



Prevalence and Pathology of Sub-clinical Abomasal Coccidiosis (*Eimeria gilruthi*) in adult West African Dwarf goats from three localities in Oyo and Ogun States, Nigeria.

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ABSTRACT

Coccidiosis is a neglected disease that affects small ruminants, including goats, in sub-Saharan Africa. This study aimed to assess the prevalence, risk factors, and abomasal morpho-pathological changes associated with sub-clinical *Eimeria gilruthi* infection in adult goats from three localities in Oyo and Ogun States, Nigeria. A total of 103 abomasal samples were collected from West African Dwarf (WAD) goats in the aforementioned states. Data on husbandry system, housing, sex, age, and colour of the animals were recorded. Examination for the presence of *Eimeria* schizonts, gross and histopathological analyses were conducted. The overall prevalence of *E. gilruthi* infection in WAD goats was 67.0%. Ogun State exhibited a higher prevalence (87.5%) compared to Oyo State (12.5%). Among the three localities, Ayetoro had the highest prevalence (88.2%), followed by Odeda (86.8%), while Beere had the lowest prevalence (16.1%). The semi-intensive grazing system and housing with wood and scrap materials were associated with higher infection prevalence. Age and skin color did not significantly influence the infection rate. Gross and histopathological examinations unveiled thickening of the abomasal wall with prominent nodules. Schizonts, surrounded by inflammatory cells and edema fluid, were observed within enterocytes. Morphological and morphometric analysis of the schizonts revealed diverse developmental stages and characteristic features. In conclusion, sub-clinical *E. gilruthi* infection is prevalent among adult goats in Oyo and Ogun States, Nigeria. This study provides vital insights

into the prevalence, risk factors, and morpho-pathological changes related to sub-clinical coccidiosis in goats, which can contribute to improved management and control strategies for this disease.

Keywords: Prevalence, risk-factors, Sub-clinical, *Eimeria gilruthi*, Goats, Nigeria.

INTRODUCTION

Over the years, diseases have posed significant challenges to goat production in sub-Saharan Africa (Jahnke *et al.*, 1988). In this region, goats are susceptible to various diseases, including but not limited to helminthiasis, bacterial infections, Peste des Petits Ruminants (PPR), mange, coccidiosis, and trypanosomosis (Jahnke *et al.*, 1988; Emikpe *et al.*, 2010). Among these diseases, coccidiosis appears to be one of the significant neglected diseases of small ruminants in the sub-sahara Africa.

Coccidian organisms are intracellular protozoa parasites responsible for gastrointestinal infection in vertebrates and invertebrates (Soulsby, 1986). The parasite possesses significant nutritional, economic and zoonotic importance affecting humans, domestic and wild animals (Long, *et al.*, 1984). In ruminants, clinical and sub-clinical forms of coccidian infection have been recognized (Chartier and Paraud, 2011). The clinical presentation primarily occurs in pre-weaned and recently weaned lambs and kids, characterized by increased oocyst excretion (Long, 1984). Typically, this manifests through clinical symptoms of pasty watery diarrhea and dehydration (Levine, 2001). Such occurrences are often linked to the diminished immunity of young animals, exacerbated by factors like poor nutrition, inadequate sanitation, overcrowding, damp and wet environments, as well as the physiological stress of weaning, shipping, sudden dietary changes, or extreme weather conditions like heat and cold (Soulsby, 1986). In contrast, sub-clinical coccidian infection has been

associated with adult ruminants (Chartier and Paraud, 2011) and its economic impact on small ruminants has not been well-documented in tropical regions. Sub-clinical manifestation of *Eimeria* infection in adult sheep and goats can produce a subtle significant impact on the development, health and productivity of goats and sheep (Faizal and Rajapakse, 2001). Thus, causing a reduction in weight gain, reduced feed efficiency, increased disease susceptibility, decreased fertility and milk yield (Pout, *et al.*, 1973; Gross *et al.*, 1999; Gelberg, 2012).

In Nigeria, few reports on a faecal examination of coccidiosis in sheep and goats have been documented (Fabiya, 1980; Majaro and Dipeolu, 1981; Ikpeze, 2009). Despite the economic and nutritional importance of goats in sub-Saharan Africa, there is a paucity of information on the prevalence, risk factors and pathogenicity of sub-clinical *E. gilruthi* infection in the abomasum of adult WAD goats (Ammar, *et al.*, 2019). Therefore, this study is designed to determine the prevalence, risk factors and abomasal morpho-pathological changes of sub-clinical *E. gilruthi* infection in adult goats from three localities in Oyo and Ogun States, Nigeria.

MATERIALS AND METHODS

Study location

The study was carried out in Ayetoro (7°12'N, 30°3'E) and Odeda (7°13'N, 3°31'E), in Ogun State and Beere (7°23'47"N, 3° 55' 0"E) in Ibadan, the capital of Oyo State (Fig.1) in southwest Nigeria.

The study was carried out between April 2016 and February 2019.



Figure 1. Map of the study area where the samples were collected

The two states are close and adjacent to one another and share relatively the same rainfall per year (750 mm²). The average daytime temperature in both states is relatively high generally above 28°C with an average relative humidity of 74% (Adekunle and Agbaje, 2011). Aiyetoro and Odeda are localities in Ogun State while Beere is within the city of Ibadan where most of the indigenous people owned goats as pets, sources of income, and protein.

