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Leptospirosis, bovine viral diarrhea and infectious bovine rhinotracheitis: prevalence in Colombian cattle and buffaloes

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ABSTRACT. One of the limiting factors of productive efficiency in cattle and buffalo herds is related to the high prevalence of infectious diseases which affect reproduction. The aim of this study was to determine the prevalence of anti-*Leptospira* antibodies for bovine viral diarrhea virus (BVDV) and bovine herpesvirus type 1 (BoHV-1) in bovine and buffalo herds in Colombia. Blood serum samples were collected from 1100 buffaloes and 1000 cattle. The ELISA technique was used to detect antibodies against BVDV and BoHV-1, and the microscopic agglutination technique to detect anti-*Leptospira* antibodies. The prevalence of anti-*Leptospira* antibodies and of BVDV and BoHV-1 in bovine samples was observed in 16, 39.7, and 65% of animals, respectively, while the positivity in samples for the same antibodies in buffalos was detected in 18.7, 27.5 and 51.5%, respectively. Exposure of cattle and buffaloes to BoHV-1 was positively associated with age, higher prevalence rates were observed in older ages. Seropositivity of cattle for BVDV and BoHV-1 was higher in male animals. Activities such as embryo transfer, milking, and needle reuses, as well as the presence of cats and rodents are factors which favor positivity of the herd for BVDV and BoHV-1.

Keywords: *Bovidae*; Colombia; reproductive infectious diseases; risk factors; serology.

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Introduction

Colombia's territory has an extension of 114 million hectares, from which approximately 39.2 million are used for livestock production (Jiménez, Miranda, Gantiva, & Escuela Colombiana de Ingeniería, 2008). The growth in the cattle production in Colombia has shown a slow and sustained pace with an average annual rate of 0.02% (Cervantes, Espitia, & Prieto, 2010; Mahecha, Gallego, & Peláez, 2016).

One of the limiting aspects in the productive efficiency in cattle and buffalo herds is the high prevalence of infectious reproduction diseases (Alfieri & Alfieri, 2017). The most prevalent pathogens with reproductive impact on ruminants include *Leptospira*, Bovine Viral Diarrhea Virus (BVDV) and Bovine Herpesvirus Type 1 (BoHV-1) (Buitrago Horta, Jimenez Escobar, & Zambrano Varon, 2018; Rivera, Rincón, & Echaverry, 2018; Vargas-Niño et al., 2018).

Leptospirosis is a zoonotic disease caused by urine-released bacteria which can infect large ruminants through their skin with or without abrasions, or through the conjunctival, oral, nasopharyngeal, and genital mucosae (Pulido-Medellín, Díaz-Anaya, & Giraldo-Forero, 2017). They can colonize the renal tubules, mammary glands and the reproductive tract of animals, leading to infertility and miscarriages (Lilenbaum & Martins, 2014). The optimal temperature for leptospira growth is 28-30°C, which may facilitate their spreading in herds of regions with tropical weather (Levett, 2001).

The etiological agent of Bovine Viral Diarrhea (BVD) is a pestivirus of the Flaviviridae family that is transmitted through direct contact with an infected animal (Lanyon, Hill, Reichel, & Brownlie, 2014). Females exposed to the virus during pregnancy may infect the fetus in the womb, resulting in reproductive loss, stillbirth, fetal malformations, embryonic absorption or birth of persistently infected calves (PI), which are the main linkage in the BVD epidemiological chain (Grooms, 2004).

Infectious Bovine Rhinotracheitis (IBR) is caused by bovine herpesvirus type 1, with the main biological characteristic of establishing latent infections (Alfieri & Alfieri, 2017). This pathogen can infect animals

through direct or indirect contact, and is the major agent involved in the respiratory complex of young cattle (Slompo et al., 2017). In addition to respiratory disease, BoHV-1 infection is associated with reproductive disease, leading to reproductive failures, which represent the most significant losses associated with the presence of this pathogen (Viu, Dias, Lopes, Viu, & Ferraz, 2014).

These diseases are not subject to official control programs in Colombia, thus hindering access to economic resources to implement reduction and eradication programs. The objective of this study was to determine the prevalence of pathogens causing Leptospirosis, BVD and IBR on farms in Colombia, and to identify risk factors associated with their presence.

