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Article

Epidemiological study of Maedi-Visna in the state of Veracruz

Jorge Ernesto Eliseo Céspedes-Rosas [a]

David Itzcoatl Martínez-Herrera [a*]

Argel Flores-Primo [a]

Violeta Trinidad Pardío-Sedas [a]

Rebeca Isabel Vergara-Reyes [a]

José Alfredo Villagómez-Cortés [a]

[a] Universidad Veracruzana. Facultad de Medicina Veterinaria y Zootecnia. Circunvalación esq. Yañez. Col. Unidad Veracruzana, 91710. Veracruz, México.

* Corresponding author: dmartinez@uv.mx

Abstract:

Maedi-Visna disease (MV) is caused by a retrovirus that affects sheep and causes pneumonic, nervous, and arthritic conditions and mastitis. Its distribution is cosmopolitan, notifiable, and endemic in Mexico. An epidemiological study was conducted in sheep farming basins in the state of Veracruz, Mexico, to determine the seroprevalence of MV and identify its risk factors and spatial distribution in the state. The sample size was 386 animals in 55 randomly selected production units (PUs). Blood serum was collected from the ewes, and the diagnosis was performed in series using two commercial ELISA kits in screening and confirmatory modalities. The distribution map was constructed with QGIS® version 3.0. The overall seroprevalence was 9.59 %, the seroprevalence by municipality was 69.23 %, and the seroprevalence by production unit was 32.14 %. Risk factors were identified as pregnant ewes, coexistence with other ruminant species, sheep aged 13 to 24 mo, the intensive production system, animals located in the Totonaca region, and animals from municipalities

with an altitude of less than 500 m asl and environmental temperature of more than 15 °C (59 °F).

Keywords: State of Veracruz, Cross-sectional study, Lentivirus, Sheep farming, Seroepidemiology.

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Introduction

The Maedi-Visna (also known as progressive pneumonia of small ruminants) is a viral pathology caused by a retrovirus that has a worldwide distribution and belongs to the subclassification of viruses called Lentivirus of Small Ruminants (LvPR)[1]. Four main routes of transmission are recognized[2]: the most frequent is the consumption of infected colostrum or milk during any period of lactation. The horizontal transmission route by respiratory aerosols is also of great importance, especially in intensive and semi-intensive production units, where overcrowding of animals is common; in addition, this type of transmission can be favored by regional practices such as mixed breeding of sheep with animals of other species or overcrowding due to climatic or environmental conditions[3,4]. Fecal-oral transmission also favors the transmission of the causal agent. It has not yet been possible to isolate the virus in feces; however, it has been possible to experimentally infect young animals through water contaminated with feces from positive animals. On the other hand, no evidence of transmission through urine has been found, although the causal agent has been isolated in samples of kidney medullary tissue from infected animals[5].

LvPR infections were first described in Mexico by Adam *et al*. in 1984[6], and since then, it has been identified in Chiapas, Jalisco, Veracruz[7], State of Mexico[8], Sonora[9], Coahuila, Nuevo León, and Tamaulipas[10]. In the state of Veracruz, sheep production is developed mainly as an activity for family consumption without appropriate hygiene measures, biosafety, or veterinary advice; therefore, this study aimed to determine the presence of MV in PUs in the state of Veracruz, Mexico.

Materials and methods

A cross-sectional, multistage, stratified study was carried out. The sample size, calculated with the WinEpi online software for a population of approximately 462,000 sheep, consisted of 386 animals. Blood samples were collected by venipuncture in tubes without anticoagulant

in order to obtain serum and perform an indirect enzyme-linked immunosorbent assay (ELISA) with IDEXX® commercial kits: CHEKIT CAEV/MVV for the screening test and CAEV/MVV VERIFICATION as the confirmatory test. Samples were collected between December 2014 and December 2015 in 13 municipalities in the state of Veracruz, comprised within three zones of the state that are representative of the state's sheep farming:

1) Region III, Totonacapan, has an altitude that ranges between 10 and 740 m asl, depending on the municipality, a temperature of 20 to 26 °C, and an annual rainfall of 1,000 to 1,500 mm. In this region, the municipalities of Coatzintla, Gutiérrez Zamora, Papantla, and Tihuatlán were selected.