Data source

Information regarding husbandry and housing systems used was gathered from indigenous individuals, sellers, and butchers in the localities of the study areas. The samples (abomasal tissue) from Odeda local government areas of Ogun State were collected from local butchers. Samples from Aiyetoro was obtained from goats brought to the animal experimental units of the College of Veterinary medicine for analysis. The samples from Beere were obtained from the local slaughterhouses

in the vicinity of where some of the goats were raised. None of the goats from Beere, Odeda and Aiyetoro showed any clinical manifestations of disease such as diarrhea, emaciation, and anaemia.

Sample size determination

The sample size was calculated based on an estimated prevalence of 13%, with an accuracy of 5% within a 95% confidence interval. The calculations followed the method described by Cannon and Roe (1982).

$$N = 1.96^2 P_{exp} (1 - P_{exp}) / d^2$$

Where N= sample size required

P_{exp}= predicted prevalence

d=desired precision (5%)

1.96= constant for stratified and simple random samplings

Sample Collection

A total of 103 abomasal samples of goats were used in this study. Thirty-one samples (n=31) were collected from the Beere in Ibadan while seventy-two abomasal samples (n=72) were obtained from Aiyetoro (n=34) and Odeda (n=38) towns in Ogun State. Samples were collected into ice packs and taken to the Department of Veterinary Pathology, Federal University of Agriculture, Abeokuta for pathological analysis. The locality, husbandry system, housing, sexes, ages and colour of the animals were documented. The dentition of each animal was used to determine its age. Sex was determined using male and female, colour by visual observation.

Gross and histopathology

The abomasum was examined for the presence of raised greyish white nodules on the mucosa surface (Maratea and Miller, 2007). The nodules on each of the abomasum were scored according to their severity. These include; 1-10 foci of nodules = mild, 10-30 nodules = moderate, and above 30 nodules = severe. Additionally, the mucosal surface of the abomasum was also examined for other gross pathological changes which were duly documented. Cut sections of the abomasum were fixed in 10% neutral-buffered formalin and processed via standard paraffin-embedding techniques. Sections were cut at 5 microns and they were routinely stained with haematoxylin and eosin stains. Slides were examined under Olympus light microscope. Histopathological grading of selected tissues was carried out according to the severity, distribution, duration of the inflammatory response and degenerative changes. The severity of the lesions was graded using mild (+), moderate (++) and severe (+++) designations depending on the degree of the changes observed. The distribution of the schizonts and the associated histopathological changes were assigned as follows; focal = only one focus of reaction with the organism, multifocal = multiple foci of reaction with the organisms (3-4 organisms) and reactions around them, locally extensive = many matured and immature schizonts and inflammatory reactions observed in a localized area, and diffuse = many schizonts and inflammatory reaction all over the areas.

Morphological and Morphometric analysis of the Schizonts

Various stages of schizogony were classified according to the method of Wacha *et al.*, (1971). These include the random arrangement of nuclei (RAN), peripheral arrangement of nuclei (PAN), early and late compartmentalization arrangement of nuclei (CAN), blastophore formation (BAN) and

early and late stages of merozoites formation (MAN). Peculiar characteristics of each of the developmental stages of various schizonts were documented. These morphological features include the presence and position of the host cell nucleus (HCN), the shape of the host cell nucleus (SHCN), the presence of host nucleoli (PHCN), the number of host cell nucleoli (NHCN), pigment in the host cell cytoplasm (PHCC), parasitophorous vacuole (PPV), the shape of Schizont (SS), inflammatory exudate (INE), severity of inflammatory reaction (SIR) and host cell cytoplasmic indentation (HCCI) were documented.

Two to five schizonts of different stages of development within the epithelium of the abomasal mucosa were morphometrically determined using Aperio Imagescope digital camera attached to Olympus microscope. Parameters such as length, diameters, area, perimeters, and host cell cytoplasmic wall (HCCW) were measured. These parameters were taken from 5 different locations on each of the schizont developmental stages. The mean average of the 5 measurements was determined by dividing the sum of the five measurements by 5.

Statistical Analysis

Data were analyzed using descriptive analysis using tables for morphological and morphometric analysis of schizonts, binary logistic regression model and Pearson Chi-Square for all the hypothesized risk factors such as state and locality, housing system and age with a p-value less than 0.05 value being considered significant.

RESULTS

One hundred and three (103) abomasal samples of West African Dwarf (WAD) goats were examined for the presence of *Eimeria* schizonts from Oyo (31) and Ogun (72) states. Analysis of all the

hypothesized risk factors by binary logistic regression model and Pearson Chi-Square analysis revealed that state and locality, housing system and age were the main factors significantly associated ($P < 0.05$) with *E. gilruthi* infection in the WAD goats (Table 1). The overall prevalence of *E. gilruthi* infection in the abomasum of WAD goats from these two states was found to be 67.0% ($n = 69/103$). Of the 103 samples collected from both states, Ogun state had the highest prevalence of 87.5% ($n = 63/72$) compared with Oyo state with a prevalence of 12.5% (5/31). There was a significant difference ($p < 0.05$) in the number of positive samples from Ogun state compared with Oyo State.

