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## Environmental factors associated with the prevalence of *Haemonchus* spp in lambs from the central zone of Sinaloa

Factores ambientales asociados a la prevalencia de *Haemonchus* spp en corderos de la zona centro de Sinaloa

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### ABSTRACT

Sheep are a species exploited in different areas of production. They are prone to different pathogens, highlighting parasites such as *Haemonchus contortus*. The prevailing climate and husbandry management practices are considered the main factors driving the spatial and temporal distribution of the nematode. Its distribution is worldwide, causing economic losses due to morbidity and mortality, and prevalence studies have been reported in different countries such as India, Spain, Nigeria, and Mexico; therefore, the aim of this study was to determine the edaphoclimatic factors of different zones from Culiacan municipality and the production system that influence the prevalence of *Haemonchus* spp. in lambs. The research was carried out in Culiacan municipality, Sinaloa, Mexico, over a period of one year. It was an observational study, which included 23 sheep production units distributed in 10 districts of the municipality, with a total of 1520 samples of feces from animals under 3 months of age. Feces were processed individually by flotation technique. The overall prevalence was 13.42 %, and the autumn season (OR 2.38 (1.69-3.34) P<0.001), valley zone (OR 2.70 (1.21-6.02); P<0.016) and extensive system (OR 4.81 (3.38-6.85); P<0.0001) were risk factors associated with the presence of the nematode in lambs, so they should be considered for the establishment of preventive measures and control of parasitosis.

**Keywords:** *Haemonchus*, prevalence, sheep, risk factor, gastrointestinal nematode.

### RESUMEN

Los ovinos son una especie explotada en diferentes ámbitos de la producción. Estos son propensos a diferentes patógenos, destacando parásitos como *Haemonchus contortus*. El clima predominante y las prácticas de manejo en la crianza se consideran los principales factores que impulsan la distribución espacial y temporal del nematodo. Su distribución es mundial, ocasiona pérdidas económicas por morbilidad y mortalidad, se han reportado estudios de prevalencia en diferentes países como en India, España, Nigeria, México; por ello, el objetivo de este trabajo fue determinar los factores edafoclimáticos de las distintas zonas del municipio de Culiacán y sistema de producción que influyen en la prevalencia de *Haemonchus* spp en corderos. La investigación se realizó en el municipio de Culiacán, Sinaloa, México, en un periodo de un año, fue un estudio observacional, para el cual se incluyeron 23 unidades de producción ovina distribuidas en 10 sindicaturas del municipio, se realizó un muestreo por época del año colectando un total de 1520 muestras de heces procedentes de animales menores a 3 meses de edad. Las heces se procesaron individualmente por técnica de flotación. La prevalencia general fue de 13.42 %, y la época de



otoño (OR 2.38 (1.69-3.34)  $P < 0.001$ ), zona de valle (OR 2.70 (1.21-6.02);  $P < 0.016$ ) y sistema extensivo (OR 4.81 (3.38-6.85);  $P < 0.0001$ ) resultaron factores de riesgo asociados a la presencia del nematodo en los corderos, por lo que deben considerarse para el establecimiento de medidas preventivas y de control de la parasitosis.