Material and methods

The study was conducted in the Colombian states of Cordoba, Antioquia and Sucre (Table 1). Cattle (n = 1000) and buffalo (n = 1100) blood serum samples were collected. Simple-random sampling was performed, which was calculated through WinEpiscope® 2.0 epidemiological software for veterinary medicine and considering the population size based on the Colombian national livestock census of 2017 (Instituto Colombiano Agropecuario [ICA], 2017). Farms involved in the study did not adopt vaccination programs against the assessed pathogens. This study was approved by the Institutional Committee for the Care and Use of Animals of the Corporation for Health Studies (CES), under protocol number 162 of November 17th, 2017.

Blood was collected by puncturing the coccygeal vein using a closed system collection (Vacutainer®). Samples were centrifuged at room temperature for 15 min. at 2000 g and the isolate blood serum were transferred to 1.5 mL transport tubes and stored in a freezer at -20°C until further processing. An epidemiological questionnaire was applied to the owners and/or managers of the 59 selected farms with questions about the type of production system, management practices and nutritional supplementation to assess the risk factors.

Next, commercially available ELISA kits were used following the manufacturer's instructions (INgezim PESTIVIRUS Compac p80/p125 Antibody and INgezim IBR Compac Total Antibody - INGENASA©) to detect antibodies against BVDV and BoHV-1. The microscopic agglutination test (MAT) was performed using a culture of live antigens in Ellinghausen-McCullough-Johnson-Harris (EMJH) culture medium to diagnose serological variants of *Leptospira*: Autumnalis, Bataviae, Bratislava, Canicola, Celledoni, Cynoptery, Copennhageni, Grippotyphosa, Hardjo, Mini, Pomona, Shermani and Tarassovi. Titers were determined according to the standard criteria as the highest serum dilution which agglutinated at least 50% of the cells for each serotype tested (Blanco, Santos, Galloway, & Romero, 2016).

Data were analyzed using SAS University Edition® statistical software (2019). The seroprevalence of each disease was calculated by the chi-squared test. The association of seropositive animals with management issues and presence of other animal species was analyzed by chi-squared (X^2) and logistic regression tests. Values of $p < 0.05$ and $OR > 1$ were considered statistically significant.

Table 1. Colombian locations used for collecting blood serum samples from cattle and buffaloes for serological analyses for the presence of antibodies against BVDV, BoHV-1 and *Leptospira* spp.

State	Municipality	Specie	No. of samples	No. of farms
Córdoba	Montería	Cattle	1000	8
Córdoba	Ayapel	Buffalo	281	11
Córdoba	Montelíbano	Buffalo	157	7
Córdoba	Buenavista	Buffalo	86	5
Córdoba	Planeta Rica	Buffalo	30	1
Córdoba	Pueblo Nuevo	Buffalo	27	1
Córdoba	Tierralta	Buffalo	25	1
Córdoba	Montería	Buffalo	25	1
Córdoba	San José de Uré	Buffalo	25	1
Córdoba	La Apartada	Buffalo	10	1
Antioquia	Caucasia	Buffalo	205	12
Antioquia	Cáceres	Buffalo	99	5
Antioquia	Nechí	Buffalo	30	1
Antioquia	Tarazá	Buffalo	30	1
Sucre	San Marcos	Buffalo	40	2
Sucre	San Benito Abad	Buffalo	30	1
Total			2100	59

Data were analyzed using SAS University Edition® statistical software (2019). The seroprevalence of each disease was calculated by the chi-squared test. The association of seropositive animals with management issues and presence of other animal species was analyzed by chi-squared (X^2) and logistic regression tests. Values of $p < 0.05$ and $OR > 1$ were considered statistically significant.

Results and discussion

Seroprevalence of Leptospirosis was observed in 160 cows (16%) and 206 buffaloes (18.7%). In turn, antibodies against BVDV were found in 397 cows (39.7%) and 302 buffaloes (27.5%). For BoHV-1, 650 (65%) positive cattle reagents were detected, while 565 samples (51.5%) from buffaloes were positive. Significant differences between the two species were observed for BVD and IBR prevalence ($p < 0.001$) (Table 2).

Table 2. Prevalence of antibodies against Leptospira, BVDV and BoHV-1 in cattle and buffaloes from Colombia.

