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Status of *Eimeria* infections of sheep and goat sold at small ruminant markets in Lagos State, Southwest, Nigeria.

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Abstract

Coccidiosis, an intestinal disease caused by apicomplexan parasites of the genus *Eimeria*, continues to challenge profitable small ruminant production worldwide. This study was carried out to determine the occurrence of *Eimeria* and identify the species infecting sheep and goat sold at small ruminant markets in Lagos State. Fresh faeces were collected from 83 sheep and 51 goats. The faecal samples were grossly examined for their consistency and subjected to salt floatation to detect the presence of *Eimeria* oocysts. Morphometric measurements of oocysts were taken for species identification. The number of oocysts/ml of faeces were estimated using a haemocytometer. Results revealed that 67 (80.7%) sheep and 34 (66.7%) goats were positive for *Eimeria*. There was no difference in the prevalence of infection between male and female of both animals ($P > 0.05$). The difference in infection rates between the breeds of sheep were also insignificant ($P > 0.05$). Goats with diarrheic stool had higher faecal oocyst counts (20400 oocysts/ml) when compared to those with normal stool (5250 oocysts/ml). *E. faurei* (41%), *E. ovinoidalis* (36%) and *E. intricata* (31%) were the most common of the 11 species infecting sheep in this study. Of the 8 species recovered from goats, *E. ninakhloyakimovae* (51%) and *E. alijevi* (39%) were the most frequent. The high incidence of pathogenic *Eimeria* species detected in the study area poses serious economic risks to stakeholders in Lagos State and Nigeria at large. Small ruminant farmers and traders are advised to improve biosecurity and sanitary conditions where livestock are kept.

Keywords: *Eimeria* parasites; coccidiosis; goats; sheep; small ruminant markets; Nigeria.

Introduction

Sheep (*Ovies aries*) and goats (*Capra aegagrus hircus*) are economically important livestock animals bred for their meat, milk, wool, hides and skin [1]. They make valuable contribution to household income and food security, especially among the poor living in rural areas of the developing world [2]. The production of these small ruminants is most significant in Sub-Saharan Africa and South-East Asia where livestock industries strive to meet the increasing protein demand of growing human populations [3,4].

Diseases are major constraints in livestock production worldwide [1,5]. Coccidiosis, a notable disease of farm animals is widespread among small ruminants, and is responsible for significant economic losses associated with high morbidity, mortality and increased management costs [6,7]. It is an enteric disease characterized by malabsorption, reduced weight gain, poor feed conversion efficiency, reduced productivity and death of untreated animals [8].

Coccidiosis is caused by obligate, monoxenous apicomplexan parasites of the genus *Eimeria* that live and develop within cells of the intestinal mucosa.

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About 15 and 16 distinct species of *Eimeria* are recognized worldwide to affect sheep and goat, respectively [8]. Infections with *E. ninakohlyakimovae* and *E. arloingi* are considered the most pathogenic in goats while *E. ovinoidalis, E. crandallis* and *E. ashata* are the highly pathogenic species of sheep [8]. Susceptible animals become infected when they ingest feed and/or water contaminated with sporulated oocysts of *Eimeria* found in the environment.

Although, *Eimeria* infection is prevalent among sheep and goats of all ages and breeds, it is more severe in kids and lambs, resulting in clinical coccidiosis with signs such as bloody and/or watery diarrhea, dehydration, emaciation, weakness, appetite loss and death being common [9]. Sub-clinical infections seldom result in illness or death of affected animals, but significant reduction in productivity is common [8].

Studies related to the occurrence and diversity of *Eimeria* parasites of sheep and goat in Nigeria are scarce, and not up to date [10,11]. Prevalence rates ranging from 1.8 – 80% have been reported in the north [1,11] and south [10,12]. [12] also identified seven species, with pathogenic species occurring more frequently. More recent updates on the status of these parasites will provide valuable information that underpin the effective and efficient control of small ruminant coccidiosis in Nigeria.

Small ruminant markets situated in southwestern Nigeria deal in the sales of goats and sheep bred across different regions of the country [2]. Therefore, parasitological surveys conducted in such markets will present findings that are likely to be representative of the country at large [12]. This study therefore seeks to determine the prevalence of *Eimeria* and identify the species in trade sheep and goats sold at selected small ruminant markets in Lagos, Southwest, Nigeria.