**Palabras clave:** *Haemonchus*, prevalencia, ovinos, factor de riesgo, nematodo gastrointestinal.

## INTRODUCTION

Gastrointestinal nematode infections affect the health of small ruminants compromising their production and reproduction, more frequently in young developing animals, causing low weight gain and growth retardation, making them one of the main causes of economic losses (González *et al.*, 2011; Asmare *et al.*, 2016; Kuma *et al.*, 2019), mainly in the costs incurred in treatment and control (Tramboo *et al.*, 2015). Sheep are generally more prone to gastrointestinal parasitism, due to their feeding on larvae 3 contaminated pastures (Tariq *et al.*, 2008). Among gastrointestinal parasites, *Haemonchus* is the species with the highest economic importance (Rinaldi *et al.*, 2015). This nematode is located in the abomasum, feeds on the blood of sheep and goats, can be found in other ruminants such as cattle (Getachew *et al.*, 2007), it is among the most pathogenic in sheep (Besier *et al.*, 2016), its extensive geographical distribution and resistance against anthelmintic control measures makes it a threat to the sustainability of sheep farming (Saccareau *et al.*, 2017). The rainy season favors its frequency, with animals grazing during the early morning hours (Mederos *et al.*, 2010), the prevailing climate (temperature, rain and humidity) and management practices in husbandry are considered the main factors driving its distribution (Rinaldi *et al.*, 2015); its distribution is heterogeneous and depends on variables that differ from one area to another, even from one farm to another, such as management, prevention and control (Musella *et al.*, 2011). On the other hand, studies on the prevalence in grazing animals is high in areas with tropical climates in both hemispheres (O'Connor *et al.*, 2006), young animals and pregnant females are more susceptible to helminths as opposed to adult animals due to their nutritional status and low level of immunity (Vieira *et al.*, 2014). The prevalence of *Haemonchus* has been reported worldwide, in India Tramboo *et al.* (2015) out of a total of 1200 animals sampled 55 % were positive to the nematode; in Mexico, 32 % were found positive out of 219 sampled from grazing sheep (Hernández *et al.*, 2007); in the Sinaloa region when analyzing 120 sheep from an extensive production system a frequency of 17.5 % was reported (Gaxiola *et al.*, 2010). Therefore, the aim of this study was to determine the edaphoclimatic factors of different zones from Culiacán municipality and the production system that influence the prevalence of *Haemonchus* spp. in lambs in the central zone of Sinaloa.



## MATERIAL AND METHODS

### Study area

The study was carried out in Culiacán municipality, Sinaloa, Mexico (24° 46' 13" NL and 107° 21' 14" WL). The region is characterized by a BS1 (h') w(w)(e) climate, defined as semi-dry, very warm, with rainfall in summer, according to the Köppen classification and modified by García (2004); with an average annual temperature of 25.9 °C, with an average maximum of 30.4 °C in June and July, and an average minimum of 20.6 °C in January; the average relative humidity is 68%, with a maximum of 81% in September and a minimum of 51% in April; the average annual rainfall is 688.5 mm (CIAPAN, 2002).

### Type of study and sample size

This is an observational, cross-sectional, descriptive study. Twenty-three extensive, semi-intensive and intensive ranches were sampled. In the municipality of Culiacán, 125 Sheep Production Units (SPU) are registered (SIAP, 2013), so the sample represented 18.4% of the SPUs. Ten of the 18 districts of Culiacán municipality, Sinaloa were considered (Figure. 1), the selection of the production units was made by convenience, based on the cooperation of the owner and ease of access. The sample size was determined with the following formula:

$$n = \frac{Z^2 pq}{B^2}$$

Where: n is the sample size, Z is 1.96 for 95 % confidence, p is the expected frequency of the factor to be studied, q is 1-p, B is precision or admitted error (Jaramillo & Martínez, 2010). The calculated sample size was 380 feces samples in each season (summer, fall, winter and spring) and because the number of lambs in the production unit was unknown at the time of the visit, the number of adults was considered (Table 1), and a number of lambs representing at least 10% of the adults in each production unit was sampled, which were randomly selected to complete the calculated sample size. Sampling was conducted by time of year and a total of 1520 samples were obtained from lambs less than ninety days (d) of age.

### Sample collection and laboratory analysis

During each visit to the production units, a form was filled out to record information on the following factors: time of year (summer, fall, winter or spring), location (highlands or valley), production system (intensive, semi-intensive, extensive). The lambs were chosen randomly. Feces were taken directly from the rectum with a latex glove, individually identified, and refrigerated in containers at 4 °C with ice and refrigerants for transfer to the Parasitology laboratory of the Faculty of Veterinary Medicine and Animal Husbandry for processing and analysis.