Specie	Pathogen					
	BVD		IBR		Leptospirosis	
	Positive	%	Positive	%	Positive	%
Cattle	397	39.7[a]	650	65[a]	160	16
Buffalo	302	27.4[b]	565	51.3[b]	206	18.7

Different letters in each column indicate statistically significant differences ($p < 0.05$).

The BVDV seroprevalence rate in cattle agrees with that reported by Vargas, Jaime, and Vera (2009), who ratified the virus as endemic in most Colombian bovine herds with rates between 40 and 80%. In addition, the present study confirmed the presence of BVDV antibodies in buffaloes from Colombian herds, unlike the results of Giraldo, Hoyos, García, and Abelado (2014), who did not detect seropositive buffaloes for this pathogen in Colombia.

The increase in BVDV antibody titers had a potential association ($p < 0.001$) with the increased chance of those animals being seropositive for BoHV-1. This association was identified in 272 cows (27.2%) and 174 buffaloes (15.8%). The finding suggests that animals transiently infected with BVDV could present immunosuppression due to the virus' affinity for defense cells for their viral replication, thus generating lymphoid depletion and neutropenia and increasing the susceptibility of the host to infection by other pathogens which affect the reproductive system (Headley et al., 2014; Lanyon et al., 2014).

Seroprevalence of BoHV-1 was also higher in bovine animals compared to buffaloes. Studies carried out in Colombia have confirmed the presence of the virus in cattle from different states, showing the enzootic behavior of the disease due to the latent characteristic of the pathogen in the infected animal which could be naturally reactivated in stress situations throughout its life (Alfieri & Alfieri, 2017; Ruiz Sáenz, Jaime, & Vera, 2010; Vargas-Niño et al., 2018).

The seroprevalence in buffaloes was lower compared to the previously reported 80.3% in Monteria state (Giraldo et al., 2014), however this result confirms the susceptibility of buffalo to BoHV-1 infection and therefore its role as host/reservoir of the virus (Scicluna et al., 2010).

A positive correlation was observed in both species between the age of the animals and the seropositivity to BoHV-1 infection ($p < 0.001$). In cattle, animals younger than one year old had a prevalence of 38.7% (24/62), and animals between 1 and 2 years had a prevalence of 27% (65/241), while the prevalence rates in the age groups of 2 to 3 and over 3 years old were 81.5% (527/646) and 66.6% (34/51), respectively. Buffaloes younger than one year had a prevalence of 43.4% (86/198) and of 23% (56/243) for between one and two years of age. Furthermore, the prevalence in the age group between 2 and 3 years was 51% (74/145), while seropositivity in animals older than 3 years was higher, with a final prevalence of 68.4% (349/510).

A higher prevalence in adult than in young animals has been reported (Aragaw et al., 2018; Erfani, Bakhshesh, Fallah, & Hashemi, 2019). Buffaloes and cattle between 2 and 3 years old represent the most active population of the herd from the productive and reproductive points of view. This characteristic allows greater possibility for natural infection due to the greater chances of sexual contact (Eiras, Diéguez, Sanjuan, Yus, & Arnaiz, 2009). Additionally, higher stress conditions as a consequence of production requirements provide immunosuppression conditions which are crucial for viral reactivation (Piedrahita, Montoya, & Pedraza, 2010).

Regarding sex, male cattle showed higher seropositivity for BVDV (41.8%) and BoHV-1 (66.5%), suggesting that infected bulls represent an important source of venereal transmission. The use of antimicrobials in the semen freezing process for artificial insemination does not affect the potential for infection of these viruses

and could therefore remain active for their transmission. Other practices such as natural breeding with sires with latent BoHV-1 represent a major problem because the semen collected during episodes of viral reactivation could suffer contamination (Viu et al., 2014).

The occurrence of leptospirosis in tropical areas of South America has endemic characteristics for the disease (Brasil et al., 2015). In this study, the results obtained regarding the presence of anti-*Leptospira* spp. antibodies showed positivity of 18.7% for buffalo and 16% for cattle. Higher results were described in another study conducted in Colombia, in which researchers reported 37.3% and 28% seroprevalence in buffalo and cattle, respectively, with a predominance of Gryppotyphosa serotype (Giraldo et al., 2014).