Odeda with a prevalence of 86.8% (33/38) while Beere had the least prevalence of 16.1% (5/31). There was a significant difference ($p < 0.05$) in the number of positive samples from Ayetoro compared with other localities.

Based on the husbandry system, those in the semi-intensive grazing system had the highest prevalence of 67.6% (40/61) compared with those reared on the open-grazing system with the prevalence of 66.7% (28/42) but there was no significant difference ($P > 0.05$).

Table 1. Prevalence and Risk Factors Associated with *Eimeria gilruthi* in West African Dwarf Goats

Risk factors		N tested (n=103)	N (+ve)	Prevalence (%)	P-value	OR
State	Oyo	31	05	12.5	0.000	36.4
	Ogun	72	63	87.5		
Locality	Beere	31	05	16.1	0.000	-
	Odeda	38	33	86.8		
	Aiyetoro	34	30	88.2		
Husbandry system	Open grazing	42	28	66.7	0.908	1.05
	Semi-intensive	61	40	67.6		
Housing system	No House	40	17	42.5	0.000	5.75
	Wood and scrap	63	51	81.0		
Sex	Male	63	42	66.7	0.862	1.077
	Female	40	26	65.0		
Age (Years)	1-2	22	19	86.4	0.001	-
	3-4	65	34	52.3		
	>4	16	15	93.8		
Colour	Black ± white	95	64	67.4	0.609	-
	Brown	06	03	50.0		
	White	02	01	50.0		

Of the three localities considered, Ayetoro had the highest prevalence of 88.2% (30/34) followed by

The goats housed with wood and scrap exhibited a highly significant difference ($P > 0.05$) in prevalence

at 79.7% compared to those without housing, which had a prevalence of 20.3%. The male goats showed the highest prevalence of 84.1% than female goats (15.9%), although there was no significant statistical difference between sexes and the *E. gilruthi* infection. Goats between the ages of 2-3 years had the highest prevalence of 49.3%. This was followed by goats between ages 1-2 years, and goats above 3 years of age had the least prevalence. On the basis of skin colour, goats with black and white patches had the highest prevalence of 91.3%, although those with black and white patches were over-represented. Sex, husbandry system and skin colour of goats had no significant ($p>0.05$) effect on prevalence of *E. gilruthi* infection in this study.

Gross and Histopathology

The abomasal wall was diffusely thickened and the mucosal surface was faintly rough (Plate I). The mucosal surface revealed numerous, widespread and prominent nodules which were small, whitish, non-hyperaemic and non-pedunculated with 4 mm to 8mm in diameter in severely infected cases (Plate I). The larger nodules were visible from the external surface of the abomasum through the tunica serosa. Upon incision, the abomasa wall was diffusely edematous and thickened. No other parasites were detected in abomasa surfaces or contents.



Plate I: Normal abomasal mucosa surface (1st Image) and diffusely thickened and faintly rough abomasal mucosal surface with widespread and prominent nodules which were small, whitish, non-hyperaemic and non-pedunculated (2nd Image)

Histopathological examination of the nodules revealed papillary hyperplasia of the abomasal mucosal epithelium. The most prominent microscopic lesions were degenerative changes and inflammatory reactions. The mucosa was multifocally effaced by the accumulation of inflammatory cells centered on fragments of schizonts. The lamina propria and submucosa were diffusely expanded by oedema fluid and moderately infiltrated by lymphocytes, plasma cells and a few lymphoid nodules with few but sparse neutrophils, eosinophils, and macrophages in the lamina propria. Eosinophils were not observed at the earlier stages of schizogony. It was only observed toward the late stages of schizont's development. The degree and the nature of inflammatory reactions depend on the stage of schizont development. Cystic dilatation of the secretory glands and fibroplasia was common in many samples. Many schizonts were intact, surrounded by neutrophils and macrophages, and sometimes by oedema fluid (Plate II).

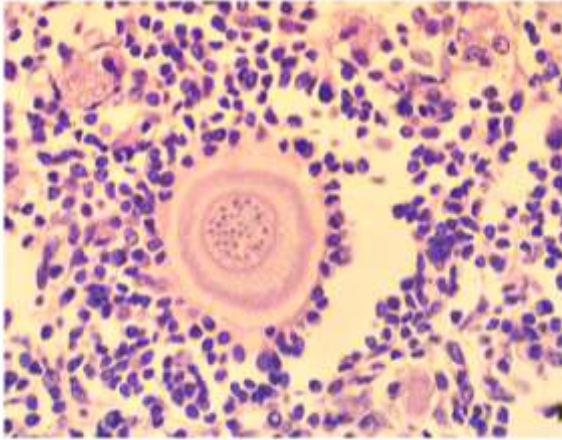


Plate II: Schizonts was intact, surrounded by neutrophils, macrophages and few plasma cells, and by oedema fluid. H&E Stain. Scale bar = 100µm

There were atrophy and effacement of the glandular structures as well as compression of adjacent epithelial cells in samples that were positive for *E. gilruthi* infection (Plate III). Schizonts were composed of numerous banana-shaped merozoites within enterocytes (Plate IV).