2) Region V, Capital, is located at an altitude ranging between 260 and 2,400 m asl, with a temperature of 10 to 25 °C and an annual rainfall of 400 to 2,000 mm; in this region, the municipalities of Altotonga, Emiliano Zapata, Jalacingo, Perote, and Ayahualulco were selected.

3) Region IX, Los Tuxtlas, has an altitude ranging between 10 and 340 m asl, a temperature of 23 to 25 °C, and an annual rainfall of 1,800 to 2,000 mm; in this region the municipalities of Catemaco, San Andrés Tuxtla, Santiago Tuxtla, and Ángel R. Cabada were utilized.

A survey was also conducted to gather information about management and PU in order to identify risk factors. Each PU was georeferenced with a GPS device to establish the distribution of MV in the municipalities considered in this study.

The online statistical software Vassarstats (http://www.vassarstats.net/) was used to calculate seropositivities, 95% confidence intervals, and odds ratios for the different variables as proposed by Thrusfield[11]. The significance value was set at 0.05.

Results

Of the 386 animals sampled, 37 were ELISA positive; thus, the overall seropositivity was 9.5% (Table 1). Likewise, seropositive animals were identified in 9 of the 13 municipalities where samples were collected and in 18 of the 56 herds from which animals were bled, and the seropositivity was 69.2 % and 32.1 %, respectively.

Table 1: Maedi-Visna seropositivity in sheep from the state of Veracruz, Mexico

Seropositivity	Samples (n)	Seropositive (n)	Seropositive (%)	95%CI
Overall	386	37	9.5	6.9 - 13.0
By municipality	13	9	69.2	38.8 - 89.6
By herd	56	18	32.1	20.6 - 46.0

95%CI= 95% confidence interval.

According to their productive stage, pregnant ewes exhibited a seropositivity with a higher odds ratio than that of empty ewes ($P<0.05$) (Table 2). The other groups considered had higher seropositivity and odds ratios, but the values were not significant.

Table 2: Maedi-Visna seropositivity in sheep from the state of Veracruz, Mexico, by sex and productive stage

Productive stage	Total (n)	Seropositive (n)	(%)	95%CI	OR	*P* value
Open	131	7	5.3	2.3 - 11.1	1.0	-
Yearling	17	2	11.7	2.0 - 37.7	2.36	0.27
Pregnant	125	15	12.0	7.1 - 19.3	2.41	0.04
Lactating	24	4	16.6	5.4 - 38.1	3.37	0.07
Weaned	5	1	20.0	1.05 - 70.1	4.42	0.26
Sire	84	8	9.5	4.4 - 18.4	1.86	0.18

95%CI = 95% confidence interval. OR= odds ratio.

When analyzing Maedi-Visna seropositivity according to the characteristics of the animals and their management, it was found that in herds where there is no coexistence with other animals, in this case only goats, the seropositivity and odds ratios were lower. Similarly, ewes managed in a semi-intensive system exhibited lower seropositivity than those raised in stalls or those kept in pastures. Interestingly, older animals exhibited a lower seropositivity than other age groups. The seropositivity determined among males was similar to that determined in females (Table 3).

Table 3: Maedi-Visna seropositivity in sheep in the state of Veracruz, Mexico, as a function of animal characteristics and management

Variable	Total (n)	SP (n)	SP (%)	95%CI	OR	*P* value
Coexistence with other ruminants:						
No	280	16	5.7	3.4 - 9.2	1.0	-
Yes	106	21	19.8	12.9 - 28.9	4.07	0.00007
Production system:						
Semi-intensive	128	7	5.4	2.4 - 11.3	1.0	-
Grazing	220	22	10.0	6.5 - 14.9	1.92	0.09
Stabled	38	8	21.5	10.1 - 37.7	4.61	0.007
Age range (months):						
6 – 12	73	6	8.2	3.3 - 17.4	1.46	0.35
13 – 24	94	14	14.8	8.6 - 24.0	2.85	0.02
25 – 36	98	10	10.2	5.2 - 18.3	1.85	0.16
>36	121	7	5.7	2.5 - 11.9	1.0	-
Sex:						
Male	84	8	9.5	4.4 - 18.4	1.0	-
Female	302	29	9.6	6.6 - 13.6	1.0	0.58

SP= seropositive; 95%CI= 95% confidence interval, OR= odds ratio.

