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Original Article

Losses caused by gastrointestinal nematode infections in Dorper lambs under two nutritional status

Prejuízos causados pelas infecções por nematoides gastrintestinais em cordeiros Dorper submetidos a dois manejos nutricionais

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Abstract

The aim of this study was to evaluate the effect of two nutritional statuses on the productive performance of Dorper lambs naturally infected with gastrointestinal nematodes. Thirty-two lambs, grazing together on the same pasture, were allocated into four experimental groups: (G1) infected-supplemented diet, (G2) control-supplemented diet, (G3) infected-basal diet, and (G4) control-basal diet. Control animals received suppressive treatment with monepantel every two weeks, while precautionary anthelmintic treatments were given to all lambs of the infected groups with packed cell volume (PCV) <23%. There was reduction in the PCV means of all groups, which was more pronounced in the infected lambs that also presented reduction in total plasma protein values in comparison with the controls. Weight gain was affected by diet and infection status ($P < 0.05$). Daily body weight gain was 0.170 kg in the G1, 0.205 kg in the G2, 0.085 kg in the G3, and 0.116 kg in the G4. The cold carcass weight was 4.1% and 13.7% higher in controls in comparison with infected lambs, respectively, in the supplemented and basal diets. The infected groups, despite receiving precautionary anthelmintic treatments to prevent deaths due to haemonchosis, presented reduction in the production parameters in comparison with the controls.

Keywords: *Haemonchus*, nutrition, monepantel, *Trichostrongylus*, sheep.

Resumo

O experimento teve por objetivo determinar o efeito de dois níveis de nutrição no desempenho produtivo de cordeiros Dorper naturalmente infectados por nematoides gastrintestinais. Trinta e dois cordeiros, mantidos juntos na mesma pastagem, foram alocados em quatro grupos experimentais: (G1) infectado-suplementado, (G2) controle-suplementado, (G3) infectado-dieta basal e (G4) controle-dieta basal. Os cordeiros suplementados receberam diariamente concentrado em quantidade equivalente a 2% do peso corporal (PC), enquanto na dieta basal receberam apenas uma pequena quantidade de concentrado (0,35% do PC). Os animais controles receberam tratamento supressivo com anti-helmíntico a cada duas semanas e os infectados foram tratados individualmente quando apresentaram volume globular (VG) <23%. Houve redução nas médias de VG em todos os grupos, as quais foram mais pronunciadas nos animais dos grupos infectados, que também apresentaram redução nos valores de proteína plasmática total em comparação com os controles. Houve efeito significativo da dieta e da infecção no ganho de peso (P <0,05). O ganho em peso diário foi de 0,170 kg no G1, 0,205 kg no G2, 0,085 kg no G3 e 0,116 kg no G4. Os grupos infectados, apesar de receberem tratamentos anti-helmínticos preventivos que evitaram mortes por haemonchose, apresentaram redução nos parâmetros produtivos em comparação com os controles.

Palavras-chave: *Haemonchus*, nutrição, monepantel, *Trichostrongylus*, ovinos.

Introduction

The Dorper is a hardy South African composite nonwoolled, mutton breed of sheep. Its development in the early 1940s occurred because of the need for a relatively easy-care sheep breed with an acceptable meat carcass suitable for the production of slaughter lambs under the adverse conditions of the arid, extensive regions of South Africa (CLOETE et al., 2000; MILNE, 2000). Dorper was developed through the crossing of the Blackhead Persian ewes with the Dorset Horn rams. The Blackhead Persian has small litter sizes, slow growth rates, and poor mutton characteristics, but it was selected as the mother breed because of its outstanding

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performance, especially under harsh environmental conditions of the semi-desert extensive sheep production systems in South Africa (SCHOEMAN & BURGER, 1992). The Dorset Horn was selected because it demonstrated a longer breeding season in comparison with other British sheep breeds (MILNE, 2000).

In South Africa, both Dorper and Dorper cross-breeds are superior in reproductive and growth traits to woolled and other indigenous breeds. They reach a target slaughter weight of approximately 40 kg earlier than Merino, Afrino, Suffolk, and Ile de France crosses. The average daily gain to slaughter was 44% higher in the Dorper than the average of the groups it was compared to (SCHOEMAN, 2000). However, in comparison with the Red Maasai sheep, an indigenous breed of Kenya and Tanzania, Dorper are highly susceptible to *Haemonchus contortus* infection (MUGAMBI et al., 1996, 1997; WANYANGU et al., 1997). For this reason, although generally considered as being 'well-adapted', the Dorper proved to be unsuitable for commercial use in the African tropics (SCHOEMAN, 2000).

Despite this problem related to susceptibility to haemonchosis, Dorper sheep have been introduced into tropical areas of Brazil in recent years, where they have quickly become widespread. Cross-breeding of Dorper with local breeds, especially Santa Ines hair sheep, resulted in lambs with reasonably acceptable meat carcass and with a satisfactory degree of resistance against gastrointestinal nematodes (AMARANTE et al., 2009; ISSAKOWICZ et al., 2018). No need for shearing is another advantage of Dorper sheep in tropical environments. For these reasons, there is a high acceptance of Dorpers, which today has the highest number of registered stud sheep breeders in central and northeast areas of Brazil.

Gatrointestinal nematode (GIN) infection is one of the main health problems of the sheep industry, causing significant losses due to reduction in productivity and mortality of animals. *H. contortus* is the most important parasite in regions with tropical and subtropical climates. It is a blood-sucking parasite that causes, in acute infections, mortality, which is considered the most important economic effect attributable to this parasite (BARGER & COX, 1984). In addition, it presents high prolificacy and great capacity of developing anthelmintic resistance (ALBUQUERQUE et al., 2017; MARTINS et al., 2017; OLIVEIRA et al., 2017). Other trichostrongylid parasites produce significant reductions in weight gain, which is the case of *Trichostrongylus colubriformis*, considered the second most important nematode in sheep in Brazil (WILMSEN et al., 2014; ALMEIDA et al., 2018). *T. colubriformis* is a parasite of the small intestine that can cause inappetence and a disruption of the normal digestive process in lambs (KIMAMBO et al., 1988).

The indiscriminate use of anthelmintics for controlling GIN infections leads to the fast development of anthelmintic resistance (STARLING et al., 2017). For this reason, a combination of different control methods has been investigated to maximise animal productivity and reduce the use of antiparasitic drugs. The optimisation of host nutrition to increase resistance/resilience against GIN infections has received increasing attention (KNOX & STEEL, 1996; BRICARELLO et al., 2005; HOSTE et al., 2016). GIN has a negative effect on dry matter intake and live-weight change of lambs. However, it has been suggested that the metabolic cost of GIN infection can be covered by supplying an additional requirement for protein and energy in the diet of infected lambs (MÉNDEZ-ORTÍZ et al., 2019).