The diagnosis of *Haemonchus* spp was performed by coproparasitoscopic analysis using the qualitative Faust flotation technique (Zajac & Conboy, 2011), being a useful and widely used means in preliminary studies to determine what types of parasites are present in fecal samples (Medeiros *et al.*, 2018), for parasite detection, optical microscopy with 10x and 40x objectives was used, eggs were identified based on their characteristic morphology, dark brown blastomeres and size described by Ljungström *et al.*, 2018.

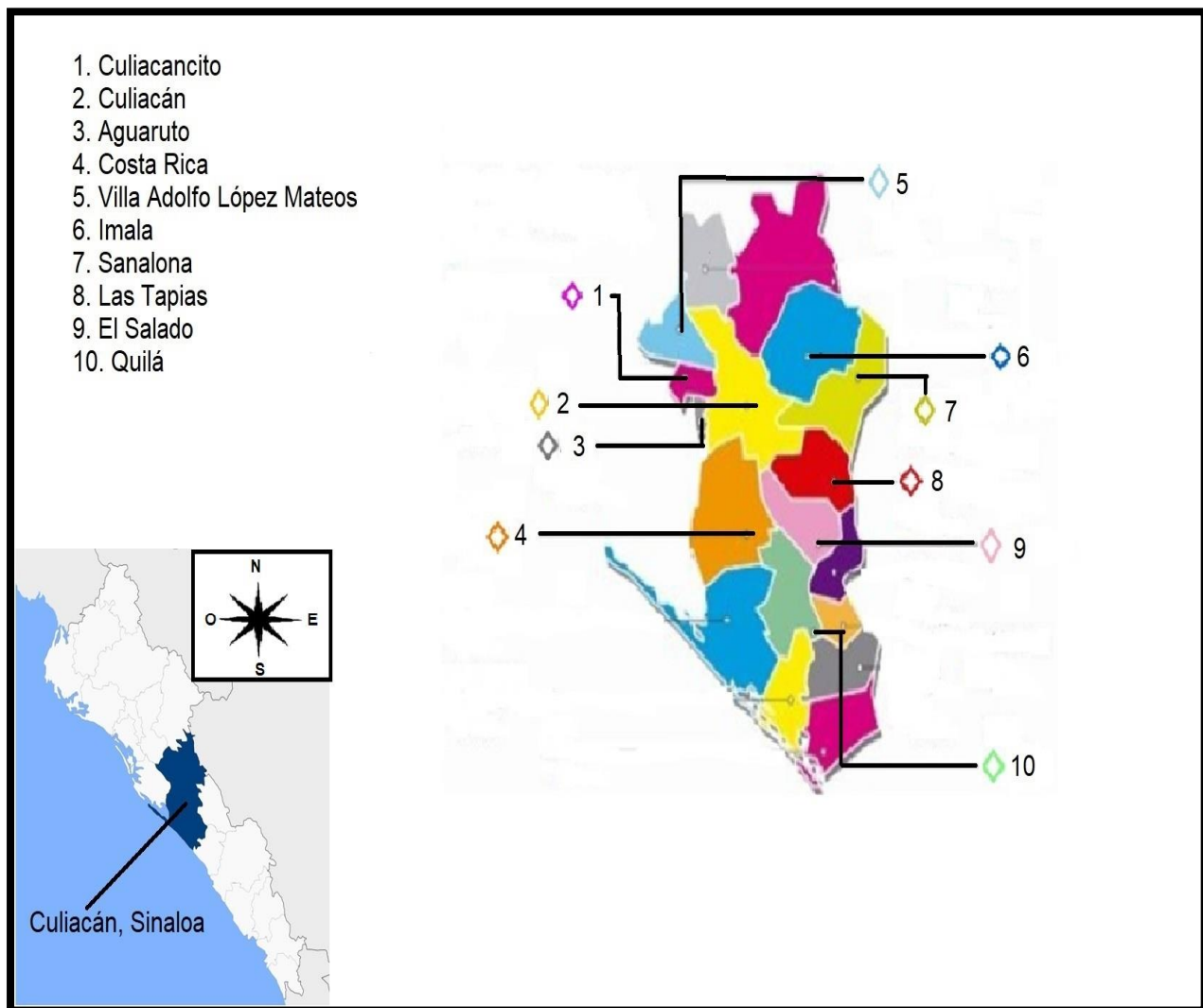


Figure 1. Location of the sheep production units sampled in Culiacán Municipality, Sinaloa, Mexico.