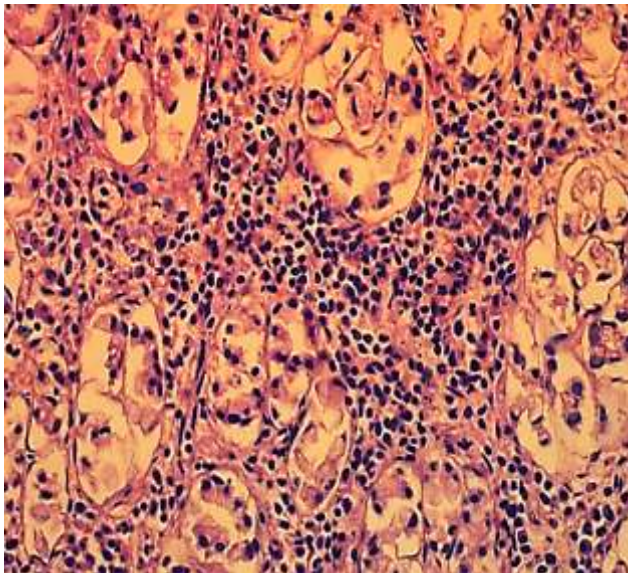


Plate III: Atrophy and effacement of the glandular structures (arrows). H&E Stain. Scale bar = 100µm

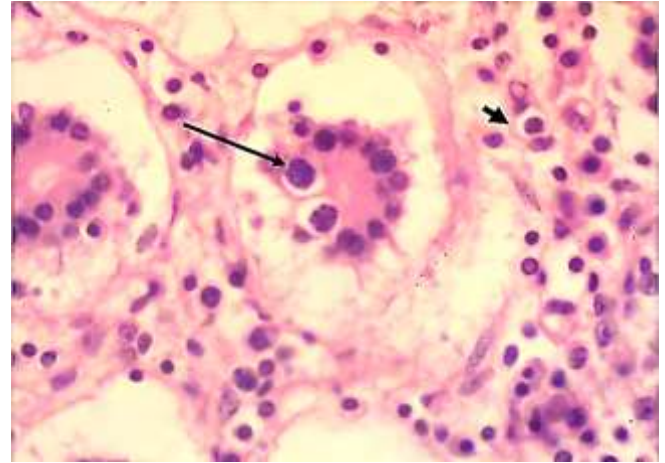


Plate IV: Microgamonts were round and peripherally located in the nuclei (arrow). H&E Stain. Scale bar = 50µm

Morphological and morphometric Analysis of the Schizonts

The morphological and morphometric parameters are depicted in Tables 2 and 3. Many intact and degenerate protozoal schizonts were observed in the upper and deep part of the mucosa and few were found in the underlying submucosa. Schizonts were ovoid, thick to thin-walled and contained thousands of immature cytomeres (Eimeria parasites) within a large parasitophorous vacuole (Plate V).

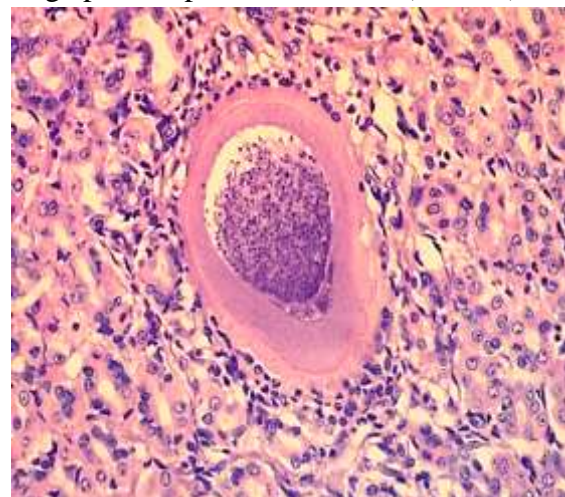


Plate V: Schizonts were ovoid, thick walled and contained thousands of immature cytomeres (Eimeria parasites) within a large parasitophorous vacuole. H&E Stain. Scale bar = 50µm

Table 2: Morphological features of various Schizogonic developmental stages of *Eimeria gilruthi* in the abomasum of WAD goats.

Developmen tal stages	Characteris tic features	NE	HC N	SHC N	PH NC	NH CN	PH CC	PP V	SS	INE	SIR	TCE	HCCI
1	Random nuclear arrangement	5	+	Oval to ellipsoidal	+	1-3	+	+	Oval	+	+1	LY, MQ	+
2	Peripheral nuclear arrangement	5	+	Oval to circular	-	1-2	+	+	Oval to circular	+	+2	LY, MQ	+
3	Nuclear Compartmentalization.	5	-	-	-	-	-	+	Oval	+	+2	LY, MQ	+
4	Blastophore formation	5	-	-	-	-	-	-	Oval to Circular	+	+2	NQ, EQ, MQ, LY	+
5	Merozoite formation	5	-	-	-	-	-	-	Oval to Ellipsoidal	+	+3	NQ, EQ, MQ, LY	+

HCN= Host cell nucleus. *SHCN*= Shape of host cell nucleus, *PHNC* = presence of Host nucleoli, *NHCN*=Number of Host cell nucleoli, *PHCC*= pigment in host cell cytoplasm, *PPV*=Parasitophorus vacuole, *SS*= Shape of Schizont, *INE* = inflammatory exudate, *SIR* = Severity of inflammatory reaction, *TCE* = Types of cellular exudate, *HCCI* = Host cell cytoplasmic indentation.