Experiments conducted under grazing conditions are more challenging than those carried out indoors under controlled conditions of infection and nutrition. In the field, the major difficulty encountered is in maintaining uninfected control groups as worm-free. It has been demonstrated that persistent challenge with *H. contortus* and *T. colubriformis* infective larvae does not affect the growth of grazing, meat-breed lambs when suppressively treated with effective anthelmintics, indicating that the use of suppressively treated sheep could be a valid substitute for worm-free lambs in field experiments (DEVER et al., 2015). However, the absence of anthelmintics with a high degree of efficacy has been a problem when designing studies that require comparisons between infected and noninfected control groups in the field. The recent launch of monepantel in Brazil gave us an opportunity to evaluate the losses caused by GIN in sheep using lambs suppressively treated with monepantel as a control group and substituting GIN-free lambs in a field experiment.

Because of the little information about the effects of nutrition and its interactions with gastrointestinal parasitism in grazing Dorper lambs in Brazil, this study was conduct to evaluate the performance of young weaned Dorpers in an environment with a high rate of nematode parasitic infection, as well to determine the effect of the supplementation in the outcome of such infections.

Materials and Methods

All procedures involving animals in this study were conducted in accordance with international ethical standards and were approved by the local Ethics Committee on Animal Use (protocol number 159/2015-CEUA, FMVZ/UNESP).

Experimental area and climatic measurements

The experiment was conducted at São Paulo State University (UNESP), Botucatu, Brazil. According to the Köppen classification system, the region's predominant climate is the Cwa type (humid subtropical), which is characterized by warm, rainy summers and dry winters (ALVARES et al., 2014).

The 32 experimental lambs grazed together in a pasture of *Cynodon dactylon* and *Urochloa decumbens* in an area with 4,092.6 m^2 divided into four fenced paddocks of similar sizes. Each paddock was grazed for 9 days, with a 27-day interval between grazing. Animals had free access to drinking water. After the 9 days of grazing, the pasture was fertilised with 20 kg of mineral fertilizer (NPK 20-05-20), corresponding to a quantity equivalent to 1000 kg per hectare/year. The pasture had been grazed by sheep before the trial. Therefore, it was contaminated with free living stages of gastrointestinal nematodes.

Pasture bromatological analyses were performed monthly according to the methodology described by Van Soest et al. (1991). The first analysis was performed on 10 October, before the beginning of the experiment (Table 1). Samples were collected at four sites using a metal square, each with an area of 0.25 m^2, as described by Piza et al. (2019).

Table 1. Bromatological analysis (% of dry matter) of the herbage (*Cynodon* spp. and *Brachiaria decumbens*) grazed by the experimental lambs during the trial.

Date	CP (%)	EE (%)	A (%)	NDF (%)	ADF (%)	Hemicellulose (%)
10/Oct/2015	7.28	1.82	11.15	68.46	31.84	36.62
27/Nov/2015	11.86	3.71	8.16	78.28	41.93	36.35
25/Dec/2015	13.57	4.01	8.88	76.54	39.27	37.27
29/Jan/2016	12.92	3.97	9.35	75.26	37.18	38.08
21/Feb/2016	13.19	2.89	9.64	77.99	35.97	42.02

CP - Crude protein, EE - Ethereal Extract, A – Ash content, NDF - Neutral Detergent Fiber, ADF - Acid Detergent Fiber.

Description of the experiment

Thirty-two Dorper lambs, 3 months old, with an initial body weight of 22.47 ± 1.57 kg, were acquired from a commercial farm where they had been raised indoors since birth. Lambs arrived in the experimental area of the university on 20 November 2015. On this occasion, faecal samples were collected from all animals for parasitological examinations. Among the 32 lambs, 17 were shedding eggs of strongyles with values ranging from 100 to 2700 eggs per gram of faeces (EPG). For this reason, all animals received treatment with monepantel (2.5 mg/kg; Zolvix®; Zoetis). No nematode eggs were found in the faecal examinations performed 14 days after treatment. All lambs received a clostridiosis vaccine (2 mL/animal, single dose; Ultrachoice 8; Zoetis®) and toltrazuril (15 mg/kg, Isocox®, Ourofino) to prevent coccidiosis.

The experiment used a completely randomized 2 × 2 factorial design (infected or control × supplemented or basal diet). The 32 animals were randomized to the four treatments, 8 animals for each one, on 10 November:

Group 1: infected-supplemented.

Group 2: control-supplemented. In this group, near the end of the trial, one lamb suffered a limb fracture. For this reason, its data were excluded, and this group had seven animals.

Group 3: infected-basal diet.

Group 4: control-basal diet.

Control lambs were treated every 2 weeks with monepantel (2.5 mg/kg; Zolvix®; Zoetis) to keep them as free as possible of parasitic infection. To prevent deaths, precautionary anthelmintic treatment with the same drug was administered to lambs of the infected groups wherever they presented packed cell volume (PCV) <23%.

Supplemented lambs were given a daily live weight–based allowance (2% of their live weight) of a high-quality food. The lambs under the basal diet received a small daily amount of concentrate (0.35% of their live weight) to supply them with minerals and also to provide the same daily management of the supplemented lambs (i.e., all lambs were kept in individual stalls for approximately 1 h in the afternoon, where they received the concentrate). Taking into consideration the first bromatological analysis of the pasture, the two concentrates were formulated in the software Small Ruminant Nutrition System (SRNS) based on the structure Cornell Net Carbohydrate and Protein System (2000) for sheep production to provide estimated gains of 225 g/day and 90 g/day for the supplemented and basal diet groups, respectively. Ingredients and nutritional composition of the concentrates supplied daily to lambs under basal or supplemented diets are presented in Table 2.

Table 2. Ingredients and nutritional composition of the concentrates supplied daily to lambs under the basal or supplemented diet.

	Basal Diet	Supplemented Diet
Ingredientes, % of dry matter (DM)		
Ground corn	87.39	78.84
Soybean meal	0.00	7.98
Peanut meal	0.00	7.98
Calcitic Limestone	0.00	1.33
Urea	0.00	0.74
Mineral[1]	12.61	3.14
Chemical composition, DM basis[2]		
Dry matter, %	89.50	88.61
Crude protein, %	8.10	18.00
Rumen degradable protein, % PB	91.90	74.51
Metabolizable protein, %	6.31	9.51
Metabolizable energy, Kcal/kg	2747	2884
Ethereal Extract, %	3.50	4.00
Calcium, %	2.70	1.00
Phosphor, %	0.90	0.50

[1]Mineral composition (kg of the product) 200 g Ca, 75 g P, 50 g Mg, 10 mg S, 24 mg Se, 3060 mg Zn, 1000 mg Mn, 2500 mg I, 20 mg Co.; [2]Values calculated by CNCPS – Sheep software.