When considering the effect of some geographical and climatic factors on Maedi-Visna seropositivity, a higher frequency was observed in the Totonaca region than in the capital. It was also observed that there is a greater chance of finding seropositive animals at altitudes below 500 m asl than at other altitudes. The climate also seems to exert some influence, since seropositivity is higher in localities where the mean annual temperature exceeds 15 ^{o}C and where the mean annual rainfall exceeds 500 mm.

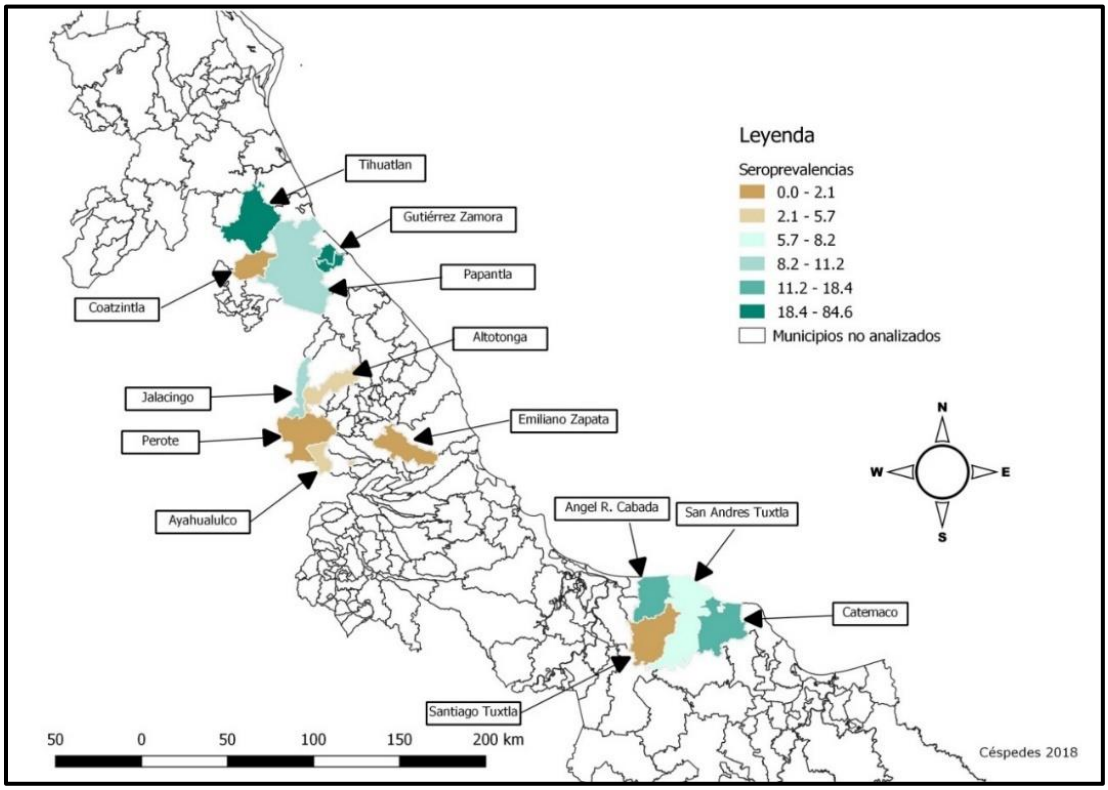
Table 4: Maedi-Visna seropositivity in sheep from the state of Veracruz, Mexico, as a function of geographic and climatic factors