Table 3: Morphometric Analysis of various developmental stages of *E. gilruthi* schizonts in the abomasum of West African Dwarf goats

S/N	Developmental stages	NE	Length(μm) (Range)	Diameter (μm)	AS (μm^2)	PS(μm)	HCW (μm)
1	Random nuclear arrangement	5	217.55 \pm 7.7	158.7 \pm 34.7	139,475.8 \pm 11.0	611.3 \pm 274.98	29.61 \pm 16.71
2	Peripheral nuclear arrangement	3	221.4 \pm 0.03	175.6 \pm 23.1	159,072 \pm 57.06	646.15 \pm 150.80	24.69 \pm 8.3
3	Nuclear compartmentalization	5	280 \pm 9.33	201.13 \pm 6.3	167,668.0 \pm 209.2	788.6 \pm 47.7	17.66 \pm 5.4
4	Blastophore schizont	5	415.5 \pm 129.3	306.3 \pm 171.0	400,296 \pm 257.1	1180.0 \pm 345.1	13.98 \pm 16.7
5	Merozoite schizont	3	283.0 \pm 70.6	342.4 \pm 157.25	415,201 \pm 370.1	1179.5 \pm 565.7	11.31 \pm 4.1

NE = Number examined, AS= Area of schizont, PS = Perimeter of schizont, HCW = Host cell wall

Schizonts with the random nuclear arrangement (RNA) showed an oval to slightly elongated shape. The schizont occupied an area of $139,475.8 \pm 11.0 \mu\text{m}^2$, a diameter of $158.7 \pm 34.7 \mu\text{m}$, and a perimeter of $611.3 \pm 274.98 \mu\text{m}$. The host cytoplasmic wall was thickened ($29.61 \pm 16.71 \mu\text{m}$) and stratified into two layers. Both layers were lightly and variably eosinophilic and homogenous. These were separated in some specimens by a thin layer that was intensely eosinophilic. The inner layer showed numerous diffusely basophilic pigments than the outer layer. The external layer showed radial striations that extended outward to form host cell cytoplasmic indentations (HCCI) with the adjacent structures (Plate VII). The hypertrophied nucleus of the host cell was oval to ellipsoidal in shape. It was compressed and displaced toward one extremity. It revealed margined chromatin and the presence of 1-3 nucleoli. The nuclei of the parasites (cytomeres) were randomly scattered within the parasitophorous vacuole of the host cell cytoplasm. All the schizonts with RNA were surrounded by a few inflammatory cells, mostly lymphocytes, macrophages, neutrophils.

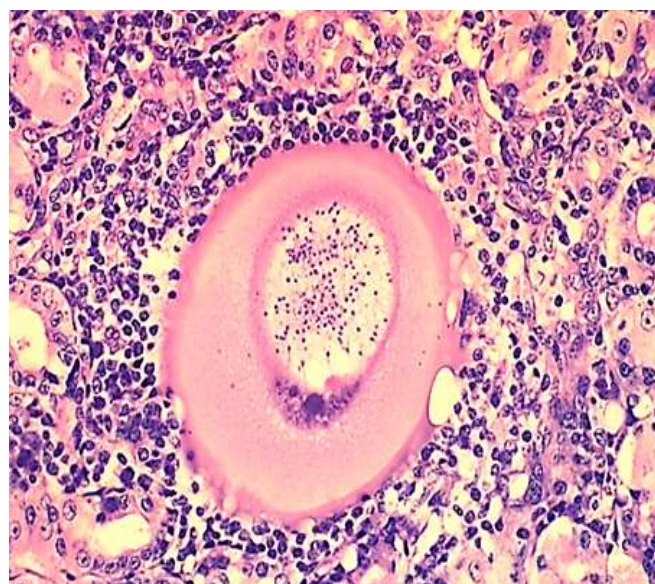


Plate VI: Schizonts with HCCI on the outer layer (thick) and eosinophilic material in the inner layer (thin). H&E Stain.
Scale bar = 50 μm

Schizont with peripheral arrangement displayed closely attached nuclei materials to the wall of the host cell cytoplasm. Schizonts with the peripheral nuclear arrangement (PNA) showed an oval to spherical shape. The schizont occupied a cellular area of $159,072 \pm 57.06 \mu\text{m}^2$, with a diameter of $175.6 \pm 23.1 \mu\text{m}$ and perimeter of $646.15 \pm 150.80 \mu\text{m}$. The host cell cytoplasmic wall was slightly

thickened without stratification like Schizont with RNA. The host cell cytoplasmic wall was eosinophilic and homogenous and contained a single hypertrophied host cell nucleus without visible nucleoli. The cytoplasmic indentations on the outer part of the host cell were considerably reduced in size ($24.69 \pm 8.81 \mu\text{m}$). The hypertrophied nucleus of the host cell was oval, compressed and displaced toward one side of the extremities. The nuclei of the parasites were peripherally arranged. Schizont with PNA were moderately surrounded by inflammatory cells, mostly lymphocytes, macrophages with few neutrophils and eosinophils (Plate VII).