By late afternoon, lambs were confined in individual stalls (3 m^2), with concrete floors and an individual water fountain and trough, where they were fed with the predetermined amount of concentrate. Every 7 days, the lambs were weighed individually, which allowed for adjustment of the amount of concentrate supplied to each of them. After they had finished the concentrate consumption, the lambs were conducted again to the paddock.

Lambs started to graze in the experimental paddocks on 10 November, when they started to receive a progressive increase in the amount of the concentrate over 2 weeks to adapt them to the diets. Daily body weight gain was calculated after this adaptation period, from 23 November 2015 to 29 February 2016, for a total of 98 days (14 weeks). Lambs were slaughtered on 1 March.

Quantification of infective larvae on pasture

To determine the number of third-stage larvae (L3) of nematodes per kilogram of dry matter (L3/kg DM), grass samples were collected, starting at 8 a.m., following a predetermined W-shaped tracing in each paddock (TAYLOR, 1939). The collector followed this route, manually collecting a sample of grass every four steps, in an approximate distance of 4 m. Samples were collected on the ground level using scissors and packed in plastic bags. The grass samples were processed according to Niezen et al. (1998), and the larvae were separated from the sediment, recovered, and quantified as previously described by Carneiro & Amarante (2008).

Faecal examination

Faecal samples were collected every 14 days from the rectum of each animal. Samples were individually processed using the modified McMaster technique with a sensitivity of 100 EPG (UENO & GONÇALVES, 1998). On the same collection days, composite faecal cultures were prepared for each group to obtain and differentiate third-stage larvae into parasitic genus (UENO & GONÇALVES, 1998).

Haematology

Blood samples (5 mL) were collected every 2 weeks by jugular vein puncture into Vacutainer® tubes containing anticoagulant (EDTA). The PCV was determined by microhematocrit centrifugation, and the total plasma protein (TPP) levels were estimated using a refractometer (Refractometer SPR-N, Atago).

Helminths recovering and counting

At the end of the experiment, the animals were slaughtered, and the gastrointestinal tract was collected and taken to the laboratory. Worm identification and counting procedures were performed on samples of the abomasums and intestine contents as described by Ueno & Gonçalves (1998).

Behaviour of lambs

The behaviour of the lambs was evaluated individually over an uninterrupted period of 24 hours beginning at 8 p.m. on 14 December according to the descriptions of Jamieson & Hodgson (1979). The animals were evaluated by two observers for a period of 4 consecutive hours. At the end of this period, the duo was replaced by another and involved a total of six people. The variables recorded were time of pasture intake, rumination time, idleness time, and water intake. The animals were confined in individual stalls at 4:00 p.m., when they received the concentrate, and at 5:00 p.m. they were relocated to the paddock where all animals grazed together.

Statistical analysis

Data were analysed by analysis of variance for the variables measured just once (body weight, carcass and behaviour variables) using the General Linear Models (GLM procedure) of the Statistical Analysis Systems, version 9.4 (SAS Institute, 2016). For the weekly measures (PCV and TPP), the repeated measures analysis of GLM procedure was used. Diet and infection status were the classes evaluated. Means were compared by Tukey's test at the 5% significance level, and only significant interactions at the 5% significance level are reported in the results. Data regarding EPG and parasite worm burden were analysed using nonparametric tests. Mann-Whitney test was used in the case of strongyle variables, because there were only two groups infected with those parasites, while the Kruskal-Wallis test was used in the variables related to *Strongyloides,* present in the four groups.

Results

The presence of infective larvae on pasture during the trial (Figure 1) demonstrated that lambs had been challenged continuously by GIN infections. *Haemonchus* was the predominant parasite in pasture with a maximum of 1939 L3/kg DM on 18 December (paddock 3). In fecal cultures, *Haemonchus* larvae displayed percentages between 54% and 100% in the basal diet group and from 67% to 99% in the supplemented group (Table 3). Larvae of *Trichostrongylus* had a maximum value of 356 L3/kg DM in the pasture of paddock 3 on 29 January (Figure 2) and the highest percentage in faecal culture (46%) in the basal diet group at the end of the study (Table 3). *Cooperia* spp. was recorded in only three coprocultures, with a maximum percentage of 7% (Table 3).

The lambs under suppressive treatment did not shed strongyle eggs in the faeces during the trial, whereas the infected groups presented a progressive increase in EPG (Table 3) associated with a progressive reduction in PCV and TPP values (Figure 2). To prevent deaths, all lambs in the basal diet group required precautionary treatment with anthelmintic: six lambs once and two lambs twice. In the supplemented diet group, five lambs required treatment: three lambs once and two lambs twice. In this group, three lambs were able to cope with the parasitism without anthelmintic treatment: all three showed the highest faecal egg count (FEC) on 4 January (6,300 EPG, 15,500 EPG, and 5,700 EPG for lambs 1, 7, and 8, respectively) and no eggs in the last sampling in February (lambs 1 and 7) or 200 EPG (lamb 8). Lambs that required treatment showed FEC values between 6,200 EPG and 25,000 EPG and PCV values between 11% and 22%. Two weeks post the 17 doses of precautionary anthelmintic treatments given to lambs of the infected groups, no strongyle eggs were detected in the faecal examination of 10 lambs, while the other seven lambs remained shedding eggs. Infected lambs showed an increase in PCV two weeks posttreatment, with values between 26% and 35%.

In general, EPG counts had similar trends in both infected groups. The highest values were recorded on 21 December followed by a progressive decrease in FECs. There was a significant difference ($P < 0.05$) only in the last sampling, when the supplemented group presented a lower median FEC (100 EPG) in comparison with the basal diet group (250 EPG) (Table 3).