Methodology

Study area

This study was conducted in three daily urban small ruminant markets located in Gbagada, Agege and Alaba-Rago in Kosofe, Agege and Ojo Local Government Areas (LGAs) of Lagos State, respectively. Lagos State is located in the southwestern region of Nigeria, lying approximately on longitude 20 42'E and 32 2'E, and between latitude 60 22'N and 60 2'N. She has a total land mass of 3, 577sq. km. and is bounded in the North and East by Ogun State, in the West by the Republic of Benin and the south by the Atlantic Ocean. Lagos State is a wetland region consisting mainly of fresh water and mangrove swamp forests and has two notable climatic seasons: dry (November to March) and wet (April to October). She continues to play a significant role in the economy of Nigeria, serving as the nation's major financial nerve centre (www.lagosstate.gov.ng).

Sampling design and collection

This cross-sectional study spanned three months, between July and September, 2018. A total of 83 sheep and 51 goats were sampled at random from the three selected small-ruminant markets. Each market was visited only once throughout the sampling period.

About 10g of freshly egested faeces were collected from each of the selected sheep and goats presented for sale at the markets. The samples were collected in clean, sterile universal bottles and preserved temporarily in a cool-box. The breed and sex of the sampled animals were noted, including the consistency of their stools whether 'formed/normal' or 'watery/diarrheic'. The samples were transported immediately to the Parasitology laboratory in the Department of Zoology, University of Lagos for analyses. Samples that could not be processed immediately were stored in a refrigerator at 4°C for maximum of two days post collection.

Sample processing, examination and oocyst enumeration

The faecal samples were concentrated using saturated NaCl(aq) solution and directly examined under ×10 and ×40 objective lens for the detection of Eimeria oocysts [11]. The number of oocysts per ml of faeces in positive samples were counted using the haemocytometer method earlier described by [13].

Species identification of Eimeria

Oocysts were harvested from each of the positive faecal samples and sporulated in 2.5% $K_2Cr_2O_7$(aq) solution according to [13]. Morphological characteristics such as length, width, shape and colour of oocysts, size and shape of sporocysts, the presence and absence of stedia, granular pole micropyle and micropyle cap of at least 100 random sporulated oocysts per sample were used in species determination [8, 14].

Data analysis
Data were analysed on SPSS 16.0 (SPSS Inc., Chicago, IL, USA) statistical software package. Statistical differences in prevalence rates and mean intensity between breeds and sexes of both animals were determined using the Fisher's Exact test and student's T-test respectively. P-values less than 0.05 ($P<0.05$) were considered significant.

Results
Out of the 83 sheep sampled, 70 were males and 13 were females, while 74 were Balami breeds and 9 were Uda breeds. All the 51 goats sampled were West African Dwarf (WAD) goats and comprised of 39 males and 12 females (Table 1).

Eimeria spp. were detected in 67 (80.7%) and 34 (66.7%) sheep and goats, respectively in this study (Table 1). As shown in Table 1, there was no statistical difference in infection between sexes of both goats ($P>.05$) and sheep ($P>.05$) and the breeds of sheep ($P>.05$) studied.

Mean intensity was higher in males than females of both sheep (8245.6 oocysts/ml) and goats (8538.5 oocysts/ml), and among the Balami sheep breed (8000 oocysts/ml) compared to the Uda breed (7142.9 oocysts/ml) (Table 2). These observed differences were however statistically insignificant (Table 2).

The nature of the stool samples, in relation to the oocyst output of the infected ruminants is presented in Table 3. Majority of the infected sheep and goat had formed stools with oocysts/ml of faeces at an average of 8033 and 5250 respectively. Goats with diarrheic stool (20400 oocysts/ml) had higher *Eimeria* counts than those with normal stool (5250 oocysts/ml) whereas higher oocyst levels were determined among sheep with normal stool (8033 oocysts/ml) compared to those that suffered from diarrhea (6857 oocysts/ml). There was however no statistical difference in oocyst output of the infected animals presenting with formed or watery stools ($P>.05$).

Eleven different species of *Eimeria* were recovered from sheep in this study, with *E. faurei* (41%), *E. ovinoidalis* (36%) and *E. intricata* (31%) having the highest prevalence rates (Figure 1). The least prevalent were *E. crandallis* (6%), followed by *E. marsica* (5%), *E. bakuensis* (2%) and *E. granulosa* (1%). The occurrence of each *Eimeria* species according to the sex and breed of sheep are given in Figure 2 and 3. *E. ovinoidalis* occurred more frequently among rams (Figure 2) and among the Balami breed (Figure 3).

