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Original Article

The prevalence of Strongylida/strongyles in small ruminants in Manatuto Municipality in central region of Timor-Leste

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Abstrak

Tujuan: Tujuan dari penelitian ini untuk mengetahui tingkat prevalensi cacing Strongylida/Strongyles pada ternak ruminansia kecil (domba dan kambing) di wilayah sentral Timor-Leste.
Metode: Metode yang digunakan dalam penelitian ada 2 yaitu metode *multistage sampling* untuk menentukan lokasi penelitian dan metode *purposive sampling* untuk pengambilan sampel. Total sampel sebanyak 192. Dari total sampel 192, 96 sampel berasal dari kambing dan 96 sampel berasal dari domba. Untuk menentukan sampel size berdasarkan desain prevalensi 50% dengan *level confidence* 95% dan tingkat keakuratan 10%. Sampel yang diuji adalah feses segar ternak domba dan kambing dengan menggunakan metode *natif* untuk mengetahui telur cacing yang ada di dalam feses. Hasil perhitungan prevalensi akan dihitung *confidence interval* 95% dengan menggunakan *exact binomial method*.
Hasil: Berdasarkan diagnosa laboratorium menunjukkan bahwa tingkat prevalensi cacing *Strongylida/strongyles* tertinggi pada ternak domba sebanyak 21,88% (14,1%-31,5%) dan terendah pada ternak kambing 20,83% (13,2-30,3%). Tingkat prevalensi berdasarkan pada jenis kelamin tertinggi pada jenis kelamin jantan pada ternak domba 25% (13,6-39,6%) dan 18,75% (8,9-32,6%) pada betina. Sedangkan untuk ternak kambing tingkat prevalensi cacing *Strongylida/strongyles* tertinggi pada jenis kelamin betina 25% (13,6-39,6%) dan 16,67% (7,5-30,2%) pada jantan. Selanjutnya tingkat prevalensi berdasarkan umur ternak domba lebih tinggi pada usia 9-12 bulan yaitu 31,25% (16,1-50,0%) dibandingkan dengan ternak kambing tertinggi pada umur 0-4 bulan yaitu 28,13% (13,7-46,7%).
Kesimpulan: Dari penelitian ini, diketahui bahwa tingkat prevalensi cacing *Strongylida/strongyles* pada ternak ruminansia kecil (domba dan kambing) di wilayah sentral Timor-Leste adalah 21,35% (15,8-27,8%).

Kata Kunci: Domba; Kambing; Prevalensi*; Strongyles; Strongylida*

Abstract

Objective: This research aimed to identify the prevalence of Strongyles in small ruminants (sheep and goats) in the central region of Timor-Leste.

Methods: There were two methods used in this research, i.e. *multistage sampling* to determine research sites and *purposive sampling* to collect samples. The total samples collected were 192. From the 192 samples collected, 96 samples were from goats and another 96 samples from sheep. The sample size was determined based on 50% design prevalence and 95% confidence level and absolute precision of 10%. Samples examined were fresh fecal samples from sheep and goats. The samples were tested using a native smear to identify the presence of worms' eggs in the feces. The 95% confidence interval of the prevalence was calculated using the exact binomial method.
Results: Based on laboratory examination, it was found that the prevalence of *Strongylida/strongyles* in sheep was 21.88% (14.1-31.5%) and in goats was 20.83% (13.2-30.3%). Based on sex, the highest prevalence of *Strongylida/strongyles* of sheep was found in ram 25% (13.6-39.6%) compared to 18.75% (8.9-32.6%) in the ewe. In goats, however, the prevalence of *Strongylida/strongyles* was 25% (13.6-39.6%) in doe and 16.67% (7.5-30.2%) in bucks. Based on age, the highest prevalence in sheep was found in the age group of 9-12 months old 31.25% (16.1-50.0%), whereas in goats, the highest prevalence was found in the age group of 0-4 months old 28.13% (13.7-46.7%).
Conclusions: Based on this research**,** it was identified that the prevalence of *Strongylida/strongyles* in small ruminants (sheep and goats) in the central region of Timor-Leste was 21.35% (15.8-27.8%).