There was reduction in the PCV means of all groups (Figure 3), which was more pronounced in the infected groups, with a significant difference ($P < 0.05$) in relation to the controls on 21 December, 18 January, and 1 February. The lowest mean was recorded in the supplemented-infected group (PCV = 23.4%) on 21 December. Infected animals showed a reduction in TPP values from the third sampling until the end of trial in comparison with the controls ($P < 0.05$). The diet had no effect on PCV values

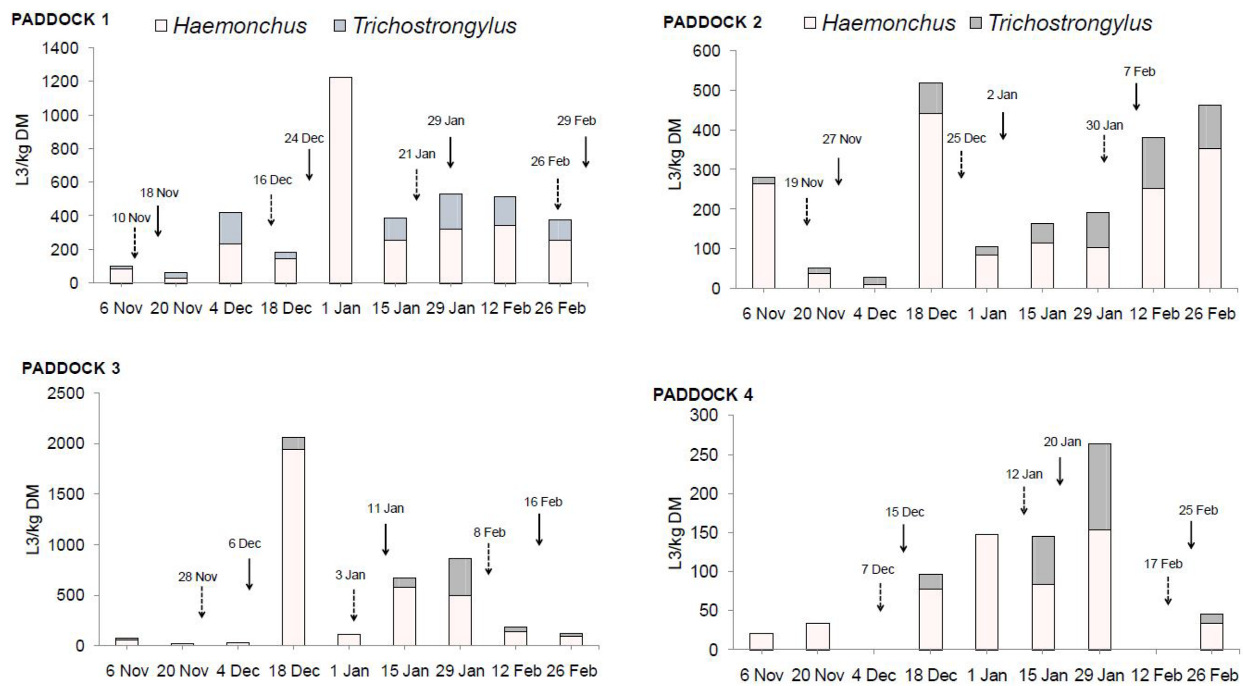

Figure 1. *Haemonchus* spp. and *Trichostrongylus* spp. infective larvae by kilogram of herbage dry matter (L3/kg DM) recovered from each of the four paddocks rotationally grazed by the experimental lambs. Dashed arrows indicate the beginning of the grazing in each paddock and the solid arrow, the exit.

Table 3. Number of lambs of the infected groups, fed with a basal or a supplemented diet, that received precautionary anthelmintic treatments (PAT) during the trial due to PCV < 23%. Median (minimum – maximum values) of eggs per gram of faeces (EPG) and third-stage larvae (%) of *Haemonchus*, *Trichostrongylus* and *Cooperia* in faecal cultures.

Date	Infected Groups	PAT	EPG	*Haemonchus*	*Trichostrongylus*	*Cooperia*
23 Nov	Supplemented	-	0 (0-0)	-	-	-
	Basal Diet	-	0 (0-0)	-	-	-
7 Dec	Supplemented	0	950 (0-5900)	97	3	0
	Basal Diet	1	200 (0-7500)	100	0	0
21 Dec	Supplemented	3	9700 (1800-16900)	99	1	0
	Basal Diet	4	3300 (0-33500)	100	0	0
4 Jan	Supplemented	2	6150 (0-25000)	90	3	7
	Basal Diet	1	250 (0-23400)	99	1	0
18 Jan	Supplemented	0	1150 (0-16100)	99	1	0
	Basal Diet	3	1250 (0-9600)	95	5	0
1 Feb	Supplemented	1	250 (0-10100)	89	5	6
	Basal Diet	1	0 (0-7000)	97	1	2
15 Feb	Supplemented	1	0 (0-11600)	99	1	0
	Basal Diet	0	0 (0-1600)	100	0	0
29 Feb	Supplemented	0	100* (0-300)	67	33	0
	Basal Diet	0	250 (200-900)	54	46	0

*There was significant difference (P<0.05) between group medians only in the last sampling by the Mann Whitney test.

(P > 0.05) and an effect on TPP only at the first sampling, when the supplemented animals showed an overall average lower than those of the basal diet (Figure 2).

Only a few *Haemonchus* worms were found in lambs of the control groups, with a maximum of 21 specimens in one lamb of the control-basal diet group. For this reason, Table 4 presents data only of the infected groups. The reduction in the degree of infection, observed at the end of the trial, based on FEC, was confirmed at slaughter, when lambs displayed relatively low worm counts (Table 4). Among species, *H. contortus* presented the highest infection intensity, with predominance of immature worms (Table 4). The worm-counting values were higher in the basal diet compared with the supplemented group (Table 4), with a significant difference in numbers of early L5, adult females, and total *Haemonchus* worm

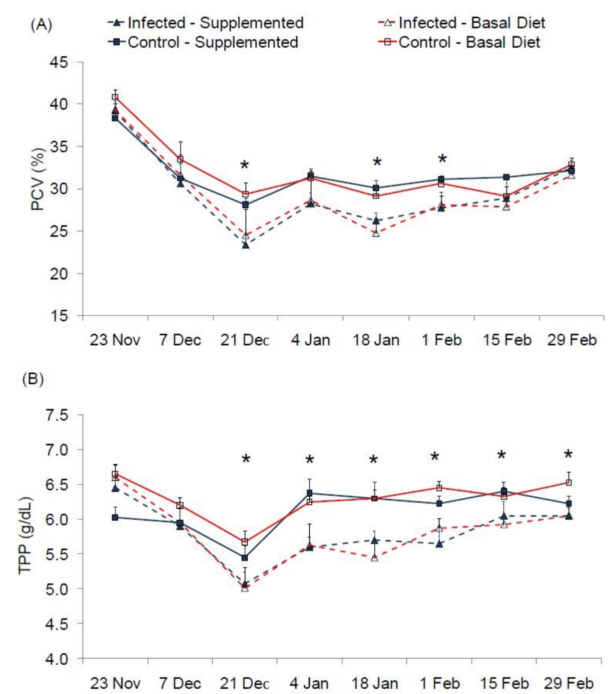

Figure 2. (A) Mean values of packed cell volume (PCV) and (B) total plasma protein (TPP) of the infected and control lambs fed with a supplemented or a basal diet. Bars: standard error. *Indicates moments with significant infection status effect (P < 0.05).