**Table 1. Location of sheep production unit by district and population of adult sheep**

District	*SPU	Adult sheep	Lambs sampled
Villa Adolfo L.M	Agrícola Limón	80	10
	La Hacienda	150	20
	La granjita	120	17
	Alboradas	180	18
El Salado	El alacrán	70	11
Costa Rica	El trabajo	20	5
	Agrícola Sanfer	150	15
	Agrícola Tabachines	180	18
Quilá	Naranjos	70	12
	La Loma	30	5
Aguaruto	Fetasas	200	22
	Agrícola del Río	45	5
	Los Cabritos	200	20
Culiacancito	San Sebastián	250	25
	Agrícola Quiroz	250	25
	Los Otates	180	20
Las Tapias	Santa María	130	13
Sanalona	Baldomero	20	5
Imala	Guayacanes	200	24
Culiacán	Guásima	230	23
	Agrícola Mojolo	300	32
	Ganadera Verdugo	80	15
	Campo Morelia	180	20

\*SPU= Sheep Production Units



## Statistical analysis

Lambs were considered positive with at least one *Haemonchus* spp egg; prevalence was estimated as the number of positive sheep among the total sheep sampled according to category.

The results of microscopic observation (positive or negative) were summarized in contingency tables by factor and analyzed for association between the result and the factor, using the Chi-squared test. Statistical difference was considered statistical difference with a  $P \leq 0.05$ .

For factors with more than two categories, the results were dichotomized. Next, to determine the risk factors for positive results, multivariate logistic regression analysis was applied. The general model was:

$$\pi(x) = \frac{\exp(\alpha + \sum \beta_i x_i)}{1 + \exp(\alpha + \sum \beta_i x_i)}$$

Where:  $\pi(x)$ , the value of  $\pi$  can vary as the value of  $x$  changes, and we want to describe its dependence; the values of  $x_i = (x_1, \dots, x_p)$  are the predictor variables  $p$ ,  $x_i$  represents the vector of independent variables;  $\exp$  is the base of the natural logarithms 2.71828;  $\alpha$  is the value of the intercept;  $\beta_i$  are the values of the regression coefficients. For this analysis, the LOGISTIC procedure (SAS, 2001) with the backward option was used to estimate the degree of association [odds ratio (OR)] and confidence intervals. The alpha level to consider association between the factor with the positive outcome, and to estimate the risk factor was  $P \leq 0.05$ .

## RESULTS AND DISCUSSION

From a total of 1520 samples analyzed, 204 were positive for *Haemonchus* spp representing a prevalence of 13.42 % in the annual period analyzed; the method used to detect the nematode was the flotation technique, a test mainly used in stool examination in animal diagnosis, it concentrates eggs, parasite oocysts and separates the debris present in the sample (Zajac & Conboy, 2011; Rinaldi *et al.* 2011; Medeiros *et al.*, 2018), one of the main advantages of using this test is that it has a high egg recovery rate (Medeiros *et al.*, 2018), causes less damage to cysts and eggs (Zajac & Conboy, 2011), which allows proper morphological identification facilitating the characteristic structure observation, egg dimensions to be identified (Indre *et al.*, 2010; Mahmood *et al.*, 2019), the technique prevents flotation of trematode eggs and it is not as specific to determine the species of parasites observed (Zajac & Conboy, 2011; Ljungström *et al.*, 2018), therefore results using this technique report only the *Haemonchus* genus. Factors studied





are presented in Table 2, Chi-Square tests indicated that the three factors were significant ( $P < 0.05$ ), year time, production zone, production system; similarly for the analysis of risk factors were significant ( $P \leq 0.05$ ) in the multivariate logistic regression model (Table 3). The results by year time showed a higher prevalence in autumn with 20.53 %, no difference was observed between winter and spring and the lowest prevalence was found in summer with 7.89 % ( $P < 0.0001$ ).