Variable	Total (n)	SP (n)	SP (%)	*95%CI	OR	*P* value
Region:						
Capital	165	7	4.2	1.8 - 8.8	1.0	-
Los Tuxtlas	153	12	7.8	4.3 - 13.6	1.92	0.13
Totonaca	68	18	26.4	16.4 - 38.7	8.13	0.000003
Altitude, m:						
<500	197	30	15.2	10.6 - 21.1	3.72	0.0008
501-1000	37	0	0	0 - 11.7	-	-
>1001	152	7	4.6	2.0 - 9.6	1.0	-
Annual temperature, °C:						
10-15	152	7	4.6	2.0 - 9.6	1.0	-
>16	234	30	12.8	8.9 - 17.9	3.04	0.004
Annual rainfall, mm:						
<500	79	2	2.5	0.4 - 9.6	1.0	-
501-1000	77	10	12.9	6.7 - 23.0	5.37	0.018
1001-1500	85	9	10.5	5.2 - 19.6	4.56	0.037
>1501	145	16	11.0	6.6 - 17.5	4.77	0.018

SP= seropositivity; 95%CI= 95% confidence interval; OR= odds ratio.

Only in three of the municipalities included in the study were no positive animals identified. However, they belong to different regions and exhibit different Maedi-Visna seropositivity. Figure 1 shows the distribution of the municipalities, and Table 5 shows the descriptive analysis.

Figure 1: Localization and seropositivity to Maedi-Visna in sheep from tested municipalities in the state of Veracruz, Mexico



The seropositive animals were found in 9 of the 13 sampled municipalities, which are located within the three regions considered in this study. The Maedi-Visna virus was found in sheep in the three main sheep-producing areas of the state of Veracruz, although, except for the municipality of Gutiérrez Zamora, the seropositivity rate in the municipalities was less than 25 % (Table 5).

Table 5: Maedi-Visna seropositivity by municipality in sheep in the state of Veracruz, Mexico

Municipality	Total (n)	SP (n)	SP (%)	95%CI	OR	*P* value
Gutiérrez Zamora	13	11	84.6	53.7 - 97.2	107.25	0.0000005
Tihuatlán	16	4	25.0	8.3 - 52.5	6.5	0.046
Papantla	32	3	9.3	2.4 - 26.1	2.01	0.38
Coatzintla	7	0	0.0	0 - 43.9	-	-
Jalacingo	36	3	8.3	2.1 - 23.5	1.77	0.437
Altotonga	37	2	5.4	0.4 - 19.5	1.11	0.652
Ayahualulco	41	2	4.8	0.8 - 17.4	1.0	-
Perote	38	0	0.0	0 - 11.4	-	-
Emiliano Zapata	37	0	0.0	0 - 11.7	-	-
Ángel R. Cabada	38	6	15.7	6.5 - 31.9	3.65	0.108
Catemaco	32	4	12.5	4.0 - 29.8	2.78	0.227
San Andrés Tuxtla	27	2	7.4	1.2 - 25.8	1.56	0.522
Santiago Tuxtla	32	0	0.0	0 - 13.3	-	-

SP= seropositivity; 95%CI= 95% confidence interval; OR= odds ratio.

Discussion

Of the 386 ewes sampled, 37 were found to be seropositive for Maedi-Visna, resulting in an overall seropositivity of 9.5 %. This proves the presence of this agent in sheep in the state of Veracruz, since, according to information provided by the producers, the animals were born within the flocks, or were acquired in nearby localities or municipalities. This identification is important because epidemiological information on lentivirus in small ruminants in Mexico is scarce[7,10], and the state of Veracruz is the third largest sheep producer in the country[12]. The overall seropositivity obtained in this study was lower than the 25.4 % reported in sheep in northeastern Mexico[10]. However, this research only considered sheep, goats, or mixed flocks as a management variable; therefore, it is not possible to infer whether this difference can be attributed to other variables. In this sense, it is necessary to better understand the specific characteristics in the herds that promote the viability and dispersion of the agent,

since control programs must be precise and adapted to the conditions of each PU[1,13]. On the other hand, it was possible to identify seropositive animals in several municipalities located in the three study regions (Figure 1), so the Maedi-Visna virus may be assumed to be endemic and widely distributed in Veracruz, with the municipalities of Tihuatlán and Gutiérrez Zamora being the most severely affected; these are located in the Totonaca region, in the north of the state, which exhibited the highest seropositivity, possibly due to the intensive production systems that predominate in that region, and showed a higher seropositivity to MV than other production systems, such as pasture or semi-intensive grazing. In this regard, it has been established that this type of management predisposes the spread of the MV virus[14,15] as a consequence of permanent overcrowding and poor ventilation, which favor the propagation of aerosols; it is therefore advisable to implement management alternatives to prevent herd infection.