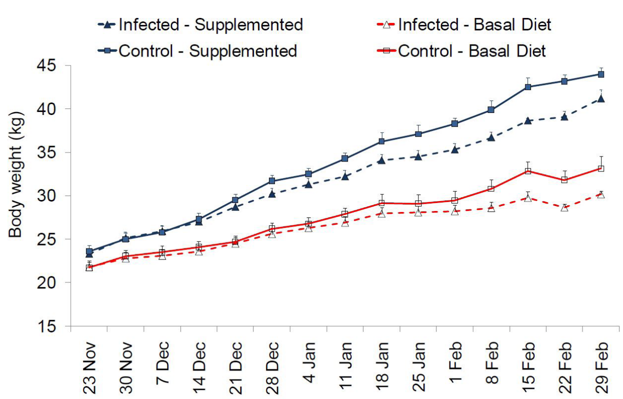

Figure 3. Mean body weight of the infected and control lambs fed with a supplemented or a basal diet. Bars: standard error.

burden ($P < 0.05$). *T. colubriformis* and *Cooperia curticei* presented similar low infection intensity in both infected groups (Table 4).

Because the anthelmintic used (monepantel) did not present efficacy against *Strongyloides papillosus*, lambs of all groups presented infection by this parasite, beginning to shed *Strongyloides* eggs on 4 January. The highest FEC medians were recorded in the last sampling: 700 (0-1,600), 350 (0-1,900), 200 (100-600), and 300 (0-1,000) EPG in the infected-basal, control-infected, infected-supplemented, and control-supplemented diet groups, respectively. *S. papillosus* worm burden was similar in all groups ($P > 0.05$; Kruskal-Wallis test), with a median of 745 (330-1870) worms in the infected-supplemented group, 780 (150-1082) in the control-supplemented group, 700 (70-1870) in the infected-basal diet group, and 402 (87-1602) in the control-basal diet group.

All groups of lambs presented weight gain throughout the trial, which was affected by diet and infection status. The highest body weight gains were recorded in groups fed a supplemented diet, including the 2 weeks of the diet adaptation period. For this reason, they were significantly ($P < 0.05$) heavier at the beginning of the data collection on 23 November (Table 5). The difference between the growth of the lambs under the supplemented and basal diets increased with the progression of the trial (Figure 3), with a significant Time × Diet interaction ($P < 0.0001$) with regard to body weight. Similarly, there was a significant Time × Infection interaction ($P < 0.0001$), and the difference between the control and infected groups increased with the progression of the trial. There was a statistically significant diet effect and infection status effect on daily body weight gain, final body weight, and carcass weight (Table 5). The daily body weight gain was 0.170 kg (±0.009) in the infected-supplemented group, 0.205 kg (±0.007) in the control-supplemented group, 0.085 (±0.008) in the infected-basal diet group, and 0.116 kg (±0.010) in the control-basal diet group. The cold carcass weight was 4.1% and 13.7% higher in controls in comparison with infected lambs, respectively, in the supplemented and basal diets. Carcass yield was higher in supplemented animals ($P < 0.05$), without an influence of the infection status (Table 5). The percentage loss from cooling was similar ($P > 0.05$) regardless of diet and infection status (Table 5).

The supplemented lambs grazed for less time and spent more time in idleness than animals from the basal diet group ($P < 0.01$). The lambs of the infected-supplemented, control-supplemented, infected-basal, and control-basal groups spent, on average, 27.9%, 29.3%, 36.5%, and 35.7%, respectively, of the time grazing.

Table 4. *Haemonchus contortus*, *Trichostrongylus colubriformis*, and *Cooperia curticei* worm burden (medians followed by minimum and maximum values in parentheses) in lambs of the infected groups fed with a basal or a supplemented diet.

Species	Stage of development	Supplemented Diet	Basal Diet	P-value#
H. contortus	Early L4	20 (0-140)	55 (0-440)	0.457
	Late L4	105 (0-1500)	275 (30-570)	0.156
	Early L5	30 (0-1010)	170 (50-530)	0.024
	Adult males	25 (0-50)	60 (10-350)	0.091
	Adult females	10 (0-170)	95 (20-470)	0.020
	Total burden	270 (0-2710)	705 (270-1790)	0.038
T. colubriformis	Total burden	165 (0-260)	220 (20-390)	0.528
C. curticei	Total burden	0 (0-20)	10 (0-30)	0.418

#Mann Whitney test at the 5% significance level; L4: fourth stage larvae; L5: fifth stage larvae in early development.

Table 5. Means (± standard error) of the variables related to the performance of the infected and control lambs fed with a supplemented or a basal diet.

Variable	Supplemented Diet		Basal Diet		Effects (P-value)	
	Infected (n=8)	Control (n=7)	Infected (n=8)	Control (n=8)	Diet	Infection status
Body weight* on 23/Nov	23.3^{a} ±0.55	23.9 ^{a} ±0.83	21.8^{b} ±0.70	21.7^{b} ±0.75	P< 0.02	P=0.85
Body weight on 29/Fev	39.9^{a} ± 0.99	44.0^{a} ± 0.88	30.2^{b} ± 0.55	33.1^{b} ±1.36	P<0.01	P<0.01
Daily weight gain	0.170^{a} ± 0.01	0.205^{a} ± 0.01	0.085^{b} ± 0.01	0.116^{b} ± 0.01	P< 0.01	P<0.01
Body weight after fasting (1/Mar)	38.2^{a} ± 0.94	40.9^{a} ± 2.37	28.4^{b} ± 0.48	31.7^{b} ± 1.38	P< 0.01	P<0.02
Warm carcass weight	18.8^{a} ± 0.49	19.6^{a} ± 0.56	12.8^{b} ± 0.45	14.8^{b} ± 0.81	P< 0.01	P<0.01
Cold carcass weight	18.5^{a} ± 0.49	19.3^{a} ± 0.56	12.6^{b} ± 0.46	14.6^{b} ± 0.81	P< 0.01	P<0.01
Loss of carcass weight on cooling (%)	1.40^{a} ± 0.36	1.39^{a} ± 0.44	1.40^{a} ± 0.31	1.70^{a} ± 0.39	P=0.29	P=0.28
Warm carcass yield (%)	49.3^{ab} ± 0.57	50.1^{a} ± 2.15	44.9^{b} ± 1.07	46.7^{ab} ± 0.57	P<0.01	P=0.22
Cold carcass yield (%)	48.6^{a} ± 0.62	49.4^{a} ± 2.01	44.3^{b} ± 1.10	45.9^{b} ± 0.60	P<0.01	P=0.27

*Weight in kg; In each row, results with different letters are significantly different by Tukey's test (P<0.05); There was no significant Diet × Infection status interaction (P > 0.05).