The prevalence in this study was higher in buffalo, possibly due to the extensive management commonly practiced in Colombian herds, which allows the animals to have free access to lakes, wetlands and ciliary forests, along with the habit of buffaloes staying protected in flooded areas during the hottest hours of the day, thus facilitating contact with water which has been previously contaminated with urine from rats, other wildlife animals, or even other infected buffaloes (Calderón, Rodrígues, Máttar, & Arrieta, 2013; Giraldo et al., 2014; Nagel et al., 2019).

The highest percentage of reactive animals in the two species included in this study was for the Hardjo serovar (7.1% and 3.8% for cattle and buffalo, respectively). The presence of Canicola (5.7%), Copenhageni (3.7%) and Pomona (0.2%) serovars was verified in cattle, while buffaloes were reactive for 11 of the 13 serovars analyzed, with seropositivity rates of between 0.09 and 3.8% (Table 3).

Table 3. Serovars of *Leptospira* spp. in blood serum samples from cattle and buffalo with positive reaction to the MAT technique.

Specie	Sorovar	Cattle	%	Buffaloes	%
L. interrogans	Autumnalis	0	0%	37	3.4%
	Bataviae	0	0%	5	0.4%
	Bratislava	0	0%	31	2.8%
	Copenhageni	37	3.7%	1	0.1%
	Canicola	57	5.7%	35	3.2%
	Hardjo	71	7.1%	42	3.8%
	Pomona	2	0.2%	31	2.8%
L. kirshneri	Gryppotyphosa	0	0%	28	2.5%
	Cynopteri	0	0%	2	0.2%
L. borgpetersenii	Mini	0	0%	1	0.1%
	Tarassovi	0	0%	0	0%
L. santarosai	Shermani	0	0%	1	0.1%
L. Weillii	Celledoni	0	0%	0	0%

Multivariate logistic regression analysis for BVDV and BoHV-1 pathogens in cattle and buffaloes is presented in Table 4. The presence of antibodies to BoHV-1 in both species and BVDV in cattle was significantly associated with the practice of not using individual needles per animal. These observations confirm the importance of transmitting pathogens by direct pathways and the epidemiological role played by materials, vehicles or contaminated equipment as potential sources of transmission (Han, Weir, Weston, Heur, & Gates, 2018).

Table 4. Risk factors associated with the presence of BVDV and BoHV-1 in cattle and buffalo from Colombia analyzed by logistic regression.

Specie	Predictors	Odds Ratio	95% CI	P-*value*
	BVDV			
Cattle	Reuse of needles	3.07	1.20 - 7.85	0.01
	Presence of rodents	2.04	1.36 - 3.05	0.0005
	Clinical history of diarrhea	2.42	1.50 - 3.93	0.0003
Buffalo	Use of embryo transfer technique	6.76	2.74 - 16.68	<0.001
	Presence of cats	3.04	1.4 – 6.2	0.0026
	Milking	1.87	1.22 – 2.85	0.0038
	BoHV-1			
Cattle	Presence of rodents	1.92	1.01 - 3.62	0.04
	Reuse of needles	4.03	1.20 - 13.55	0.02
	Reuse of needles	1.40	1.02 – 1.92	0.03
Buffalo	Clinical history of diarrhea	2.06	1.40 – 3.02	0.0002
	History of breathing problems	1.5	1.05 – 2.33	0.02

The clinical history of diarrhea in buffaloes was correlated with positivity for BVDV and BoHV-1. In addition to the wide variety of respiratory and reproductive clinical manifestations, these pathogens can generate diarrhea during the acute stage of the disease, which predisposes the animals to debilitating secondary infections (Takiuchi, Alfieri, & Alfieri, 2001). Diarrhea may additionally represent an important source of environmental contamination for this pathogen, as the presence of active viral particles in feces from BoHV-1-positive buffaloes has been proven (Scicluna et al., 2010).

This study also correlated the history of respiratory problems with the presence of BoHV-1 in buffaloes ($p < 0.05$). The virus can trigger changes associated with breathing problems with symptoms such as coughing and nasal discharge which are a source of direct contamination for other animals. Along with other viral and bacterial pathogens, the BoHV-1 is also recognized for its contribution to developing Bovine Respiratory Disease (BRD) due to the adverse effects they produce on the respiratory tissues (Moore, O'Dea, Perkins, & O'Hara, 2015).

