuária e Abastecimento. (2017). *Programa Nacional de Sanidade de Caprinos e Ovinos*. MAPA. <https://www.gov.br/agricultura/pt-br/assuntos/sanidade-animal-e-vegetal/saude-animal/programas-de-saude-animal/sanidade-de-caprinos-e-ovinos>
- Nascimento, C. B., Pinheiro, R. R., Alves, F. S. F., Brito, R. L. L. D., Rodrigues, A. D. S., Bezerra e Silva, R. A., Paula, O. R. N., & Batista, M. D. C. D. S. (2014). Ferramentas diagnósticas de Lentivirose de pequenos ruminantes: padronização da técnica de ELISA indireto. *Arquivos do Instituto Biológico*, 81(1), 9-15. doi: 10.1590/S1808-16572014000100003
- Perez, M., Biescas, E., Andres, X. de, Leginagoikoa, I., Salazar, E., Berriatua, E., Reina, R., Bolea, R., de Andrés, D., Juste, R. A., Cancer, J., Gracia, J., Amorena, B., Badiola, J. J., & Lujan, L. (2010). Visna/maedi virus serology in sheep: survey, risk factors and implementation of a successful control programme in Aragon (Spain). *Veterinary Journal*, 186(2), 221-225. doi: 10.1016/j.tvjl.2009.07.031
- Peterson, K., Brinkhof, J., Houwers, D. J., Colenbrander, B., & Gadella, B. M. (2008). Presence of pro-lentiviral DNA in male sexual organs and ejaculates of small ruminants. *Theriogenology*, 69(4), 433-442. doi: 10.1016/j.theriogenology.2007.10.013
- Pinheiro, D. N. S., Costa, J. N., Souza, T. S., Santos, V. W. S., Azevedo, D. A., Costa, A. O., Neto, & Pinheiro, R. R. (2018). Serum epidemiological survey and risk factors investigation for lentivirus in goats from Sisal Region, Bahia, Brazil. *Arquivos do Instituto Biológico*, 85(1), 1-6. doi: 10.1590/1808-1657000812016
- Ramírez, H., Reina, R., Bertolotti, L., Cenoz, A., Hernández, M., Román, B. S., Glaria, I., de Andrés, X., Crespo, H., Jáuregui, P., Benavides, J., Polledo, L., Pérez, V., García-Marín, J. F., Rosati, S., Amorena, B., & de Andrés, A. (2012). Study of compartmentalization in the visna clinical form of small ruminant lentivirus infection in sheep. *BMC Veterinary Research*, 8(8), 1-12. doi: 10.1186/1746-6148-8-8
- Ravazzolo, A. P., Reischak, D., Peterhans, E., & Zanoni, R. (2001). Phylogenetic analysis of small animal lentiviruses from southern Brazil. *Virus Research*, 79(1-2), 117-123. doi: 10.1016/s0168-1702(01)00339-2
- Reina, R., Berriatua, E., Lujan, L., Juste, R., Sanchez, A., Andres, D. de, & Amorena, B. (2009). Prevention strategies against small ruminant lentiviruses: an update. *The Veterinary Journal*, 182(1), 31-37. doi: 10.1016/j.tvjl.2008.05.008
- Secretaria da Agricultura de do Abastecimento do Paraná (2008). *Ovino e caprino - programa estadual de apoio à estruturação das cadeias produtivas*. SEAB. <http://www.ovinocaprino.pr.gov.br/>

- Souza, K. C., Pinheiro, R. R., Santos, D. O., Brito, R. L. L., Rodrigues, A. S., Sider, L. H., Paula, N. R. O., Avila, A. A., Cardoso, J. F. S., & Andrioli, A. (2013). Transmission of the caprine arthritis-encephalitis virus through artificial insemination. *Small Ruminant Research*, 109(2-3), 193-198. doi: 10.1016/j.smallrumres.2012.07.031
- Tu, P. A., Shiu, J. S., Lee, S. H., Pang, V. F., Wang, D. C., & Wang, P. H. (2017). Development of a recombinase polymerase amplification lateral flow dipstick (RPA-LFD) for the field diagnosis of caprine arthritis-encephalitis virus (CAEV) infection. *Journal of Virological Methods*, 243(1), 98-104. doi: 10.1016/j.jviromet.2017.01.023
- Valas, S., Benoit, C., Guionaud, C., Perrin, G., & Mamoun, R. Z. (1997). North american and french caprine arthritis-encephalitis viruses emerge from ovine maedi-visna viruses. *Virology*, 237(2), 307-318. doi: 10.1006/viro.1997.8800
- World Organization for Animal Health (2017). *Manual of diagnostic tests and vaccines for terrestrial animals*. WOA. https://www.woah.org/fileadmin/Home/eng/Health_standards/tahm/3.08.02_CAE_MV.pdf
- World Organization for Animal Health (2022). *Diseases, Infections and Infestations Listed by the OIE*. WOA. https://www.woah.org/en/what-we-do/standards/codes-and-manuals/terrestrial-code-online-access/?id=169&L=1&htmlfile=chapidre_oie_listed_disease.htm