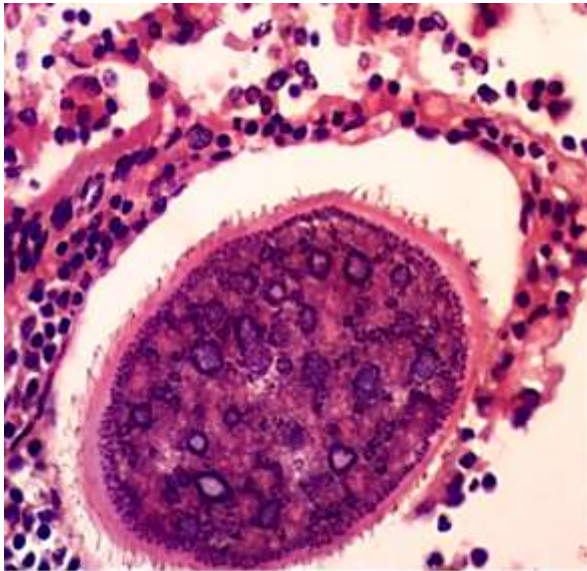


Plate VII: Schizonts with the compartmentalization of nuclear arrangement (CNA) showed an oval-shaped appearance, moderately surrounded by inflammatory cells, mostly lymphocytes, macrophages and neutrophils. H&E Stain. Scale bar = $20 \mu\text{m}$

Schizonts with the compartmentalization of nuclear arrangement (CNA) showed an oval-shaped appearance (Plate VIII). The schizont occupied a cellular area of $167,668.0 \pm 209.27 \mu\text{m}^2$, diameter of $201.13 \pm 6.3 \mu\text{m}$, and perimeter of $788.6 \pm 47.7 \mu\text{m}$.

The HCW was slightly thickened ($17.66 \pm 5.40 \mu\text{m}$) without stratification. The cytoplasmic wall was eosinophilic and homogenous. The cytoplasmic indentations on the outer part of the host cell wall were not prominent, and the host cell nucleus was not visible. There was compartmentalization of the cytomeres nuclei with infolding of peripheral arranged nuclei within the parasitophorous vacuole. Schizont with CNA was moderately surrounded by inflammatory cells, mostly lymphocytes, macrophages, neutrophils and eosinophils (Plate VIII).

Schizont at the blastosphere stage revealed a spherical shape and was moderately larger than other schizonts. The schizont occupied a cellular area of $400,296 \pm 257.1 \mu\text{m}^2$, a diameter of $306.3 \pm 171.0 \mu\text{m}$ and a perimeter of $1,180.0 \pm 345.1 \mu\text{m}$. The host cytoplasmic wall was slightly thickened (13.98 ± 16.19) without stratification. The cytoplasmic wall was eosinophilic and homogenous with mild visible cytoplasmic indentations and the host cell nucleus. There was the formation of small rings of parasite nuclei. There was no parasitophorous vacuole. Schizonts were moderately and diffusely surrounded by inflammatory cells, mostly neutrophils eosinophils, lymphocytes and macrophages. (Plates VIII).

Schizonts of the merozoite stage revealed oval to spherical shape. The schizont occupied a cellular area of $415,201 \pm 370.1 \mu\text{m}^2$, the diameter of $342.4 \pm 157.25 \mu\text{m}$ and perimeter of $1179.5 \pm 565.7 \mu\text{m}$. The host cytoplasmic wall was very thin ($11.31 \pm 4.34 \mu\text{m}$) without stratification and host cell nucleus. The cytoplasmic wall was eosinophilic and homogenous without visible cytoplasmic indentations and the host cell nucleus. There was the formation of rows of nuclei in longitudinal and tangential forms without parasitophorous vacuole. Merozoite schizonts were severely surrounded and

invaded by inflammatory cells, mostly eosinophils, neutrophils, and macrophages. The HCW in some appeared disintegrated as many inflammatory cells entered into the schizont. Each of the merozoites had various degree of inflammatory cells invasion.

DISCUSSION

This study revealed a high prevalence of *E. gilruthi* infection (formerly *Globidium gilruthi*) in the abomasum of WAD goats in the study area. Most of the available works on abomasal coccidiosis due to *E. gilruthi* have been in sheep (Fox *et al.*, 1991; Maratea and Miller, 2007). Few case reports have been documented in goats (Hermosilla *et al.*, 2016), but prevalence studies with a large number of goats on abomasal coccidiosis are rare. To the best of the author's knowledge, this is the first prevalence study on abomasal coccidiosis in the WAD goats in sub-Saharan Africa. This study is in agreement with the findings of Mc Dougald, (1979) who reported that *E. gilruthi* parasitized the abomasum of sheep though in this case, the host is different. The millet-sized nodular mucosa surface used for gross morphological identification of the organism is consistent with the previous reports (Maratea, and Miller, 2007; Hermosilla *et al.*, 2016) who observed the same lesion on gross examination. The identification of the schizonts on abomasal mucosa using microscopy is consistent with the work of Se'naud *et al.*, (1984), who reported that infections are generally believed to be incidental and are characterized by the presence of giant schizonts within the mucosa of the abomasum and less commonly in the duodenum.

In this study, the overall prevalence of 67% of the *E. gilruthi* parasite in the abomasa of WAD goats was found to be higher compared with previous studies on intestinal coccidiosis in sheep and goats (40%) (Soliman, 1960). However, this prevalence is lower than the studies of Kambarage *et al.*, (1996) who reported 97.3% from faecal samples of goats in

Tanzania and Kheirandish *et al.*, (2014) who obtained 89.9% in goats from Southeastern Iran.