Table 1: Prevalence of *Eimeria* spp. based on sex and breed of sheep and goats sold at small ruminant markets in Lagos State

Small ruminant	Variables	No. sampled	No. infected	Prevalence (%)	P value
Sheep	Total	83	67	80.7	-
	Sex				
	Male	70	57	81.4	0.708
	Female	13	10	76.9	
	Breed				
	Balami	74	60	81.1	1.000
	Uda	9	7	77.8	
Goat	Total	51	34	66.7	
	Sex				
	Male	39	26	66.7	1.000
	Female	12	8	66.7	

Table 2: Intensity of *Eimeria* spp. infection in relation to sex and breed of sheep and goats sold at small ruminant markets in Lagos State

Small ruminant	Variables	Min. – Max. intensity	Mean intensity ± SD	P value
Sheep	*Sex*			
	Male	2000 – 70000	8245.61 ± 12306.04	0.702
	Female	2000 – 16000	6000 ± 4618.8	
	Breed			
	Balami	2000 – 70000	8000 ± 12028.22	0.211
	Uda	2000 – 18000	7142.86 ± 5398.41	
	Total	2000 – 70000	7910.45 ± 11491. 4	-
Goat	*Sex*			
	Male	2000 – 80000	8538.46 ± 15651.79	0.860
	Female	2000 – 12000	4750 ± 3991.06	
	Total	2000 – 80000	7647.06 ± 13843.02	-

Table 3: Intensity of *Eimeria* spp. infection in relation to faecal consistency of sheep and goats sold at small ruminant markets in Lagos State

Stool consistency	Sheep		Goat	
	No. infected (%)	Oocysts/ml	No. infected (%)	Oocysts/ml
Normal	60 (89.55)	8033.33 ± 11894.97[a]	29 (85.29)	5250 ± 6041.52[a]
Soft/Diarrheic	7 (10.45)	6857.14 ± 7733.66[a]	5 (14.71)	20400 ± 33686.79[a]

Dissimilar superscript letters within column for 'oocysts/ml' indicate significant difference (P<0.05)

As presented in Figure 4, 8 species of *Eimeria* infected goats in this study. Infection with *E. ninakohlyakimovae* were found to be the most prevalent, occurring in more than half of the sampled goats. Other prevalent species were *E. alijevi* (39%), *E. christenseni* (20%), *E. caprovina* (18%) and *E. arloingi* (16%) while *E. apsheronica* (8%) were the least occurring species. *E. ninakohlyakimovae* and *E. arloingi* were also observed to be most prevalent among bucks than does (Figure 5).

Higher percentage of the sheep (65.6%) and goats (76.5%) were positive for more than one species of Eimeria when compared to those with single species

infection (Table 4).

Results also revealed that more than half of the sheep (57.1%) and goats (60%) with diarrhea were infected with *E. ninakohlyakimovae* and *E. ovinoidalis* respectively (Table 5).

The various sizes of sporulated oocysts of *Eimeria* recovered from the sheep and goats are summarized in Tables 6 and 7.

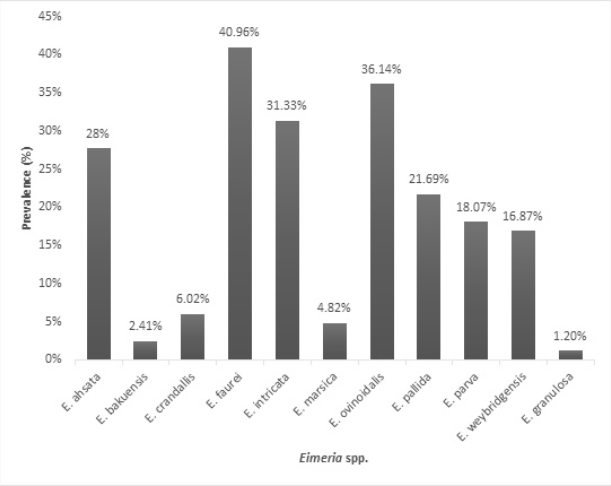

Figure 1: Prevalence of *Eimeria* species infecting sheep sold at small ruminant markets in Lagos State

