

GASTROINTESTINAL PARASITISM AND USEFULNESS OF FAMACHA® IN GOATS FROM LOJA PROVINCE, SOUTHERN ECUADOR

Parasitismo Gastrointestinal y Utilidad del Sistema Famacha® en Caprinos de la Provincia de Loja, Ecuador

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ABSTRACT

The infection with gastrointestinal (GI) parasites represents one of the main constraints for small ruminant production, health and welfare. Based on official information about mortality of goats related to a two-year drought, the aim of the present investigation was to evaluate GI parasitism, packet cell volume (PCV) values and compared them with FAMACHA® score system in Garza Real locality at Loja Province, Southern Ecuador. A total of 77 individual faecal samples from seven goat farms were analyzed by quantitative and qualitative parasitological methods; for differential larval count, coproculture were carried out with a pool of faeces from each one of the seven farms. From the total of samples, 91% were found to be infected with GI parasites. A mixed infection with two to four types of GI parasites was found in 87.82% of the animals. Based on morphology, eggs from order Strongylida, genera *Strongyloides*, *Trichuris* and *Moniezia* and oocysts of genus *Eimeria* were identified by flotation technique. Level of infection was high in four of the farms. In cultures, six genera of larvae were identified: *Haemonchus contortus*, *Trichostrongylus* spp., *Oesophagostomum* spp., *Teladorsagia* spp., *Cooperia* spp. and *Strongyloides* spp., *Haemonchus contortus* and *Trichostrongylus* spp. were the predominant species in six of the farms. A strong and negative correlation (-0.9418; P< 0.001) was established between PCV values and score assignment and a strong and positive correlation between fecal egg counts and FAMACHA® scores (0.9763; P< 0.005). The results suggest that a combination of effective treatments, supplementary food and application of FAMACHA® system should be applied in the locality of study and furthermore, in the goat farms of the entire municipality.

Key words: Gastrointestinal parasites; FAMACHA®; goat; Ecuador.

RESUMEN

La infección por parásitos gastrointestinales (GI) representa una de las mayores limitaciones en la producción, sanidad y bienestar en la cría de pequeños rumiantes. Con base en información oficial sobre la mortalidad de caprinos relacionada con dos años de sequía, el objetivo de la presente investigación fue evaluar el parasitismo GI, los valores de hematocrito y compararlos con el sistema FAMACHA® en la localidad de Garza Real en la provincia de Loja, Ecuador. Un total de 77 muestras individuales de heces obtenidas en siete explotaciones caprinas fueron analizadas por métodos parasitológicos cuantitativos y cualitativos; para la identificación de larvas, se llevaron a cabo coprocultivos utilizando un pool de heces de cada una de las siete fincas. Del total de muestras, 91% estaban infectadas con parásitos GI. Se encontraron infecciones mixtas con dos a cuatro tipos de parásitos GI en 87,82% de los animales. Con base en la morfología, se identificaron mediante técnicas de flotación, huevos del Orden Strongylida, géneros *Strongyloides*, *Trichuris* and *Moniezia* y oocistes del género *Eimeria*. El nivel de infección fue alto en cuatro de las siete explotaciones. En los cultivos, se identificaron seis géneros de larvas: *Haemonchus contortus*, *Trichostrongylus* spp., *Oesophagostomum* spp., *Teladorsagia* spp., *Cooperia* spp. y *Strongyloides* spp., *H. contortus* y *Trichostrongylus* spp. fueron las especies predominantes. La correlación entre los valores de hematocrito y la asignación de la categoría en FAMACHA® fue fuerte y negativa (-0,9418; P< 0,001) y entre el conteo de huevos y los criterios en FAMACHA® fuerte y positiva (0,9763; P< 0,005). Estos resultados sugieren que una combinación de tratamientos efectivos, suplementos alimenticios y la aplicación del sistema FAMACHA® debería ser aplicado sistemáticamente en la localidad de estudio y extensivamente, a los rebaños caprinos de toda la provincia.

Palabras clave: Parásitos gastrointestinales; FAMACHA®; caprinos; Ecuador.