According to the State and locality of infection, the reason for the high prevalence of the disease in Ogun State (Aiyetoro and Odeda) at 89.6% while Oyo State (Beere) showed the least prevalence of 16.1% is unknown. This reason for this disparity in prevalence according to locality might have been due to accessibility to Veterinary care (anti-coccidial agents) at Beere, a locality within the city of Ibadan compared with Aiyetoro and Odeda which are remote towns from the city of Abeokuta which might not have had access to veterinary services over the years.

In previous studies, age has been considered an important risk factor for different species of Eimeria infection (Bawm *et al.*, 2020), however, in this study, all ages of animals were equally exposed and there was no association ($P > 0.05$) between age and the risk of *E. gilruthi* infection.

The type of husbandry system has been shown to influence the prevalence of coccidian infection in both small ruminants (Ikpeze *et al.*, 2009; Bawm *et al.*, 2020). Previous studies have demonstrated a high prevalence of coccidian infection in both semi-intensive systems (Ikpeze *et al.*, 2009). Despite the high prevalence of coccidian infection in goats under the semi-intensive system in this study, there was no significant association ($P > 0.05$) between the husbandry system and the risk of *E. gilruthi* infection. The high level of coccidian infection in the semi-intensive mode of rearing might be due to contamination from bare soil from which goats feed directly in this part of the country.

Animals housed in wood and scrap showed a significant rate of infection compared to those without houses. This might be a result of faecal contamination of water and feed, especially in places where animals fed on bare un-concrete floors which is very important in the transmission of many

protozoa and favouring sporulated oocysts ingestion (Sharma *et al.*, 2017).

A previous study has shown that sex is a non-determinant factor in the epidemiology of coccidiosis in goats (Kheirandish *et al.*, 2014). This is in agreement with this study, in which there was no significant association between sex and the risk of *E. gilruthi* infection. Although other workers have shown either male or female goats to be significantly linked to *E. gilruthi* infection. This might be due to variations and imbalances in the number of samples collected during their studies.

The result of this study is not in tandem with the findings of Paperna (1999) who reported *Globidia* parasites exclusively confined to the small intestine of the Australian gecko (*Heteronotia binoei*) and were absent in other parts of the digestive tract. The distinct ovoidal shape of the organism reported in this study is consistent with the description given by Scholtyseck (1979).

Some of the tissues that showed appreciable gross lesions of grayish-white nodules were also positive at histopathology. Thickened abomasal wall and nodular mucosa surface seen in this study were consistent with the findings of Maratea and Miller (2007) who reported the same lesions in the abomasum of a sheep, but the presence of hyperaemic mucosal surface and petechiations documented in their report were not observed in all the tissues examined in this study.

Microscopically, abomasal mucosa was diffusely thickened and lined by hyperplastic mucous neck cells and contained markedly decreased numbers of parietal cells. This agrees with the findings of Maratea and Miller (2007) who reported the same lesions in the abomasum of a sheep.

Other histopathological changes observed such as lymphocytic infiltrations, presence of polymorphs, fibrosis, atrophy and effacement of the glandular structures are consistent with the findings of Maratea and Miller (2007) and Khodakaram-Tafti

and Hashemnia, (2017) who reported similar histopathological changes in *E. gilruthi* infected abomasum of sheep. Interestingly, eosinophils were not observed at the earlier stage of schizont development in this study, but at the later matured phase of the organism. This is in agreement with the report of Ruiz *et al.*, (2013) in which eosinophilia was not observed in experimental kids orally infected with *E. nina* until the 3-week p.i. The reason for this is unknown, but it is possible to speculate that the cascade of molecular events between the parasite and the surrounding tissues has been obscured by the intracellular presence of the organism and the thickened HCW. The thickened HCW might have shielded the organism from interacting with the tissue environment and consequently de-activated or prevented the natural innate cellular response against the parasite.

Tissues from goats that were dubbed as 'negative' but with similar histopathological changes might not have been negative samples due to omission of the organism during sectioning, and consequently not observed microscopically. Fibroplasia was specific to the abomasal mucosae of goats that were positive and negative. This could be due to the effect of recurrent healing or administration of coccidiocidal agents previously.

The effect of the organism on the secretory glands (gastric and fundic) causing degenerative and necrotic changes with effacement could negatively impact the secretion of digestive enzymes with its effect on glandular functions of the abomasum and subsequently digestion. The destruction of the parietal cells could lead to a decrease in hydrochloric acid secretion which might lead to an increase in the pH (Alkalinization) of the abomasum and consequently, the proliferation of pathogenic organisms with subsequent diarrhea as stated by Pout *et al.*, (1973) and Maratea and Miller,

(2007) who postulated that coccidiosis causes a reduction in weight gain, feed efficiency and increased susceptibility to other diseases.

In this study, no mortality was recorded due to *E. gilruthi* infection in all the WAD goats examined since many of the goats were slaughtered in the local abattoirs. This is not in agreement with the previous reports in which mortalities were recorded in cases of abomasal coccidiosis in Gran Canaria goats (Gómez-Villamandos *et al.*, 1992).