Keywords: Sheep; Goats*; Prevalence; Strongyles; Strongylida*

INTRODUCTION

The majority of the population in Timor-Leste keep goats and sheep with various objectives. These objectives include cultural ceremonies; for savings and household income. The Census data in 2015 indicated that 87% of the population raised animals [1]. According to animal census data [2] the growth rate of goats was 0.8% and sheep 0.01% in Timor-Leste from the year 2004 to 2015 compared to Bali cattle 7.4% and buffaloes 6.0%. Production of small ruminants such as goats and sheep are not great due to some factors such as poor management, lack of feeding, and bad quality of feeding and also due to parasite infestations [3].

Geographically Manatuto Municipality (the research site for this study) has a smaller grazing field (approximately 13,000 ha) compared to other municipalities. For example, Lautem has approximately 39,994 ha and Covalima 34,339 ha [2]. The majority of livestock farmers in Manatuto still adopt the traditional way of rearing animals. This is because most grazing field in Manatuto is categorized as communal grazing field (Figure 1). The traditional system of raising animals in communal land gives more opportunities for disease agents to infect animals. Parasite is one of the agents that cause animal diseases. One of the most common parasites infecting livestock is worms [4].

There are three types of worms that infect livestock. These are cestode, trematode and nematode. One example of nematode is the worm that belongs to the order of *Strongylida/strongyles*. The order *Strongylida/ strongyles* includes five superfamilies. i.e. the Diaphanocephaloidea, Ancylostomatoidea, Strongyloidea, Trichostrongyloidea, and Metastrongyloidea [4]. These families of *worms are composed of* many important parasites of grazing small ruminants. Trichostrongylidae, for example, include species such as *Haemonchus contortus; Teladorsagia circumcincta; Strongyles axei; T. colubriformis; T. vitrinus; T. rugatus, and Cooperia curticei* [4,5]. In the example, members of the superfamily Trichostrongyloidea is also called trichostrongylids [6].

Figure 1. Sheep grazing in communal land in Manatuto Municipality

These worms when they infect livestock can be diagnosed by looking at the presence of their eggs in feces in the fecal examination. Adult worms of *strongylida/strongyles* live in

abomasal mucous, small and large intestines [4]. Some species such as Haemonchus, they suck blood and cause the death of animals due to anemia [7].

In general, the *Strongylida*/*strongyles*' eggs—except for those of Nematodirus and Bunostomum are similar in size and structure and cannot easily be identified in feces examination [5]. The shell of nematode eggs varies in thickness, usually consisting of three layers (inner, middle, and outer layer), and the eggs vary widely in size and shape [8]. A typical *strongylid*/*Strongyles* type egg has the following characteristics: has a smooth surface, an ellipsoidal-shaped shell, and contains an embryo in the morula (cluster of cells) stage of development when passed out through feces. Their shape is not easily distinguished, but with careful examination, they may be distinguished by their size, shape, color, the thickness of shells, and stage of development [9] and can also be differentiated via larval culture. Because the eggs of most strongylid and trichostrongylid species are identical in appearance and size, it is difficult to identify *Strongylida*/*strongyles* eggs to the genus level. If identification is needed, the feces sample must be cultured to get L3 larvae for further study[10,11].

In this study, we did not differentiate between species in the five superfamilies of *Strongylida*/*Strongyles* but simply looked at all the *Strongylida*/ *Strongyle* shape eggs under the light microscope. Therefore, this research aimed to know the prevalence of *Strongylida*/ *Strongyle worms* of small ruminants (sheep and goats) in Manatuto municipality in the Central region of Timor-Leste.

MATERIALS AND METHODS

Materials

The materials used in this research were: fresh fecal samples from goats and sheep collected directly from the rectum in the central region of Timor-Leste. The fresh samples were preserved in 10% formalin. The types of equipment used were a coolbox, gloves, masks, markers, plastic for samples, sticky tape, spatula, pipette, glass slide, cover glass, microscope, lab coats, fridge, notebooks, pens, questionnaire, and digital camera.

Site selection and sample collection

Multistage sampling was used to select the research sites. The selection started from the municipality, administration posts down to villages (suco). Based on this procedure, Sau village and Aiteas were selected. Samples collection, however, was done based on Purposive Sampling. Samples were collected based on criteria such as sheep faecal samples were taken from Sau village and goats fecal samples from Aiteas village.