Discussion

The present experiment was carried out during the rainy season, when frequent rainfall and average temperatures of about 23°C boost abundant fodder production, allowing lambs to be kept under a relatively high stocking rate (2.52 animal unit/ha at the end of the trial). Such conditions also created a favourable environment for occurrence of massive infections by gastrointestinal nematodes, demonstrated in the present study, with several lambs of the infected groups presenting high FEC associated with reduced PCV values. This reveals that the blood loss caused by *H. contortus* infection was considerable, with a critical period of haemonchosis from 7 December until 18 January, when most of the precautionary treatments were given (8 drenches in December and 6 in January). Without these treatments, certainly, most of the lambs would have died before the end of the trial. Following this critical period, at the end of the trial in February, most of the lambs displayed low FEC, indicating the development of immune response. Despite a slight reduction in PCV and TPP means in the first month of the trial, the control groups presented these variables within the normal range in sheep.

The precautionary anthelmintic treatments prevented deaths due to haemonchosis; however, they did not avoid reduction in the production of the infected lambs in comparison with the controls. Although clinical symptoms of trichostrongylosis, such as diarrhea, were not seem at any time, *T. colubriformis* may also have contributed to the reduction in the body weight gain of the infected lambs, because it is known that this species interferes in the performance of animals, even in subclinical infections (VAN HOUTERT et al., 1995; CARDIA et al., 2011; SILVA et al., 2019). The lambs of all groups were infected by *S. papillosus*, because monepantel has no claim of efficacy against this parasite. In any case, infections by *S. papillosus* were mild and apparently had no negative impact on animal performance.

In the supplemented diet, control lambs showed a daily body weight gain 17.1% higher than the infected groups, and in the basal diet, the difference was 26.7%. Similarly, the cold carcass weight was 4.1% and 13.7% higher in controls in comparison with infected lambs, respectively, in the supplemented and basal diets. These results are substantiated by findings of several studies involving sheep that reported significant reductions in weight gain and carcass weight due to GIN infections (JACOBSON et al., 2009; SUTHERLAND et al., 2010; MILLER et al., 2012; MAVROT et al., 2015).

Despite differences in management, environmental conditions, and nematode species, our results regarding the production losses were similar to those reported in New Zealand, where the production costs of anthelmintic resistance were evaluated in sheep managed within a monthly preventive drench program to prevent losses caused by *Teladorsagia circumcincta* and *Trichostrongylus* spp. The presence of anthelmintic resistance resulted in a 14% reduction in the carcass value of lambs treated with albendazole (poor efficacy) in comparison with those treated with monepantel (high efficacy) (SUTHERLAND et al., 2010). In another study, lambs under a treatment with a drench with high efficacy (derquantel and abamectin) showed a 4.7-kg increase in carcass weight, with a 10.4% increase in carcass value, and they required less time (by 17 days) to reach a target live weight of 38 kg than those lambs treated with albendazole, an anthelmintic with poor efficacy due to anthelmintic resistance (MILLER et al., 2012).

Of the 17 precautionary drenches, 10 were given to lambs on the basal diet and 7 to supplemented lambs, which explains the similar means of EPG of the infected-basal diet and infected-supplemented groups. In this last group, three lambs performed well without the need for anthelmintic treatment. Despite the individual variation, it was possible to observe that in general, the well-nourished animals presented higher capacity to withstand the deleterious effects of parasitism, in agreement with other studies (CARVALHO et al., 2015; KHAN et al., 2017; SALGADO et al., 2017). It is necessary also to emphasize that because lambs under the basal diet received only a small quantity of concentrate, they grazed on average 1.7 hours more than the supplemented lambs. Consuming more herbage, they possibly ingested a larger number of infective larvae than the supplemented lambs. Therefore, by providing a better-quality diet associated with less exposure to infections, the concentrate supplementation favoured a better performance of the Dorper lambs, which reached a body weight of about 40 kg, considered the ideal body-weight target for this breed of sheep

(SCHOEMAN, 2000). Similarly, improvement in the productive performance and reduction in the degree of infection have been reported in lambs of other breeds supplemented with concentrate in other regions of Brazil (TONTINI et al., 2015; MELO et al., 2017; SALGADO et al., 2017).

Precautionary treatment given to lambs with PCV values lower than 23% was an approach similar to that recommended to prevent haemonchosis in the FAMACHA® method, in which the clinical category 3 corresponds to PCV = 18% to 22%. Similar to our results, in South Africa, Van Wyk (2008) observed productive losses in target selectively treated (TST)–sheep, based on the FAMACHA® method, when compared with suppressive-treated sheep. More pronounced differences were reported in Louisiana (USA), where lambs under TST treatment using FAMACHA had a daily body weight gain of 87.9 g/day, while a group under traditional anthelmintic use (four treatments with levamisole) had a gain of 113.4 g/day over 16 weeks of the trial (MILLER et al., 2011).

According to Cloete et al. (2000), in South Africa, Dorper lambs had postweaning gains in excess of 0.18 to 0.20 kg per day when weaned early at 2 to 3 months of age; at slaughter, Dorper lambs had dressing percentages of approximately 50%, and premium prices are paid for the highest-grade lamb carcasses weighing between 18 and 22 kg. The supplemented lambs reached similar records in the present study, supporting the notion that the Dorper is an adaptable sheep breed, capable of maintaining acceptable levels of production grazing under tropical conditions. However, it is important to emphasize that these values were obtained in lambs properly supplemented and that to prevent deaths, treatment with an anthelmintic showing efficacy against *Haemonchus* and *Trichostrongylus* was essential, especially during the initial months of grazing.

In conclusion, infected groups of Dorper lambs under a basal- or a supplemented-diet, despite receiving precautionary anthelmintic treatments to prevent deaths due to haemonchosis, presented reduction in the production parameters in comparison with the controls.

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References

Albuquerque ACA, Bassetto CC, Almeida FA, Amarante AFT. Development of *Haemonchus contortus* resistance in sheep under suppressive or targeted selective treatment with monepantel. *Vet Parasitol* 2017; 246: 112-117. http://dx.doi.org/10.1016/j.vetpar.2017.09.010. PMid:28969773.

Almeida FA, Bassetto CC, Amarante MRV, Albuquerque ACA, Starling RZS, Amarante AFT. Helminth infections and hybridization between *Haemonchus contortus* and *Haemonchus placei* in sheep from Santana do Livramento, Brazil. *Rev Bras Parasitol Vet* 2018; 27(3): 280-288. http://dx.doi.org/10.1590/s1984-296120180044. PMid:30133591.