INTRODUCTION

Gastrointestinal (GI) parasitism is considered one of the most serious and underestimated problems, which impedes small ruminant productivity [3] by decreasing voluntary feed intake, live weight gain, milk yield and carcass quality [2]. Parasitism of the digestive tract can be described as a nutritional disease, because of the increased nutritional demands, but has additional pathophysiological impacts. It decreases food digestibility, causes disruption of the digestive processes and nutrients absorbed via the digestive tract can be diverted from tissues in order to replace losses caused by GI parasites and to guarantee host survival [9].

Goat (*Capra hircus*) breeding in Ecuador is principally developed at the Southern Province of Loja in Southwestern Municipality of Zapotillo (4°23'11"S 80°14'37"O), characterized by a semidry climate with temperatures between 27 to 30°C and rainfall levels of 500-750 mm/year. Based on the last governmental census 2010, Zapotillo Municipality has 28,000 goats raised in an extensive way in six localities (Limonas, Garza Real, Mangahurco, Paletillas, Bolaspamba, Zapotillo) and constitutes the main economic income for the habitants [10]. According to the last Development and Territorial Plan of the Municipality, goat production has suffered a severe decline in population in the past five years [6]. An outward reason could be the construction throughout years 1999-2014 of an artificial canal from the Chira-Catamayo system [18] which enabled water supplies for agriculture but apparently diminished the available space for feeding of goats. Farmers also reported the drowning of goats when jump into the canal (unpublished data). Although there are no official reports on the impact of the canal in goat breeding, the number of goat population of 71,879 heads dropped up to 61% between 2001 to 2011 [6]. In May 2015, veterinarians from the Ecuadorian Agency for Assuring Quality of the Agriculture (Agrocalidad) suggested that deaths of more than 70 goats in farms from Garza Real locality maybe related to GI parasitism and nutritional stress after a two-year drought (unpublished data).

In this regard, FAMACHA© system was developed in South Africa and uses the estimation of anaemia, based on clinical evaluation of the colour of the lower eyelid mucous membrane, as a morbidity marker for haemonchosis [15] and therefore, indicates the urgency of antihelminthic treatments. It has been used to detect and treat GI parasitism at field in several countries [14, 16, 21, 22]. Although possible variations in the results obtained by the system can occur among management systems, animal breeds, types and ages of animals, system operators, environments and facilities, FAMACHA© would be helpful after training farmers in those localities which are distant to the laboratory or the veterinary services [14, 28], as in the case of goat farms in Ecuador. This type of evaluation has not been done previously in the country and it could be important to improve goat breeding and economic status of the farmers. Also, the present work constitutes a good example of a successful interaction between academic sector, governmental institutions and farmers.

The goal of the present investigation was to record the GI parasitism in goat farms from Garza Real locality in Zapotillo Municipality, Ecuador, and to evaluate the usefulness of FAMACHA© system on infected goats.

MATERIAL AND METHODS

Farms and animals

The locality of Garza Real (369-567 m.s.l.) was selected based on requirements of Agrocalidad related to the levels of mortality in goats. In the absence of data on GI parasitism of goats in Ecuador, results of a non-related qualitative analysis of feces of 128 goats from a neighboring locality of Limonas were used to establish sampling number (unpublished data). Based on a population of 3,109 goats in Garza Real locality, 86.3% of prevalence of GI parasites from Limonas locality and a confidence limit of 95%, sampling number was calculated in 30 animals (EpiInfo© 7.1.5, Center for Disease Control and Prevention, USA). To guarantee statistical precision, 77 cross-bred goats were sampled (75 females and 2 males, one to four years old) in seven farms with an extensive way of breeding. Only seven farmers gave their acquiescence for sampling.

Animals are allowed to feed in free lands early in the morning and return to the pens at twilight. Farmers informed about treatments with ivermectin 1% by subcutaneous route (1 mL-2 mL depending on the animal size) without veterinary assistance in those cases with notorious loss of body weight or apathy, regardless of the age or physiological state of the animal.

Parasitological analysis

Faecal samples were taken directly from the rectum into plastic bags. Samples were maintained into coolers with blue ice at field and after at 4 °C in laboratory until processing no more than three days later. Qualitative and quantitative parasitological exams were performed by flotation techniques following the standard procedures with sugar as flotation solution (density= 1.20). The identification of eggs was up to genus or order level. *Trichuris* spp., *Moniezia* spp. and *Eimeria* spp. infections were only evaluated by qualitative analysis. The individual faecal eggs counts (FECs) were performed using a modified McMaster method [7] and the eggs per gram (EPG) were calculated for parasites of order Strongylyda and *Strongyloides* spp. The infection levels by nematode infection were established as low (50-799); moderate (800-1,200) and high (>1,200) EPG [1].