Sample size

The total samples collected were: 96 fecal samples from goats, 96 fecal samples from

Table 1. The approximate sample size required to estimated prevalence in a large population with the desire fixed width confidence limits (modified from Cannon and Roe, 1982)

Expected prevalence (%)	Level of confidence 95%		
	Desired absolute precision		
	10%	***5%***	***1%***
10	35	138	3457
20	61	246	6147
30	81	323	8067
40	92	369	9220
50	96	384	9604
60	92	369	9220
70	81	323	8067
80	61	246	6147
90	35	138	3457

sheep. This sample size was based on the *expected prevalence of 50% with the Level of Confidence of 95% and Desired absolute precision of* 10% (Table 1) [12,13].

Sample testing procedures

The method used to analyze the samples was a *native smear*. First, a small number of feces was smeared on a clean microscope slide. The samples were then mixed with a few drops of water. Then a coverslip was placed over the smear. The fecal material was not left in a lump in the center of the coverslip but was evenly spread to shine through the microscope illumination. The samples then are examined under a microscope with the magnification of 10 x 10; when the egg of *Strongylida/Strongyles* was identified, the image (Figure 2) was captured using a digital camera.

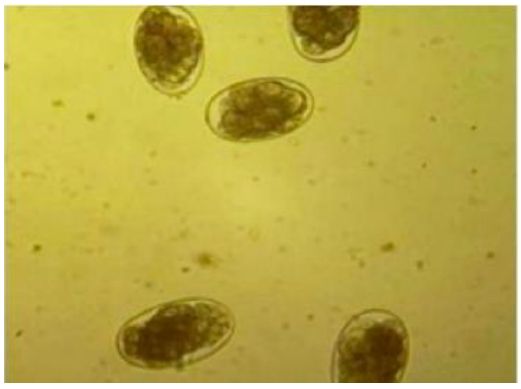

Figure 2. Strongylda eggs captured during microscopic examination

Data analysis

Excel was used to analyze the data, and the prevalence rate was calculated as follows:

Prevalence = $\frac{number\ cases}{population\ at\ risk}\ x\ 100\%$ [12,14].

RESULTS

The prevalence rate of *Strongylida/Strongyles* based on species.

The prevalence rate of *Strongyles* based on species showed that there are no significant differences between species; nonetheless, the prevalence in sheep was slightly higher (21.8%) compared to goats (20.3%, n=96) (see Table 2).

Prevalence rate of *Strongylida/strongyles* based on Sex

There are 96 samples from sheep (48 rams and 48 ewes). Based on the laboratory examination of fecal samples, it was found that the prevalence of ram is slightly higher [12 out 48–(25%) than ewes [9 out 48–(18.75%). Similarly, the prevalence in goats ranges from 16.67% to 25.00%. The total positive samples from bucks were 8 out of 48 (16.67%) compared to doe, 12 positive out 48 samples (25%) (Table 3).

The prevalence rate of *Strongylida/Strongyles* based on age groups

The laboratory result showed that 42 out of 192 **(**21.35%) samples were positive of *Strongyles* in goats and sheep. In sheep, the highest prevalence was found in the age group of 9-12 months old - 10 positive out 32 samples (31.25%). However, in goats, the highest prevalence was found in the age group of 0-4 months old - 9 positives out of 32 samples (28.13%) (Table 4).

Table 2. Prevalence rate of *Strongyles* based on species of small ruminant

Species	Total samples	Positive	Prevalence (%)	95% CI (%)
Sheep	96	21	21.88	14.5-31.5
Goats	96	20	20.83	13.2-30.3
Total	192	41	21.35	15.8-27.8

Table 3. Prevalence rate of *Strongyles* in sheep and goats based on sex

Species	Sex		Positive		Prevalence (95% CI)	
	M	F	M	F	M	F
Sheep	48	48	12	9	25.00 (13.6-39.6)	18.75 (8.9-32.6)
Goats	48	48	8	12	16.67 (7.5-30.2)	25.00 (13.6-39.6)
Total	96	96	20	21	20.83 (13.2-30.3)	21.88 (14.1-31.5)

Table 4. Prevalence rate of *Strongylida*/*Strongyles* based on age group

Species	Age group	Total samples	Diagnose		Prevalence (%)	95%CI
			+	-		
Sheep	0-4	32	4	28	12.50	3.5-29.0
	5-8	32	7	17	21.88	9.3-40.0
	9-12	32	10	14	31.25	16.1-50.0
Goats	0-4	32	9	23	28.13	13.7-46.7
	5-8	32	4	28	12.50	3.5-29.0
	9-12	32	7	25	21.88	9.3-40.0
	Total	192	41	151	21.35	15.8-27.8