Alvares CA, Stape JL, Sentelhas PC, Gonçalves JLM, Sparovek G. Köppen's climate classification map for Brazil. *Meteorol Z (Berl)* 2014; 22(6): 711-728. http://dx.doi.org/10.1127/0941-2948/2013/0507.

Amarante AFT, Susin I, Rocha RA, Silva MB, Mendes CQ, Pires AV. Resistance of Santa Ines and crossbred ewes to naturally acquired gastrointestinal nematode infections. *Vet Parasitol* 2009; 165(3-4): 273-280. http://dx.doi.org/10.1016/j.vetpar.2009.07.009. PMid:19656629.

Barger IA, Cox HW. Wool production of sheep chronically infected with *Haemonchus contortus. Vet Parasitol* 1984; 15(2): 169-175. http://dx.doi.org/10.1016/0304-4017(84)90033-5. PMid:6541834.

Bricarello PA, Amarante AFT, Rocha RA, Cabral-Filho SL, Huntley JF, Houdijk JG, et al. Influence of dietary protein supply on resistance to experimental infections with *Haemonchus contortus* in Ile de France and Santa Ines lambs. *Vet Parasitol* 2005; 134(1-2): 99-109. http://dx.doi.org/10.1016/j.vetpar.2005.05.068. PMid:16098676.

Cardia DFF, Rocha-Oliveira RA, Tsunemi MH, Amarante AFT. Immune response and performance of growing Santa Ines lambs to artificial *Trichostrongylus colubriformis* infections. *Vet Parasitol* 2011; 182(2-4): 248-258. http://dx.doi.org/10.1016/j.vetpar.2011.05.017. PMid:21641720.

Carneiro RD, Amarante AFT. Seasonal effect of three pasture plants species on the free-living stages of *Haemonchus contortus. Arq Bras Med Vet Zootec* 2008; 60(4): 864-872. http://dx.doi.org/10.1590/S0102-09352008000400014.

Carvalho N, Neves JH, Nazato C, Louvandini H, Amarante AFT. The effects of Diet and corticosteroid-induced immune suppression during infection by *Haemonchus contortus* in lambs. *Vet Parasitol* 2015; 214(3-4): 289-294. http://dx.doi.org/10.1016/j.vetpar.2015.09.012. PMid:26391820.

Cloete SWP, Snyman MA, Herselman MJ. Productive performance of Dorper sheep. *Small Rumin Res* 2000; 36(2): 119-135. http://dx.doi.org/10.1016/S0921-4488(99)00156-X. PMid:10760448.

Dever ML, Kahn LP, Doyle EK. Persistent challenge with *Trichostrongylus colubriformis* and *Haemonchus contortus* larvae does not affect growth of meat-breed lambs suppressively treated with anthelmintics when grazing. *Vet Parasitol* 2015; 209(1-2): 76-83. http://dx.doi.org/10.1016/j.vetpar.2015.02.009. PMid:25725548.

Hoste H, Torres-Acosta JFJ, Quijada J, Chan-Perez I, Dakheel MM, Kommuru DS, et al. Chapter seven - Interactions between nutrition and infections with *Haemonchus contortus* and related gastrointestinal nematodes in small ruminants. *Adv Parasitol* 2016; 93: 239-351. http://dx.doi.org/10.1016/bs.apar.2016.02.025. PMid:27238007.

Issakowicz J, Issakowicz ACKS, Bueno MS, Costa RLD, Geraldo AT, Abdalla AL, et al. Crossbreeding locally adapted hair sheep to improve productivity and meat quality. *Sci Agric* 2018; 75(4): 288-295. http://dx.doi.org/10.1590/1678-992x-2016-0505.

Jacobson C, Pluske J, Besier RB, Bell K, Pethick D. Associations between nematode larval challenge and gastrointestinal tract size that affect carcass productivity in sheep. *Vet Parasitol* 2009; 161(3-4): 248-254. http://dx.doi.org/10.1016/j.vetpar.2009.01.008. PMid:19217210.

Jamieson WS, Hodgson J. The effects of variation in sward characteristics upon the ingestive behaviour and herbage intake of calves and lambs under a continuous stocking management. *Grass Forage Sci* 1979; 34(4): 273-282. http://dx.doi.org/10.1111/j.1365-2494.1979.tb01479.x.

Khan FA, Sahoo A, Karim SA. Moderate and high levels of dietary protein on clinico-biochemical and production responses of lambs to repeated

Haemonchus contortus infection. *Small Rumin Res* 2017; 150: 52-59. http://dx.doi.org/10.1016/j.smallrumres.2017.03.009.

Kimambo AE, MacRae JC, Walker A, Watt CF, Coop RL. Effect of prolonged subclinical infection with *Trichostrongylus colubriformis* on the performance and nitrogen metabolism of growing lambs. *Vet Parasitol* 1988; 28(3): 191-203. http://dx.doi.org/10.1016/0304-4017(88)90107-0. PMid:3388740.

Knox M, Steel J. Nutritional enhancement of parasite control in small ruminant production systems in developing countries of south-east Asia and the Pacific. *Int J Parasitol* 1996; 26(8-9): 963-970. http://dx.doi.org/10.1016/S0020-7519(96)80072-5. PMid:8923143.

Martins AC, Bergamasco PLF, Felippelli G, Tebaldi JH, Moraes MFD, Testi AJP, et al. *Haemonchus contortus* resistance to monepantel in sheep: fecal egg count reduction tests and randomized controlled trials. *Semina: Ciênc Agrár* 2017; 38(1): 231-238. http://dx.doi.org/10.5433/1679-0359.2017v38n1p231.

Mavrot F, Hertzberg H, Torgerson P. Effect of gastro-intestinal nematode infection on sheep performance: a systematic review and meta-analysis. *Parasit Vectors* 2015; 8(1): 557. http://dx.doi.org/10.1186/s13071-015-1164-z. PMid:26496893.

Melo GKA, Ítavo CCBF, Monteiro KLS, Silva JA, Silva PCG, Ítavo LCV, et al. Effect of creep-fed supplement on the susceptibility of pasture-grazed suckling lambs to gastrointestinal helminths. *Vet Parasitol* 2017; 239: 26-30. http://dx.doi.org/10.1016/j.vetpar.2017.04.017. PMid:28495192.

Méndez-Ortíz FA, Sandoval-Castro CA, Vargas-Magaña JJ, Sarmiento-Franco L, Torres-Acosta JFJ, Ventura-Cordero J. Impact of gastrointestinal parasitism on dry matter intake and live weight gain of lambs: A meta-analysis to estimate the metabolic cost of gastrointestinal nematodes. *Vet Parasitol* 2019; 265: 1-6. http://dx.doi.org/10.1016/j.vetpar.2018.11.008. PMid:30638514.