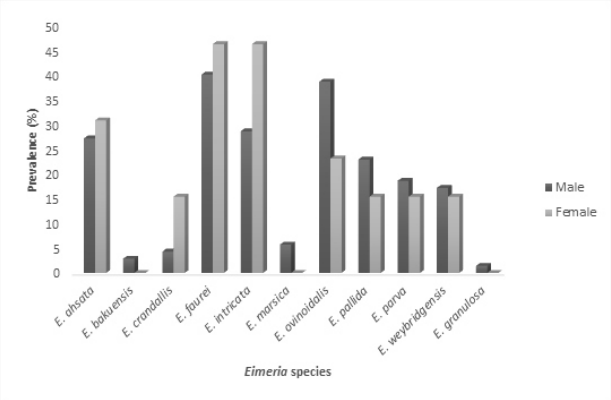

Figure 2: Gender-wise prevalence of *Eimeria* species infecting sheep sold in small ruminant markets in Lagos State

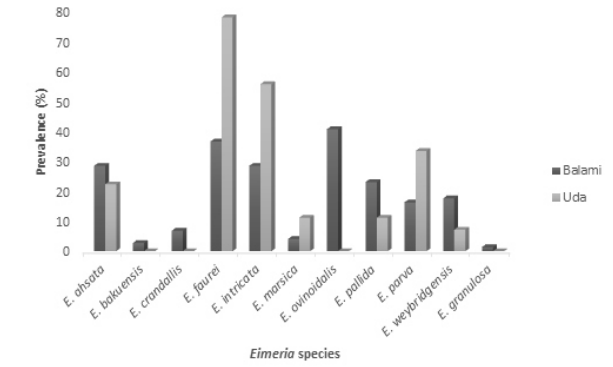

Figure 3: Breed-wise prevalence of *Eimeria* species infection of sheep sold at small ruminant markets in Lagos State

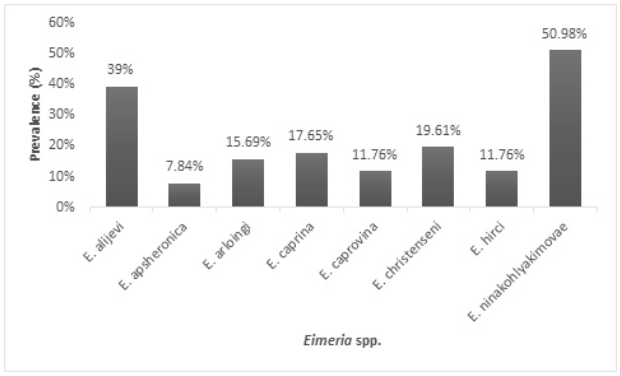

Figure 4: Prevalence of *Eimeria* species infecting goats sold at small ruminant markets in Lagos State

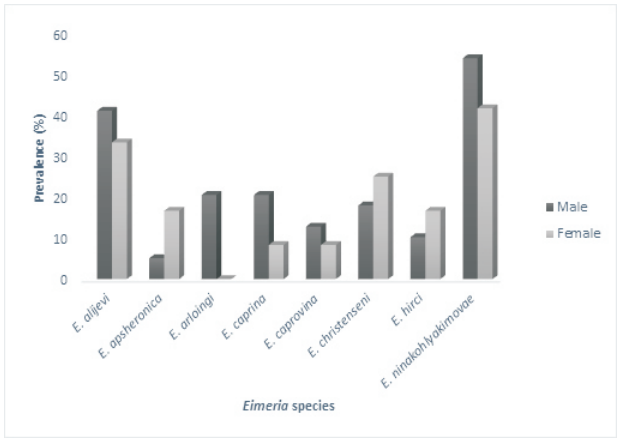

Figure 5: Sex-prevalence of *Eimeria* species infecting goats sold in small ruminant markets in Lagos State.

Table 4: Prevalence of single and mixed infections of *Eimeria* species in sheep and goats sold at small ruminant markets in Lagos State.