DISCUSSION

In general, Manatuto municipality has a tropical climate. It has a wet and humid climate because there are a lot of rice fields and rivers, which never dry during the dry season. A wet and humid grazing field favors worms' development and infestation [15]. Gastrointestinal worms cause problems to small ruminants such as sheep and goats. Gastrointestinal worms caused animals to become skinny, weak, and dead in small and young animals [16] as well as humans. In humans, the *Strongylida*/*strongyles* are nematodes of the intestine, lower and upper respiratory tract, blood vessels, and other sites [17]. *Strongyles* is a common parasite in tropical climate, sub-tropical, and temperate countries in the world [16]. So it is expected that this worm can be found in Timor-Leste, which is also a tropical country. In the Central region of Timor-Leste, it was found that the prevalence for small ruminants (sheep and goats combine) was 21.35% (15.8-27.8%). Based on sex, the prevalence of the *Strongylida*/*strongyles* of the two species (goats and sheep) ranges from 16.67-25.00%. This finding is lower compared to the finding in Colombia for *Strongylida*/*strongyles* order in sheep, which was (38.3%) [18] and in Malaysia, which ranged from 81.7% - 91.4%) in sheep and 78.1% - 83.6% in goats [19]. The lower prevalence of *Strongylida*/*strongyles* compared to other findings in other countries of similar climate is probably due to environmental factors because the samples in this study were collected during the dry season (the month of August).

In this research, the general prevalence of *Strongylida*/*strongyles* for small ruminants was 21.35% (15.8-27.8%) (41 out of 192). If we compare between the two species, the highest prevalence was found in sheep, i.e. 21.88% (14.1-31.5%) (21 positives out 96 samples), and the lowest prevalence [20.83% (13.2-30.3%)] was found in goats 20 positives out of 92 samples (see Table 2). This is probably because the grazing behavior of goats is very selective when they graze in an open field. It is well known that goats are very selective in choosing their feed; they can even travel for miles for browsing their feed [20,21] and can tolerate feed that is bitter compared to sheep [22,23]. This result maybe just a coincidence because animals are grazing in communal land where infected animals are reared with healthy ones. It was found by some experts that controlling gastrointestinal parasites is not very effective when animals are reared in communal grazing land, and contaminated grazing land is not controlled of the parasite [9,24]. That is why it is recommended to combat parasite infestation by limiting the spread of the parasite, but this isn't easy in a *communal grazing land* setting [9,25]. The highest prevalence was found in the age group of 9-12 months old. The finding of this research is similar to the finding of research in Pakistan that the worms affect more sheep aged above nine months old. Another factor observed in the field that may have contributed to the high infestation is that farmers let the animals out early when the dew is still on the grass early in the morning [26]. During our study, it was also observed that the yard in the study site was dirty and

this condition became the source of larvae transmission.

In goats, the highest prevalence was found in the age group of 0-4 months old. This is in agreement with the finding of similar research in Pakistan that goats and sheep younger than 9 months of age had a greater infestation rate than those older than 9 months [26]. This is also in agreement with the finding in Peninsular Malaysia that in sheep, mean fecal egg counts declined from 8 months onwards, whereas in goats, this happened from 12–18 months onwards. Similar to the observation in the field of sheep, the dirty or pen yard is also observed in goats', which favors the development of worms. It was reported in India that the dirty yard or pen and humid environment favors worm growth. That is why it is essential to keep the yard or pen clean, and this becomes the key to good management to prevent parasites in animals [27].

In the Manatuto Municipality in Timor-central Leste's region, we reported the first *strongylida/strongyles* infestation in goats and sheep. It will serve as a starting point for future studies on nematode infestation in small ruminants.

CONCLUSION

From the discussions above, it was concluded that the prevalence of *Strongylida/strongyles* worm from small ruminants in Manatuto of Central Region of Timor-Leste is 21.35% (15.8-27.8%). Based on species, the prevalence of 21.88% (14.1-31.5%) in sheep (21/96) and 20.83% (13.2-30.3%) (20 positive out 96 samples) in goats.

CONFLICT OF INTEREST

We (authors) state that the content addressed in this work has no conflict of interest with any financial entity. We further declare that we have no personal circumstances or interests that could be construed as improperly influencing the presentation or interpretation of stated research findings. Because this is a self-funded study, we declare that the funders had no involvement in the study design, data collection, analysis, interpretation, manuscript preparation, or the decision to publish the results.

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