Miller CM, Waghorn TS, Leathwick DM, Candy PM, Oliver AMB, Watson TG. The production cost of anthelmintic resistance in lambs. *Vet Parasitol* 2012; 186(3-4): 376-381. http://dx.doi.org/10.1016/j.vetpar.2011.11.063. PMid:22178431.

Miller JE, Burke JM, Terrill TH, Kearney MT. A comparison of two integrated approaches of controlling nematode parasites in small ruminants. *Vet Parasitol* 2011; 178(3-4): 300-310. http://dx.doi.org/10.1016/j.vetpar.2011.01.003. PMid:21333450.

Milne C. The history of the Dorper sheep. *Small Rumin Res* 2000; 36(2): 99-102. http://dx.doi.org/10.1016/S0921-4488(99)00154-6. PMid:10760446.

Mugambi JM, Bain RK, Wanyangu SW, Ihiga MA, Duncan JL, Murray M, et al. Resistance of four sheep breeds to natural and subsequent artificial *Haemonchus contortus* infection. *Vet Parasitol* 1997; 69(3-4): 265-273. http://dx.doi.org/10.1016/S0304-4017(96)01128-4. PMid:9195736.

Mugambi JM, Wanyangu SW, Bain RK, Owango MO, Duncan JL, Stear MJ. Response of Dorper and Red Maasai lambs to trickle *Haemonchus contortus* infections. *Res Vet Sci* 1996; 61(3): 218-221. http://dx.doi.org/10.1016/S0034-5288(96)90066-1. PMid:8938850.

Niezen JH, Charleston WAG, Hodgson J, Miller CM, Waghorn TS, Robertson HA. Effect of plant species on the larvae of gastrointestinal nematodes which parasitise sheep. *Int J Parasitol* 1998; 28(5): 791-803. http://dx.doi.org/10.1016/S0020-7519(98)00019-8. PMid:9650060.

Oliveira PA, Riet-Correa B, Estima-Silva P, Coelho ACB, Santos BL, Costa MAP, et al. Multiple anthelmintic resistance in Southern Brazil sheep flocks. *Rev Bras Parasitol Vet* 2017; 26(4): 427-432. http://dx.doi.org/10.1590/s1984-29612017058. PMid:29069158.

Piza MLST, Almeida FA, Pariz CM, Costa C, Amarante AFT. Comparison of two methods for the quantification of gastrointestinal nematode infective larvae from pasture. *Semina Ciênc Agr* 2019; 40(2): 713-722. http://dx.doi.org/10.5433/1679-0359.2019v40n2p713.

Salgado JA, Molento MB, Sotomaior CS, Dias LT, Castro LLD, Faisca LD, et al. Endoparasite and nutritional status of Suffolk lambs in seven production systems. *Anim Prod Sci* 2017; 58(9): 1667-1676. http://dx.doi.org/10.1071/AN16437.

SAS Institute. *SAS User's Guide. Version 9.4* [online]. Cary, NC: SAS Institute Inc.; 2016 [cited 2019 June 4]. Available from: https://support.sas.com/

Schoeman SJ, Burger R. Performance of Dorper sheep under an accelerated lambing system. *Small Rumin Res* 1992; 9(3): 265-281. http://dx.doi.org/10.1016/0921-4488(92)90156-X.

Schoeman SJ. A comparative assessment of Dorper sheep in different production environments and systems. *Small Rumin Res* 2000; 36(2): 137-146. http://dx.doi.org/10.1016/S0921-4488(99)00157-1. PMid:10760449.

Silva TPD, Bompadre TFV, Danasekaran DK, Sakita GZ, Abdalla AL Fo, et al. *Trichostrongylus colubriformis* infection: impact on digesta passage rate and lamb performance. *Vet Parasitol* 2019; 272: 17-22. http://dx.doi.org/10.1016/j.vetpar.2019.06.018. PMid:31395200.

Starling RZC, Martins IV, Alves CS, Viana MVG, Dietrich WS. *In vivo* diagnostic sensitivity of nematodes to different anthelmintics in sheep reared in semi-intensive system. *Arch Vet Sci* 2017; 22(2): 38-47.

Sutherland IA, Shaw J, Shaw RJ. The production costs of anthelmintic resistance in sheep managed within a monthly preventive drench program. *Vet Parasitol* 2010; 171(3-4): 300-304. http://dx.doi.org/10.1016/j.vetpar.2010.03.035. PMid:20434844.

Taylor EL. Technique for the estimation of pasture infestation by strongyloid larvae. *Parasitology* 1939; 31(4): 473-478. http://dx.doi.org/10.1017/S0031182000013007.

Tontini JF, Poli CH, Bremm C, Castro JM, Fajardo NM, Sarout BN, et al. Distribution of infective gastrointestinal helminth larvae in tropical erect grass under different feeding systems for lambs. *Trop Anim Health Prod* 2015; 47(6): 1145-1152. http://dx.doi.org/10.1007/s11250-015-0841-4. PMid:26003429.

Ueno H, Gonçalves PC. *Manual para diagnóstico das helmintoses de ruminantes*. 4ª ed. Tokyo: Japan International Cooperation Agency; 1998.

van Houtert MFJ, Barger IA, Steel JW, Windon RG, Emery DL. Effects of dietary protein intake on responses of young sheep to infection with *Trichostrongylus colubriformis. Vet Parasitol* 1995; 56(1-3): 163-180. http://dx.doi.org/10.1016/0304-4017(94)00668-3. PMid:7732640.

van Soest PJ, Robertson JB, Lewis BA. Methods for dietary fiber, neutral detergent fiber and nonstarch polysaccharides in relation to animal nutrition. *J Dairy Sci* 1991; 74(10): 3583-3597. http://dx.doi.org/10.3168/jds.S0022-0302(91)78551-2. PMid:1660498.

van Wyk JA. Production trials involving use of the FAMACHA® system for haemonchosis in sheep: preliminary results. *Onderstepoort J Vet Res* 2008; 75(4): 331-345. http://dx.doi.org/10.4102/ojvr.v75i4.109. PMid:19294989.

Wanyangu SW, Mugambi JM, Bain RK, Duncan JL, Murray M, Stear MJ. Response to artificial and subsequent natural infection with *Haemonchus contortus* in Red Maasai and Dorper ewes. *Vet Parasitol* 1997; 69(3-4): 275-282. http://dx.doi.org/10.1016/S0304-4017(96)01129-6. PMid:9195737.

Wilmsen MO, Silva BF, Bassetto CC, Amarante AFT. Gastrointestinal nematode infections in sheep raised in Botucatu, state of São Paulo, Brazil. *Rev Bras Parasitol Vet* 2014; 23(3): 348-354. http://dx.doi.org/10.1590/S1984-29612014058. PMid:25271455.