**Table 2. Risk factors associated with the presence of *Haemonchus* spp. eggs in feces of lambs in SPU located in the municipality of Culiacán, Sinaloa, Mexico**

Risk factor	N	Positive samples	Percentage	P <sup>1</sup>
<b>Year season</b>				0.0001
Summer	380	30	7.89 <sup>c</sup>	
Autumn	380	78	20.53 <sup>a</sup>	
Winter	380	50	13.16 <sup>b</sup>	
Spring	380	46	12.11 <sup>bc</sup>	
<b>Production zone</b>				0.0001
High	183	7	3.83 <sup>b</sup>	
Valleys	1337	197	14.73 <sup>a</sup>	
<b>Production system</b>				0.0001
Extensive	437	112	25.63 <sup>a</sup>	
Semi-intensive	248	50	20.16 <sup>a</sup>	
Intensive	835	42	5.03 <sup>b</sup>	

<sup>1</sup>Probability values of the Chi-square test. <sup>abc</sup> Different literals in the percentages of positive samples in each risk factor indicate statistical difference ( $P \leq 0.01$ ).

**Table 3. Odds ratios for risk factors associated with the presence of *Haemonchus* spp. eggs in lamb feces in SPU located in the municipality of Culiacán, Sinaloa, Mexico**

Risk factor	Odd ratio	CI 95 %	P Value
<b>Year season</b>			
S-W-S	Reference		
Otoño	2.38	1.69-3.34	0.001
<b>Production season</b>			
High	Reference		
Valleys	2.70	1.21-6.02	0.016
<b>Production system:</b>			
Intensive-Semi	Reference		
Extensive	4.81	3.38-6.85	0.0001

S=Summer, W=Winter, S=Spring; CI = Confidence interval; P = Probability.



The results of the study showed that when comparing the different seasons of the year, there is a 2.38 times higher risk of presenting *Haemonchus* spp. in autumn than in the rest of seasons ( $P < 0.001$ ). The results of the present investigation in relation to the summer season with 7.89 % are close to those described in England with 10.5 % (Broughan & Wall., 2007), the low prevalence in summer can be attributed to the high temperatures that occur at this time in the area described, which is why the larvae decrease their activity, due to the negative phototropism to intense light (Soca *et al.*, 2005), in another study related to year times in India reported the presence of the order *Strongylida* with 63.2 % in summer and 58.4 % in autumn, 52.77 % in winter and 61.3 % in spring (Tramboo *et al.*, 2015), denoting difference with the current work, since in autumn there was a prevalence of 20.52 % being higher than that of summer with 7.89 %, this can be interpreted by the maximum temperatures that occur in summer in the study area, in addition to the increased humidity in the pastures in the autumn season that favor larval migration by positive hydrotropism (Soca *et al.*, 2005), regarding winter and spring, differences were also found between the two studies, since in India the prevalence increased from winter to spring, this is attributed to the precipitation that favors the humidity of the pastures for the presence of the parasite. Contrary to the present result, there was no significant difference from winter to spring in relation to the nematode presence, this is interpreted to the state of hypobiosis in which the helminth enters, in an unfavorable period nutritionally, so it tends to lower its metabolism and activity (Soca *et al.*, 2005); in a study conducted in Iran (Moghaddar, 2008), sampled lambs under five months, during the four seasons of the year, it was reported the presence of nematodes 25.9, 22.3, 50 and 53.1 % for autumn, winter, spring and summer, respectively. The differences that stand out among the studies is the report of nematodes in general and therefore a higher percentage of positive lambs, another factor that indicates a difference between the prevalence between seasons is precipitation. In the case of Iran, there is more rainfall in March and April, so there are more nematodes in spring and summer, and in the study region of Culiacán, rainfall occurs in August and September, favoring the conditions for *Haemonchus* in autumn, coinciding with a study conducted in the same area with the protozoan *Cryptosporidium* spp. in lambs, showing 2.2 times more risk of presenting the parasite in autumn than in summer (Castro *et al.*, 2017), although they are taxonomically classified in different phylum the climatic conditions at this time favor both.