In extensive production systems, seropositivity was 10%, which is explained by the fact that the risk of contagion decreases because animals tend to graze in search of food in a larger space and interact less with each other[10,14]. Given that this type of extensive management predominates in the sheep-producing areas of Veracruz, this could be favorable to reduce the potential impact of MV in other states where the concentration of sheep in reduced spaces is prevalent. Although a lower seropositivity was determined in semi-intensive PU, it cannot be concluded that there is a relationship between this kind of management and a lower risk of infection, given the characteristics of the agent and the number of PU that develop this system and were selected to be included in this research.

On the other hand, a seropositivity of 12 % was determined in pregnant females, and the intrauterine and transplacental routes have been suggested as possible important routes of transmission of the causal agent of MV[1]; since females used as breeders tend to remain for considerable periods in the PU, they have a high capacity to infect offspring and to maintain the agent in the herds. Likewise, the presence of seropositivity in lactating lambs and weaned animals poses a risk of dissemination of the agent because the route through infected colostrum or milk is considered the most important for the spread of the virus[16]. In this sense, lambs represent a source of distribution among other PUs or localities, given that, although some of them are used for consumption at an early age, others are traded for breeding stock or exchanged as studs between producers, and the worldwide distribution of MV is attributed to the uncontrolled movement of animals[17]. In addition, infection of breeding females and lambs is implicated in direct losses caused by the MV virus: according to a review by Azevedo *et al*[18], decreased productivity is a consequence of reduced milk production, lactation period and life span, as well as of predisposition to bacterial infections in the mammary gland. Eight sires were also identified as seropositive for the agent in this study; due to their zootechnical function, these animals remain for years in the PU and can carry and transmit the infection during their long productive life. This point is of utmost relevance, given that it has been established that one of the effective measures to prevent the

spread of the virus is that sires for mating or semen collection must be confirmed as free of MV[19].

A higher seropositivity was identified in sheep from the Totonaca region than in those from the other two regions sampled; this may be related to two variables that were consistently identified in the PU of the region: the intensive production system and the climatic characteristics of altitude and ambient temperature of the region.

Coexistence with other species was identified as a risk factor (P=0.0007), in this particular case with goats, because goats can become infected and eliminate the agent through their milk[20]. Therefore, it is of utmost importance to encourage producers to separate the species or adequately clean common areas in order to reduce the risk.

Finally, it should be noted that infection can occur regardless of the management or the zootechnical purpose of the ewes[21]; moreover, MV is currently included in Group III: Endemic Diseases and Pests of Mandatory Monthly Notification in Mexico[22]. It is therefore considered a lower epidemiological and economic risk; notwithstanding, in the absence of further research and periodic and constant surveillance campaigns, MV could be an underestimated issue for Mexican sheep production. Therefore, although antibodies against the agent were detected in the present study, direct identification of the agent with other diagnostic techniques ―namely, isolation, immunofluorescence, immunohistochemistry, or molecular tests― would be relevant today both in Veracruz and in other Mexican states.

Conclusions and implications

Serological evidence of the presence of the MV virus was found in sheep from the Totonaca, Capital and Los Tuxtlas regions in the state of Veracruz, and so were various conditions that could favor endemicity, dissemination of the causal agent, and infection within the flocks, potentially causing considerable productive losses and economic impact. Although more specific studies are needed to establish this impact, it is advisable to promote adequate management practices among sheep producers in the state to avoid the spread of not only of the MV virus but also other infections that impair sheep production. Collaboration between producer associations, veterinarians, and government agencies is essential to implement monitoring campaigns and efficient control measures for the benefit of sheep producers.

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Conflict of interest

The authors declare that they have no conflict of interest.

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