Faecal samples collected in the farms were pooled and cultured at 27 °C, 70% relative humidity in an incubator (Memmert, model BE-200, Memmert GmbH + Co., Germany) for seven days [7]. After incubation, third stage larvae (L₃) were collected by the Baerman technique and identified according to Van Wyck *et al.* [27] and Roeber *et al.* [20]. The larval composition was obtained by counting and identifying 100 L₃ in each one of the seven cultures under microscope (Axiostar Plus, Zeiss, Germany).

Hematological analysis

Blood samples were individually collected by jugular venipuncture from each animal by using disposable Vacutainer© system into tubes with EDTA (ethylenediamine tetraacetic acid) (Becton Dickinson, USA). Packet cell volume (PCV) was measured on the blood samples by the microhaematocrit method (Haematocrit centrifuge model KHT-430B, Gemmy Industrial Corporation, Taiwan) and values were expressed as percentages. The FAMACHA© guide was used through the evaluation of the ocular mucosa membranes classification, performed by comparing with a laminated colour chart bearing pictures of goat classified into five categories ranging from the normal red, through pink to practically white mucous membrane in severe anaemic animals (1: red; 2: red-pink; 3: pink, mildly; 4: pink-white; 5: white) [19, 26].

Statistical analysis

Analysis of variance was used to establish statistical differences between faecal eggs counts in the farms. Level of significance was set at $P < 0.05$. PCV values and faecal egg counts were

also compared and correlated to the score assignment in the FAMACHA© system by a correlation analysis. In all the cases, Prism GraphPad v. 7.0 (GraphPad Software, Inc, USA) was used.

RESULTS AND DISCUSSION

From the total of 77 faecal samples, 91% were found to be infected with GI parasites. A mixed infection with two to four types of GI parasites was found in 87.82% of the animals (12.32, 32.87, 44% and 10.95% with one, two, three or four taxa, respectively). Based on morphology, eggs from order Strongylida (80.5%), genera *Strongyloides* (59.7%), *Trichostrongylus* (19.5%), and *Moniezia* (1.3%) were identified by flotation technique. Unsporulated oocysts of *Eimeria* were observed in 67.5% of the samples. The mean of FECs of order Strongylida and genus *Strongyloides* found in the farms are shown in TABLE I. There were not significant differences in FECs between the seven farms ($F 1.58$; $P > 0.05$). Considering the mean of type Strongylida eggs, four of the farms have a high infection level ($> 1,200$ EPG).

TABLE I
FAECAL EGGS COUNTS OF PARASITES FROM ORDER STRONGYLIDA AND GENUS *Strongyloides* IN GOAT SAMPLES FROM GARZA REAL LOCALITY

Farm (n)	Strongylida		<i>Strongyloides</i>		Level of infection ¹
	Mean \pm sd	Max - min	Mean \pm sd	Max-min	
1 (20)	1,175 \pm 2,334	10,500 - 0	585 \pm 1,958	8,850 - 0	Moderate
2 (6)	200 \pm 158	450 - 0	67 \pm 82	200 - 0	Low
3 (12)	1,750 \pm 2,173	5,950 - 0	213 \pm 377	1,150 - 0	High
4 (11)	1,505 \pm 2,467	7,400 - 50	336 \pm 519	1,600 - 0	High
5 (10)	905 \pm 1,657	5,200 - 0	72 \pm 182	550 - 0	Moderate
6 (10)	3,980 \pm 6,604	21,800 - 0	455 \pm 700	2,300 - 0	High
7(8)	3,906 \pm 5,152	15,750 - 0	313 \pm 699	2,000 - 0	High

(n) Number of animals sampled by farm

Max: maximum; min: minimum

sd: standard deviation

¹ Strongylida

FEC = Fecal egg counts (eggs per gram of faeces)

These results revealed the critical situation in the farms: a high prevalence of GI parasitism, mainly from order Strongylida, which cause a serious disease; additionally, the goats harbored up to four taxa simultaneously in practically every farm and the symptoms of GI parasitism are similar to a nutritional disease [9, 25]. Additionally, the presence of coccidia in almost all farms with an infection level up to 67.5% is remarkable. The precarious conditions of the pens (no sanitary measures, humidity and darkness) in Garza Real can lead to a clinical coccidiosis due to a massive ingestion of sporulated oocysts in an highly infected

environment and a significant asexual multiplication in goats due to a lowered resistance in animals because of prolonged period of alimentary stress [5, 16].