The presence of rodents was associated with BoHV-1 infection in cattle and BVDV in buffaloes ($p < 0.05$). Rahpaya et al. (2018) first identified BVDV in rodent fecal matter, revealing the importance of this species as a vector and reservoir of the pathogen for other productive species. The existence of a new viral species of the *pestivirus* genus was also recently proposed, denominated the *rat pestivirus*, which shares common genomic organization and protein homology with other species identified in ruminants such as BVDV-1 and -2 and the etiological agent of the border disease. However, the hypothesis of a probable association of the diseases in relation to the virus sequence, as well as identification of hosts and their consequence in animals need further confirmation (Smith et al., 2017).

The presence of cats was identified as a significant risk factor for BVDV in buffalo ($p < 0.0026$). Cats could be involved in spreading the disease due to their high intensity of contact with other infected animals, dynamically participating in distributing the pathogen in the herd (Nelson, Duprau, Wolff, & Evermann, 2016). This observed association could be indirect, since farm cats are used as a biological control method for rodents.

The implementation of biotechnological techniques may be a source of transmission for BVDV. It was observed that buffaloes are more likely to be seropositive for BVDV by using the embryo transfer technique ($p < 0.0001$). Previous studies suggest that embryos produced *in vitro* from oocytes infected with BVDV-1 and -2 can develop normally and maintain latent virus until their transfer to recipient animals (Pinto et al., 2017). The use of potentially contaminated biological materials, such as fetal bovine serum used during embryo production and transfer practices should be analyzed to determine the true association between seropositivity and the application of this biotechnological technique (Dezengrini, Weiblen, & Flores, 2006).

This study identified milking practice as a factor associated with positive BVDV status in buffaloes ($p < 0.0038$). This finding was also verified in dairy products from Switzerland, having a higher risk of infection in relation to beef or mixed production systems, probably being related to higher contact with people and the increased traffic on dairy farms (inseminator, equipment, processing facilities, milk collection trucks, food suppliers, veterinarians, etc) (Presi, Struchen, Knight-Jones, Scholl, & Heim, 2011).

A study conducted in Italy founded that BVDV predominates on dairy farms due to the implemented productive system mainly being based on the internal breeding of its own offspring, thus representing an ideal habitat for persistent and self-perpetuating virus infection (Iotti et al., 2019).

There were no identified risk factors significantly associated to the presence of leptospirosis, although it has been previously reported that its prevalence could be associated to diverse factors such as contact with other animal species, management, presence of different existing serotypes in the same region, as well as climatic and environmental conditions (Higino et al., 2013).

Conclusion

In conclusion, the three pathogens included in this study are widespread in cattle and buffalo herds from Colombia. The highest prevalence in both species was for BoHV-1 (65% and 51.5%), followed by BVDV (39.7% and 27.5%) and *Leptospira* spp. (16% and 18.7%), with there also being a higher likelihood of infection by BoHV-1 when animals were further infected with BVDV.

The presence of antibodies against BVDV in buffaloes from Colombia is reported herein for the first time, as well as the risk factors associated to the presence of BVDV, BoHV-1 and leptospirosis in Colombian buffalo herds, thus demonstrating the susceptibility of this species and their role in the epidemiological dynamic of these diseases.

Adult animals were the most affected by the BoHV-1 and BVDV viruses. The risk factor analysis determined that routine management activities such as embryo transfer, milking, and reusing needles are practices which favor spreading reproductive diseases; in addition, the presence of cats and rodents was closely related to BVDV and BoHV-1 positivity in cattle and buffaloes.

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References

Alfieri, A. A., & Alfieri, A. F. (2017). Doenças infecciosas que impactam a reprodução de bovinos. *Revista Brasileira de Reprodução Animal, 41*(1), 133–139.