Small ruminant	Variables	Single infections (%)	Mixed infections (%)
Sheep	*Sex*		
	Male	20 (35.09)	37 (64.91)
	Female	3 (30.00)	7 (70.00)
	Breed		
	Balami	22 (36.67)	38 (63.33)
	Uda	1 (14.29)	6 (85.71)
	Total	23 (34.33)	44 (65.67)
Goat	*Sex*		
	Male	4 (15.38)	22 (84.62)
	Female	4 (50.00)	4 (50.00)
	Total	8 (23.53)	26 (76.46)

Table 5: Stool consistency of sheep infected with *E. ovoinoidalis* and *E. ninakohlyakimovae* infected goats in surveyed small ruminant markets in Lagos State

Faecal appearance	*E. ovoinoidalis* N (%)	*E. ninakohlyakimovae* N (%)
Formed	26 (43.33)	23 (79.31)
Watery	4 (57.14)	3 (60.00)
Total	30 (36.14)	26 (50.98)

Table 6: Sizes of sporulated *Eimeria* oocysts recovered from sheep

Species	No. of oocysts measured	Range of oocyst sizes (µm)	Average length (µm)	Average width (µm)
E. faurei	73	11.45 – 34.35 x 11.45 – 22.90	22.63	18.72
E. intricata	44	32.70 – 45.80 x 21.80 – 45.80	39.11	31.51
E. ovinoidalis	28	10.90 – 22.90 x 11.45 – 22.90	16.90	18.72
E. ashata	24	32.70 – 57.25 x 11.45 – 45.80	42.74	51.80
E parva	22	10.90 – 11.45 x 10.90 – 22.9	11.18	15.08
E. pallida	11	10.90 – 11.45 x 10.90 – 11.45	11.18	11.80
E. webridgensis	6	21.80 – 22.90 x 21.80 -22.90	22.35	22.35
E. crandallis	5	22.90 x 11.45	22.90	11.45
E. marsica	3	11.45 – 21.80 x 11.45	16.67	11.45
E. granulosa	2	22.90 – 32.70 x 11.45 – 21.80	25.50	20.70
E. bakuensis	1	34.35 x 22.9	34.35	22.90

Table 7: Sizes of sporulated *Eimeria* oocysts recovered from goat

Species	No. of oocysts measured	Range of oocyst sizes (µm)	Average length (µm)	Average width (µm)
E. ninakohlyakimovae	30	21.80 – 22.90 x 11.45 – 22.90	22.35	18.72
E. alijevi	23	10.90 – 22.90 x 10.90 – 11.45	16.76	11.18
E. caprina	13	32.70 – 45.80 x 10.90 – 22.90	39.11	18.53
E. hirci	11	21.80 – 22.90 x 11.45 – 22.90	22.35	18.72
E. christenseni	9	34.35 x 22.90	34.35	22.90
E. apsheronica	6	32.70 – 34.35 x 21.80 – 34.35	33.53	26.35
E. arloingi	5	21.8 – 22.9 x 21.8 – 22.9	22.35	22.35
E. caprovina	4	22.9 – 32.7 x 21.8 – 22.9	27.80	22.35

Discussion

Eimeria species are one of the most common and important parasites of small ruminants bred worldwide. They are responsible for moderate to severe economic losses that result from both clinical and subclinical infections in the small ruminant industry [8]. This study revealed considerably high prevalence of *Eimeria* among market-aged sheep (81%) and goats (67%), and found 11 and 9 different species infecting the ruminants respectively.

Some authors have reported similar rates of *Eimeria* infection among sheep in Nigeria [12] and Iraq [15]. All other surveys across diverse regions of the world [5,16,17,18,19,20,21,22,23,24], including Nigeria [11] revealed much lower prevalence rates. This suggests that there are conditions favoring the efficient transmission of sheep *Eimeria* in the markets where the current study was conducted. These include the presence of relatively more susceptible breeds, unhealthy and stressed animals, poor biosecurity practices and unsanitary conditions. Findings of this study agree with those of other surveys that *Eimeria* infection is not gender-related [14,17,18,20,21,25,26]. [22] however showed that female sheep were more predisposed to infection than males. This is probably because more females were sampled than males in their study. Statistical analyses also revealed that the level of infection between the breeds of sheep sampled in this study were similar, indicating that both breeds may have comparable levels of susceptibility to Eimeria infection. Another survey carried out in Jordan however found higher mean oocyst output among indigenous breeds of sheep when compared to the exotic breeds [20].

The prevalence of *Eimeria* among goats in this study was also similar to that estimated by [22] in their study in Ethiopia. In contrast, other surveys conducted in Africa and Asia either revealed higher [15,25,26,27,28,29] or lower [5,10,11,12,16,18,19,23,30] rates of infection. These discrepancies can be attributed to differences in the study settings and methodologies employed in the various regions where these studies were conducted.