A number of 700 L₃ larvae were analyzed from coprocultures. Six genera, mainly from order Strongylida, were identified: *Haemonchus contortus* (43%), *Trichostrongylus* spp. (39%), *Oesophagostomum* spp. (6%), *Teladorsagia* spp. (1%) and *Cooperia* spp. (< 1%); *Strongyloides* larvae was observed in 11% of the cultures. *Strongyloides* larvae account for 51% of the observed larvae in culture from farm 5 (TABLE II). Based on the

results, *H. contortus* and *Trichostrongylus* spp. were the most important parasites from order Strongylida as have been reported under tropical conditions [3]. *H. contortus* is generally considered to be the most prevalent and pathogenic nematode species of small ruminants in tropical and subtropical areas [12, 17, 19]; is associated with anaemia and hypoproteinaemia due to blood-feeding activity. Two species of *Trichostrongylus* occur in small

ruminants: *T. axei*, living in the abomasum, does not appear to be an important pathogen, but *T. colubriformis* causes black scours and contributes to parasitic gastroenteritis, ill thrift and diarrhea in kids. *Strongyloides* also cause disturbances in animal health, quite frequently inducing the sudden death syndrome in young ruminants due to heart failure; the degree of the damage directly correlates with the intensity of the parasitic infection [23].

TABLE II
PARASITES FROM ORDER STRONGYLIDA OBSERVED IN THE COPROCULTURES AND PACKET CELL VOLUME (PCV) IN THE GOAT FARMS

Farm	Larvae in coprocultures						PCV (%) ¹		
	<i>H</i>	<i>T</i>	<i>Tc</i>	<i>O</i>	<i>C</i>	<i>S</i>	Mean ± sd	Max	Min
1	31	61	0	2	0	6	22 ± 6	34	10
2	55	27	8	8	0	2	24 ± 2	26	21
3	52	31	0	7	0	10	21 ± 5	27	12
4	48	50	0	1	0	1	22 ± 4	29	15
5	28	61	0	10	0	1	21 ± 5	27	13
6	29	13	1	5	1	51	17 ± 4	23	9
7	58	28	1	9	0	4	16 ± 5	23	8
Mean	43 ± 13	39 ± 19	1 ± 3	6 ± 3	0 ± 0.37	11 ± 18			

H: Haemonchus; T: Trichostrongylus; Tc: Teladorsagia; O: Oesophagostomun; C: Cooperia; S: Strongyloides

¹ Normal values: 22-38% [29].

Max: maximum value; Min: minimum value

sd: standard deviation

Mean of the PCV levels and the parasites found in the farms are also shown in TABLE II. Animals of four farms have PCV values lower than normal values [11]. Statistical differences were observed in the PCV values between farms (F 3.196; P< 0.05) and a strong and negative correlation of - 0.9176 (P< 0.001) was found between PCV and FECs.

To select animals in need for antihelmintic treatment, FAMACHA© system has been found to be a useful tool in many parts of the world [9, 14, 16, 19, 21, 28]. Results of the score

of FAMACHA©/percentage of goats in the field were 1 (5.66%); 2 (35.84%); 3 (30.18%); 4 (18.86%) and 5 (9.43%). A strong and negative correlation (-0.9418; P<0.001) was established between PCV values and score assignment in the FAMACHA© system (TABLE III): animals with the highest PCV values had been assigned score 1 and those with lowest PCV value had been assigned score 5. Also, a strong but positive correlation was found (0.9763; P< 0.005) between FECs and FAMACHA© scores (TABLE III), which shows the impact of the parasites in the hematological values.