Aragaw, K., Sibhat, B., Ayelet, G., Skjerve, E., Gebremedhin, E. Z., & Asmare, K. (2018). Seroprevalence and factors associated with bovine viral diarrhea virus (BVDV) infection in dairy cattle in three milksheds in Ethiopia. *Tropical Animal Health and Production, 50*(8), 1821–1827. DOI: http://doi.org/10.1007/s11250-018-1624-5

Blanco, R. M., Santos, L. F. d., Galloway, R. L., & Romero, E. C. (2016). Is the microagglutination test (MAT) good for predicting the infecting serogroup for leptospirosis in Brazil?. *Comparative Immunology, Microbiology and Infectious Diseases, 44*, 34–36. DOI: http://doi.org/10.1016/j.cimid.2015.12.003

Brasil, A. W. d. L., Parentoni, R. N., Costa, D. F., Pimenta, C. L. R. M., Araújo, R. P., Leal, A. B. G., ... Azevedo, S. S. d. (2015). Occurrence of anti-*Brucella abortus* and anti-*Leptospira* spp. antibodies in buffaloes from Paraíba state, Northeastern Brazil. *Semina:Ciencias Agrarias, 36*(3), 2005–2012. DOI: http://doi.org/10.5433/1679-0359.2015v36n3Supl1p2005

Buitrago Horta, E. R., Jimenez Escobar, C., & Zambrano Varon, J. L. (2018). Identificación de factores asociados con la exposición al virus de ladiarrea viral bovina (VDVB) en terneras de hatos lecheros de la sabana de Bogotá. *Revista de Medicina Veterinaria, 36*, 63–73. DOI: http://dx.doi.org/10.19052/mv.5172

Calderón, A., Rodrígues, V., Máttar, S., & Arrieta, G. (2013). Leptospirosis in pigs, dogs, rodents, humans, and water in an area of the Colombian tropics. *Tropical Animal Health and Production, 46*(2), 427–432. DOI: http://doi.org/10.1007/s11250-013-0508-y

Cervantes, A. E., Espitia, P. A., & Prieto, M. E. (2010). Viabilidad de los sistemas bufalinos en Colombia. *Revista Colombiana de Ciencia Animal - RECIA, 2*(1), 215-224. DOI: 10.24188/recia.v2.n1.2010.342

Jiménez, N. J., Miranda, F. C., Gantiva, O. H. D., & Escuela Colombiana de Ingeniería. (2008). El sector de ganadería bovina en Colombia. Aplicación de modelos de series de tiempo al inventario ganadero. *Revista de la Facultad de Ciencias Económicas, 16*(1), 165–177.

Dezengrini, R., Weiblen, R., & Flores, E. F. (2006). Selection and characterization of canine, swine and rabbit cell lines resistant to bovine viral diarrhea virus. *Journal of Virological Methods, 137*(1), 51–57. DOI: http://doi.org/10.1016/j.jviromet.2006.05.032

Eiras, C., Diéguez, F. J., Sanjuan, M. L., Yus, E., & Arnaiz, I. (2009). Prevalence of serum antibodies to bovine herpesvirus-1 in cattle in Galicia (NW Spain). *Spanish Journal of Agricultural Research, 7*(4), 800-806. DOI: http://doi.org/10.5424/sjar/2009074-1094

Erfani, A. M., Bakhshesh, M., Fallah, M. H., & Hashemi, M. (2019). Seroprevalence and risk factors associated with bovine viral diarrhea virus and bovine herpes virus-1 in Zanjan Province, Iran. *Tropical Animal Health and Production, 51*(2), 313–319. DOI: http://doi.org/10.1007/s11250-018-1687-3

Giraldo, J. L. M., Hoyos, J. A. C., García, I. W., & Abelado, M. A. (2014). Prevalencia de anticuerpos a *Brucella abortus*, *Leptospira* sp. y *Neospora* caninum en hatos bovinos y bubalinos en el Departamento de Caquetá, Colombia. *Revista de Salud Animal, 36*(2), 80–89.

Grooms, D. L. (2004). Reproductive consequences of infection with bovine viral diarrhea virus. *The Veterinary Clinics of North America: Food Animal Practice, 20*(1), 5–19. DOI: http://doi.org/10.1016/j.cvfa.2003.11.006

Han, J.-H., Weir, A. M., Weston, J. F., Heur, C., & Gates, M. C. (2018). Elimination of bovine viral diarrhoea virus in New Zealand : a review of research progress and future directions. *New Zealand Veterinary Journal, 66*(6), 273-280. DOI: http://doi.org/10.1080/00480169.2018.1509030

Headley, S. A., Alfieri, A. A., Fritzen, J. T. T., Queiroz, G. R., Lisbôa, J. A. N., Pontes Netto, D., ... Alfieri, A. F. (2014). Concomitant bovine viral diarrhea, mycotoxicosis, and seneciosis in beef cattle from northern Paraná, Brazil. *Semina: Ciências Agrárias, 35*(5), 2563-2576. DOI: http://doi.org/10.5433/1679-0359.2014v35n5p2563