Although statistically insignificant, sheep had higher *Eimeria* prevalence and infection intensity when compared to goats in this study. This agrees with the findings of other researchers [5,11,12,16,19], but is not consistent with those of [6], [18], [22] and [23],

including [15] who suggested that goats are less resistant to diseases.

The level of oocyst output might be indicative of *Eimeria* infection severity and is usually determined by the susceptibility status of the hosts. OPG levels around 50,000 – 100, 000 suggest clinical infections [8]. Average oocysts per ml of faeces recorded in this present study show that majority of the ruminants were not clinically infected and explain why very few of these animals suffered from diarrhea. Level of oocyst excretion in goats was however higher than estimated in previous studies [6,30], but lower when compared to another study carried out by [26]. The immune status of the goats in the areas where these studies were conducted, reflected by the presence of other diseases, mal- and under- nutrition, stress are all possible factors for these discrepancies. High stocking densities usually witnessed in small ruminant markets is another plausible reason why the goats sampled had higher oocyst levels than in other studies.

The general low levels of oocyst per ml of faeces observed in this study, despite the high prevalence rate of infection recorded, can be attributed to the fact that most of the animals sold in these markets have reached sexual maturity and slaughter age. Continuous exposure to low levels of infective oocysts contaminating the environment is responsible for the immunity acquired by older animals.
Majority of the ruminants in this study were suffering from sub-clinical infections of *Eimeria*. Unfortunately, farmers, marketers and non-healthcare professionals find it difficult to diagnose infection at this level since characteristic clinical signs are unapparent. These infections go unnoticed, yet they are responsible for most of the losses accrued to *Eimeria*. Furthermore, sub-clinically infected animals are good reservoirs of infection as they continue to excrete infective stages of the parasite, contaminating the environment.

There were 11 species of *Eimeria* infecting sheep in this study. Similar numbers were reported by [5] and [21] in their studies carried out in Ethiopia and Iraq respectively. This shows that *Eimeria* parasites of sheep are widespread and can survive prevailing conditions in different geographical regions of the world. [6] and [15] however detected considerably lower number of species infecting sheep in their studies. *E. ovinoidalis*, the most pathogenic sheep *Eimeria* was the second most common species encountered in this study. This finding is consistent with other surveys where pathogenic species such as *E. ahsata* [15] and *E. crandallis* [5,6] were most frequently encountered and puts sheep in the present study area at risk of high morbidity and mortality.

Eight out of 10 *Eimeria* species of goat widely reported were identified in this study. This result is similar to those of [6] and [5] who found 7 and 10 species respectively. *E. ninakohlyakimovae* the most pathogenic species of goat were the most prevalent. This agrees with the findings of some others [5,6,12] and suggests that the pathogenic species of Eimeria are common and successful parasites of small ruminants worldwide.

Despite the high prevalence of pathogenic *Eimeria* species in this study, only few animals were observed to excrete watery stool. The animals in this study must have acquired immunity to these parasites by reason of age, since most of them have reached sexual maturity and slaughter age.

The occurrence of mixed species infections in the field makes *Eimeria* diagnosis challenging and difficult for unskilled personnel and usually leads to the generation of false-positive or -negative results. More than half of the small ruminants in this study were positive for at least two species of *Eimeria*. This means that traditional tools employed in specific diagnosis of *Eimeria* infection may be inadequate in the present study area.

Conclusion

Eimeria parasites were found infecting sheep and goats at high prevalence rates in the small ruminant markets. Highly pathogenic species, *E. ninakohlyakimovae* and *E. ovinoidalis* were the most frequently encountered. It can be deduced from this study that these parasites are distributed throughout Nigeria, wherever goats and sheep are bred. Sheep and goat reared in and around Lagos are also at risk of infection through spread of human traffic moving in and out of the markets. To avoid serious economic losses, small ruminant farmers in Nigeria are therefore encouraged to intensify control and curtail the spread of these parasites.

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Competing Interests
Authors have declared that no competing interests exist.

Authors' Contributions
Adeyemi OO. conceived and designed the study, performed the statistical analyses, and wrote the first draft of the manuscript. Idowu ET. and Otubanjo OA. managed the analyses of the study and reviewed the first draft of manuscript. Ikenweiwe JC. collected sample data and conducted laboratory analyses. All authors read and approved the final version of the manuscript.

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