TABLE III
PACKET CELL VOLUME (PCV) VALUES, FAECAL EGG COUNTS (FEC) OF PARASITES FROM ORDER STRONGYLIDA AND LEVEL OF INFECTION COMPARED WITH FAMACHA© SCORES IN SAMPLED GOATS

	FAMACHA© score (PCV values)				
	1 (>28 ¹)	2 (23-27)	3 (18-22)	4 (13-17)	5 (< 12)
PCV (%) ²	34 ± 3	25 ± 1	21 ± 3	15 ± 2	12 ± 0
FEC ³	0 ± 0	250 ± 0	2,076 ± 5,082	2,622 ± 4,003	3,525 ± 2,793
Level of infection	Low	Low	High	High	High

PCV and FECs are expressed as mean values ± standard deviation

¹PCV values assigned to FAMACHA© scores

²Correlation value $r = -0.9418$; $P < 0.001$

³Correlation value $r = 0.9763$; $P < 0.005$

The percentage of success on the interpretation of FAMACHA© was 65%, mainly due to the fact that score 3 was the most recorded incorrectly to the animals with higher PCV values. However, a high level of infection by GI parasites was found in those animals with an incorrect assignment of score 3, and thus, should be treated. Sotomaior *et al.* [22] reported that the sensitivity of the test increased when score 3 animals were included as being anemic; Van Wyk and Bath [26] recommended a routine treatment of every animal judged to be in FAMACHA© categories 3–5. The value of success in assignment scores was a reasonable one because it was the first time to apply the system in Ecuador. Vilela *et al.* [28] achieved a value of 32.6% in Brazil in the first of three months sampling. Some authors pointed out that applicability of the FAMACHA© method is limited because it requires a percentage of *H. contortus* in the herd greater than 60% and needs trained technicians to perform the readings [17]. Other studies have shown that animals in greater need of anthelmintic treatment can be identified and treated selectively [19]. However, with a mean of 43% of *H. contortus* in the Garza Real farms, it represents a good method to demonstrate the need for treatment and control the populations of Strongylida, mainly *H. contortus*, and improvable if there were trained farmers [14]. *Haemonchus* females are capable of producing up to 10,000 eggs per day which is a very high reproductive rate and results in rapid contamination of the environment [8]. Certainly, the method could have a disadvantage: if control by treatment is successful and the number of *H. contortus* diminishes but anaemia is due to malnutrition or another etiology, FAMACHA© method could not be used alone in the quest of an accurate diagnosis.

The high levels of parasitism and anemia found in the farms also suggest a lack of effective treatments and malnutrition due to the long-time drought. In Garza Real, as in many localities of the country, veterinary visits - limited to governmental organisms - may not be as frequent as desirable and there are no regulations about marketing veterinary drugs to the public, so the decision about treatments often relies on the farmers. This indiscriminate use of synthetic anthelmintics cause great economical losses

due to the lack of individual evaluations increases the selection pressure towards parasite resistance and leaves residues in meat, milk and in the environment [28]. Any combination of malnutrition and digestive parasitism will lead to uncompensated pathophysiological disturbances and, subsequently, to severe clinical signs (including a high mortality rate) [9]. Under low resource conditions, hosts adopt a tolerance strategy or are simply unable to mount an effective immune response; hence, resource supplementation could increase individual resistance/resilience and reduce parasite transmission [4, 24].

Shortage in the feeding lands by the canal could also help to concentrate the availability of eggs and larvae to infect the goats or even worst, change the feeding behavior of the animals. Due to browsing, goats may have a natural advantage over grazing sheep, but if the goats are forced to graze or are limited to feeding contaminated pastures may confront burdens of *H. contortus*– as other parasites [17]. Furthermore, evidence is now accumulating to indicate that, when infected with GI parasites, ruminants can modify either the amount or the type of the food they eat; this may ameliorate the negative impact of parasitic infections, and has been described as self-medication [29].

Summing up, the results from the present investigation suggest the need of a combined strategy of treatments, supplementary food and environment [13] under a well-coordinated and permanent sanitary monitoring over the goat farms of Garza Real locality and maybe over the whole Municipality.

CONCLUSIONS AND RECOMMENDATIONS

This is the first report of GI parasites in goats from Ecuador. The study revealed the high levels of GI parasitism mainly due to *H. contortus* and *Trichostrongylus* spp. parasites in the sampled farms. FAMACHA© system could be useful in animals highly infected by *Haemonchus*, but parasitological methods are also required to evaluate and apply anthelmintic treatments.

ACKNOWLEDGMENT

The authors are thankful to goat farmers from Garza Real locality, Ecuador, who kindly offered their support for the present investigation. To Agrocalidad Loja, Ecuador, for their invaluable technical assistance. To anonymous reviewers who helped to improve the manuscript.

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