Higino, S. S. S., Santos, F. A., Costa, D. F., Santos, C. S. A. B., Silva, M. L. C. R., Alves, C. J., & Azevedo, S. S. (2013). Flock-level risk factors associated with leptospirosis in dairy goats in a semiarid region of Northeastern Brazil. *Preventive Veterinary Medicine, 109*(1-2), 158–161. DOI: http://doi.org/10.1016/j.prevetmed.2012.09.005

Instituto Colombiano Agropecuario [ICA]. (2017). *Censo Pecuario Nacional-2017*. Retrieved on September, 23, 2019 from lt;www.ica.gov.co/areas/pecuaria/servicios/epidemiologia- veterinaria/censos-2016/censo-2017.aspx&gt

Iotti, B., Valdano, E., Savini, L., Candeloro, L., Giovannini, A., Rosati, S., ... Giacobini, M. (2019). Farm productive contexts and the dynamics of bovine viral diarrhea (BVD) transmission. *Preventive Veterinary Medicine, 165*, 23–33. DOI: http://doi.org/10.1016/j.prevetmed.2019.02.001

Lanyon, S. R., Hill, F. I., Reichel, M. P., & Brownlie, J. (2014). Bovine viral diarrhoea: pathogenesis and diagnosis. *Veterinary jornal, 199*(2), 201–209. DOI: http://doi.org/10.1016/j.tvjl.2013.07.024

Levett, P. N. (2001). Leptospirosis. *Clinical Microbiology Reviews, 14*(2), 296–326. DOI: http://doi.org/10.1128/CMR.14.2.296-326.2001

Lilenbaum, W., & Martins, G. (2014). Leptospirosis in cattle: a challenging scenario for the understanding of the epidemiology. *Transboundary and Emerging Diseases, 61*(1), 63-68. DOI: http://doi.org/10.1111/tbed.12233

Mahecha, L., Gallego, L. A., Peláez, F. J. (2016). Situación actual de la ganadería de carne en Colombia y alternativas para impulsar su competitividad y sostenibilidad. *Revista Colombiana de Ciencias Pecuarias, 15*(2), 213–225.

Moore, S. J., O'Dea, M. A., Perkins, N., & O'Hara, A. J. (2015). Estimation of nasal shedding and seroprevalence of organisms known to be associated with bovine respiratory disease in Australian live export cattle. *Journal of Veterinary Diagnostic Investigation, 27*(1), 6 –17. DOI: http://doi.org/10.1177/1040638714559741

Nagel, A., Vázquez, C. L., Etulain, J., Blanco, F. C., Gravisaco, M. J., Gómez, R. M., & Caimi, K. (2019). Bovine macrophages responses to the infection with virulent and attenuated Leptospira interrogans serovar Pomona. *Veterinary Microbiology, 233*, 124–132. DOI: http://doi.org/10.1016/j.vetmic.2019.04.033

Nelson, D. D., Duprau, J. L., Wolff, P. L., & Evermann, J. F. (2016). Persistent bovine viral diarrhea virus infection in domestic and wild small ruminants and camelids including the mountain goat (*Oreamnos americanus*). *Frontiers in Microbiology, 6*, 1415. DOI: http://doi.org/10.3389/fmicb.2015.01415

Viu, M. A. d. O., Dias, L. R. O., Lopes, D. T., Viu, A. F. M., & Ferraz, H. T. (2014). Rinotraqueíte infecciosa bovina: revisão. *Pubvet, 8*(4), 1-21.

Piedrahita, L. E., Montoya, L. M., & Pedraza, F. J. (2010). Herpes Virus Bovino tipo 1 (BoHV-1) como posible causa de encefalitis en bovinos de la región del Magdalena Medio Colombiano. Estudio serológico y análisis epidemiológico. *Revista Colombiana de Ciências Pecuarias, 23*(2), 191–198.