The morphological appearance of the various stages and associated structures of schizont development in this study is in tandem with previous reports in sheep and goats (Hermosilla *et al.*, 2016; Maratea *et al.*, 2007). The early hypertrophy of the nuclei and HCW, formation of multiple nucleoli, cytoplasmic indentations, and parasitophorous vacuole, appears to be the host cell response to the invasion and development of *E. gilruthi* infection in WAD goats. Contrary to the response observed in Holstein-Friesian calves infected with *Eimeria auburnensis*, where minimal or no cytoplasmic hypertrophy was observed in the hepatic coccidial white cells (HCW) (Chobotar *et al.*, 1982), the thickened HCW in this study may have played significant roles in the propagation and development of *Eimeria* schizonts. These roles might include; a) prevention of molecular events between schizonts and the surrounding tissues, b) prevention of schizont from being invaded and destroyed by phagocytic cells (eosinophils and macrophages), thus rendering the host cellular immune response incompetent during early developmental stages, c) provision of nutrition to the schizonts and d) anchorage of the parasitized host cell by the cytoplasmic indentation after been detached from the adjacent cells.

The process by which schizonts change from one developmental stage to the other is unknown in this study, but it is possible to suggest that the maturity

of the organism from one stage to the other has brought about characteristic changes in the morphological appearance and arrangement of the cytomeres within the schizonts over time. The molecular events and other factors involved in this transformation might warrant further investigations. The mechanism of dissolution or disintegration of the HCW and the factors involved at the merozoite stage is also unknown, although invasion by leukocytes has been suggested in previous studies (Hammond *et al.*, 1966; Wacha *et al.*, 1971) and the disappearance of this barrier appears to be associated with the attraction of numerous leukocytic cells with subsequent schizont's invasion. Moreover, the idea of phagocyte invasion at the earlier stage of schizont development to cause dissolution of the HCW might not have been possible due to the thickness of the HCW. This is evident by the presence of some phagocytes entangled within the outer cytoplasmic indentations. Thus, it is not out of place to suggest the possibility of other factors which might have been involved in the dissolution of HCW at the later stage of the schizont's development. These might include; a) the internal production of degradative or lysosomal enzymes by matured merozoites which weakened and soften the HCW, b) the direct invasion of the HCW by the merozoites, c) mechanical expansion or stretching of the HCW as the schizonts developed and matured intracellularly d) the release of lysosomal enzymes by the surrounding phagocytes and e) the active movement of abomasum which might rupture the delicate wall of the merozoite stage (Wegrzyn, 1981). Further studies on the mechanism of schizont dissolution or rupture at an earlier stage are warranted in order to elucidate more on the prevention of the disease and the possibility of eradicating the parasite at the earlier phase of schizogony. An effective coccidial

agent that would target any of the premature stages of schizogony either by preventing the development of the schizonts, premature dissolution or rupture of the schizonts and the release of immature merozoites might prove effective in breaking the cycle of schizont's development in the abomasum of small ruminants.

This study has shown that *Eimeria gilruthi* infection acts as a space-occupying lesion in the abomasum of WAD goats that incites persistent or continuous inflammatory response, which is mostly sub-clinical, evidenced by the absence of clinical signs of diarrhea usually associated with coccidiosis infection and infiltration of mononuclear cells (lymphocyte and macrophages) with few neutrophil and eosinophils toward the later stage of the schizont development (blastophore and merozoite stages). The characterization of these immune cells at different stages of schizont development using immunohistochemistry is underway and this might shed more light on the immunology of abomasal caprine coccidiosis in the nearest future.

The sequellae of abomasal coccidiosis in this study are the destruction of secretory glands and their replacement by inflammatory exudate and fibrous connective tissue. This could eventually lead to complete effacement of parietal and other glandular cells and consequently cause mal-digestion and mal-absorption with permanent dysfunction of the abomasum due to diffuse fibrosis. This would greatly impact the size, weight and health status of goats in this region.

Taking into consideration the deleterious effects of *E. gilruthi*-induced abomasitis in goats, it thus becomes necessary that efforts should be geared toward a better understanding of the epidemiology and pathogenicity of the disease to reduce low weight gain, low productivity and unthriftiness in goats in this region. Awareness of the use of anti-

coccidia agents by livestock farmers with WAD goats especially in rural areas should be intensified to reduce damages caused by this coccidian agent. Apart from genetic and environmental factors which some authors have alluded to be responsible for the unthriftiness or low growth rate of WAD goats in sub-Sahara African (Alade *et al.*, 2008; Peter *et al.*, 2015), subacute and continuous chronic-active abomasitis induced by *E. gilruthi* in goats early in life might contribute immensely to the low productivity of this animal.

In conclusion, this study revealed that *E. gilruthi* infection in WAD goats is a subclinical phenomenon, its subtle and sublethal effects over time on abomasal mucosa might cause great nutritional loss and consequently reduce protein and economic value of WAD goats. The absence of clinical signs such as diarrhea, emaciation, or weight loss and other signs associated with gastrointestinal diseases in this study, suggests that goats above 1 year of age in the southern part of Nigeria are tolerant to coccidian infection ("coccidiotolerant") which possibly showed the resilience and the resistant ability of this breed of goat to infection and to survive in the sub-Sahara Africa.

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