When analyzing the geographic zone of the region, the results indicate a higher prevalence for the valley with 14.73 % as opposed to the highland zone with 3.83 % ( $P < 0.0001$ ), and 2.70 times higher risk of presenting the nematode in lambs in the valley zone ( $P < 0.016$ ), presenting similarity to what was found in Ethiopia the difference that was reported in general were gastrointestinal nematodes, in valley production zone was higher than the high zone, with 95 and 68.6 % respectively and coincide in terms of higher risk of presenting *Haemonchus* in the valley production zone (Asmare *et al.*, 2016),





likewise in Switzerland in high zones a low presence of the parasite was reported in comparison with medium and low zones in Italy and Ireland ([Rinaldi et al., 2015](#)), the water source is one of the key characteristics for the survival and dissemination of nematodes such as *Haemonchus* ([Musella et al., 2011](#); [Rinaldi et al., 2015](#)), which occurred more in valley areas mainly because of dams, these areas are lower areas that can carry contaminants through water tributaries, including parasite eggs from production units or wild animals that live in higher areas, favoring the dissemination of these when this water is used for irrigation, animal consumption, among other activities of common use. Gastroenteric parasites and especially *Haemonchus* have managed to adapt to different ecosystems, their ability to adapt and survive different environments allow the infection of new hosts ([Munguía et al., 2018](#)).

According to the production system the presence of *Haemonchus* spp was found 25.63 % in extensive production system, 20.13 % in semi-intensive system and 5.03% in intensive system ( $P < 0.0001$ ) and when analyzing the association of parasitosis this was 4.81 times more risk of occurrence in lambs under an extensive production system ( $P < 0.0001$ ). The results differ from those performed by [Zapata et al. \(2016\)](#) and [Herrera et al. \(2013\)](#) in which no statistical difference was found among the three production systems analyzed. The conditions of the present study in animals under extensive systems are not systematized, they did not manage a deworming schedule, which agrees with [Mederos et al. \(2010\)](#) who indicate that the highest levels of gastrointestinal parasites occur in production units where there is no routine deworming management; in addition to the type of grazing-based feeding facilitates the ingestion of the parasite larvae present in the vegetation, and allows transmission as the animals themselves contaminate the grazing area ([Belina et al., 2017](#); [Akyüz et al., 2019](#)); on the other hand, in the semi-intensive and intensive systems, deworming management was programmed and feeding was operated with diets prepared mainly in the intensive system which favored a lower percentage of the parasite presence in these systems due to the positive impact given by better quality diets on health; [Cériac et al. \(2019\)](#) and [Naeem et al. \(2021\)](#) point out that quality nutrition supplemented with high protein, amino acids stimulates the expression of host resistance and resilience, stimulates immunity, decreases parasite proliferation.



## CONCLUSION

The autumn season, valley area and extensive production system are the edaphoclimatic factors associated with the prevalence of *Haemonchus* spp (13.42%) in lambs in Culiacán municipality, Sinaloa; therefore, these aspects should be taken into account to develop strategies for the prevention and control of parasitosis. *H. contortus* is recognized as one of the main parasites affecting sheep, which makes it necessary to consider this agent as a possible cause of productive disorders even in early stages of the animals' age, as well as a possible infection source from young animals to the rest of the flock.

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