Pinto, V. d. S. C., Alves, M. F., Martins, M. d. S. N., Basso, A. C., Tannura, J. H., Pontes, J. H. F., ... Pituco, E. M. (2017). Effects of oocytes exposure to bovine diarrhea viruses BVDV-1, BVDV-2 and Hobi-like virus on *in vitro*-produced bovine embryo development and viral infection. *Theriogenology, 97*, 67–72. DOI: http://doi.org/10.1016/j.theriogenology.2017.04.028

Presi, P., Struchen, R., Knight-Jones, T, Scholl, S., & Heim, D. (2011). Bovine viral diarrhea (BVD) eradication in Switzerland - experiences of the first two years. *Preventive Veterinary Medicine, 99*(2–4), 112–121. DOI: http://doi.org/10.1016/j.prevetmed.2011.01.012

Pulido-Medellín, M., Díaz-Anaya, A., & Giraldo-Forero, J. (2017). Determinación de *Leptospira* spp. en humanos y bovinos pertenecientes al municipio de Toca, Boyacá. *Veterinaria y Zootecnia, 11*(2), 55–66. DOI: http://doi.org/10.17151/vetzo.2017.11.2.5

Rahpaya, S. S., Tsuchiaka. S., Kishimoto, M., Oba, M., Katayama, Y., Nunomura, Y., ... Mizutani, T. (2018). Dembo polymerase chain reaction technique for detection of bovine abortion, diarrhea, and respiratory disease complex infectious agents in potential vectors and reservoirs. *Journal of Veterinary Science, 19*(3), 350–357. DOI: http://doi.org/10.4142/jvs.2018.19.3.350

Rivera, D. C., Rincón, J. C., & Echaverry, J. C. (2018). Prevalencia de algunas enfermedades infecciosas en bovinos de resguardos indígenas del Cauca, Colombia, 2017. *Revista U.D.C.A. Actualidad & Divulgación Científica, 21*(2), 507–517. DOI: http://doi.org/10.31910/rudca.v21.n2.2018.983

Ruiz-Sáenz, J., Jaime, J., & Vera, V. J. (2010). Prevalencia serológica y aislamiento del Herpesvirus Bovino-1 (BHV-1) em hatos ganaderos de Antioquia y del Valle del Cauca. *Revista Colombiana de Ciencias Pecuarias, 23*(3), 299–307.

SAS Institute Inc. (2019). *SAS University Edition® - statistical software*. Cary, NC: SAS Institute Inc.

Scicluna, M. T., Caprioli, A., Saralli, G., Manna, G., Barone, A., Cersini, A., ... Autorino, G. L. (2010). Should the domestic buffalo (*Bubalus bubalis*) be considered in the epidemiology of Bovine Herpesvirus 1 infection?. *Veterinary Microbiology, 143*(1), 81–88. DOI: http://doi.org/10.1016/j.vetmic.2010.02.016

Slompo, D., Bertagnon, H. G., Horst, E. H., Neumann, M., Mareze, J., Souza, A. M. d., ... Askel, E. J. (2017). Manejo do complexo respiratório bovino em confinamento: revisão. *Pubvet, 11*(4), 313-423. DOI: http://dx.doi.org/10.22256/pubvet.v11n4.381-392

Smith, D. B., Meyers, G., Bukh, J., Gould, E. A., Monath, T., Muerhoff, A. S., ... Becher, P. (2017). Proposed revision to the taxonomy of the genus *Pestivirus*, family Flaviviridae. *The Journal of General Virology, 98*(8), 2106–2112. DOI: http://doi.org/10.1099/jgv.0.000873

Takiuchi, E., Alfieri, A. F., & Alfieri, A. A. (2001). Herpesvírus bovino tipo 1: tópicos sobre a infecção e métodos de diagnóstico. *Semina: Ciências Agrárias, 22*(2), 203–209.

Vargas, D. S., Jaime, J., & Vera, V. J. (2009). Perspectives to control bovine viral diarrhea virus (BVDV). *Revista Colombiana de Ciencias Pecuarias, 22*(4), 677–688.

Vargas-Niño, A., Vargas R, J., Parra-Martin, J. A., Vásquez R, M. C., Góngora O, A., & Mogollón-Waltero, E. M. (2018). Serological status of IBR, BVD, leucosis, Leptospira and Neospora caninum in bovine females of the department of Santander, Colombia. *Revista MVZ Córdoba, 23*(2), 6671–6680. DOI: http://doi.org/10.21897/rmvz.1341