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SURVEY OF GASTROINTESTINAL AND PULMONARY PARASITISM IN TWO LOCAL CATTLE BREEDS IN THE HIGH GUINEA SAVANNAH, CAMEROON

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ABSTRACT

Cattle production accounts for over sixty percent of meat requirement in Cameroon but helminthosis greatly hinders its productivity. A study on worm burdens in digestive and pulmonary organs of cattle was done between February 2018 and January 2019 at a Research Centre in Cameroon with fourteen apparently healthy adult cattle that were necropsied. Recovered worms were adequately processed and identified with standard keys. Data were subjected to descriptive statistic and worm burdens recorded according to sex of worms, as well as breeds and sex of cattle. Analysis of variance (ANOVA) in general linear model on raw values was summarized as arithmetic means with standard errors of means. Mixed-model ANOVA verified relationship of worm burdens with cattle breed and sex. Nine Nematodes (*Haemonchus contortus*, *Cooperia* species, *Trichostrongylus axei*, *Trichuris globulosa*, *Oesophagostomum radiatum*, *Nematodirus* species, *Ostertagia ostertagi*, *Dictyocaulus viviparus*, *Setaria labiatopapillosa*), two Trematodes (*Calicophoron microbothrium* and *Fasciola gigantica*), and one Cestode (*Echinococcus granulosus* Hydatid cysts) were recovered at necropsy. Total worm burden was 42252; commonest infections being *Haemonchus contortus* (40.9%) , *Cooperia* species (38.2%) , and *Trichostrongylus axei* (9.5%) . Female worms 28503(67.5%) established more than males 13644(32.3%). Hermaphrodites were only 105(0.3%) of total worm burden. Worm burden/cattle was 3018 (2966/bull and 3027/cow) but for cattle breeds, burdens were

3331/Namchi (600/Namchi bull and 4696/Namchi cow) and 2933/Gudali (5331/Gudali bull and 2693/Gudali cow). Gudali breed showed more parasitic infections than Namchi, but the difference being statistically non-significant (P > 0.05). Worm burden/cattle was 3018 with an average of 2966/bull and 3027/cow but for cattle breeds, burdens were 3331/Namchi with and average of 600/Namchi bull and 4696/Namchi cow and 2933/Gudali with an average of 5331/Gudali bull and 2693/Gudali cow. Sex wise females recorded higher infection rates than males but the difference being statistically non-significant (P > 0.05). Hypobiotic larvae were not found in the digestas. Morphological identification of helminths has provided basis for further moléculaire identification to ensure sustainable helminthosis management intervention. Meanwhile strategic anti-helminth treatments will usefully control infections and re-infections in cattle in the area.

Keywords: Endoparasites , High Guinea Savannah, Local cattle, Cameroon

INTRODUCTION

In Cameroon, cattle which represent about 16% of the national agricultural production, supplies 61% of the national meat request (Ebangi *et al*., 2001). Adamawa Region in general (with more than 5.5 million or 38% of cattle population in Cameroon), and Vina Division in particular has long been considered to be one of the greatest source of beef in Cameroon (Minepia, 2003). However, in many African countries including Cameroon, helminthosis adversely affects production and productivity. Helminth parasites can only be identified at post-mortem examination on adult cattle at municipal abattoirs (Waller,1997), the results of which may not be representative of the general cattle population because municipal abattoirs most likely have drawn animals from those showing signs of ill health or weakness (Waller, 1997). It was earlier stated that parasite burden of sick animals may be higher than those of healthy ones due to immune suppression or lowered immunity (Wymann, 2006). Although the causes of helminth parasitism of cattle are ubiquitous in Cameroon, parasite species in a particular area may not be the same in another agro-ecological zone. Some studies in Cameroon have investigated parasite burdens in goats and cattle of the North-West (Ndamukong and Sewell, 1992) and South-West (Ngole *et al*., 2003) regions. Parasitic worms in cattle are unlikely to be eradicated in the foreseeable future because of the varied nature of cattle production systems and due to the fact that worms rapidly developed resistance to deworming agents (Ebene *et al*., 2016).

To maintain production goals, Adamawa cattle owners and veterinarians need to understand and manage the relationship between parasitic worms, the host and environment. In the High Guinea Savannah of Cameroon there is paucity of published information on helminth parasite species of cattle. The multi-species nature of parasite infections in livestock is a challenge to DNA-based diagnosis but some authors still believe that microscopic identification of

genera of common helminthes of cattle is still important and very effective since all the identification methods including DNA and Immunologic diagnostic tests have their limitations (Taylor et al. , 2016; Zajac and Convoy, 2012). The accurate diagnosis of helminth parasites of ruminants, regardless of their developmental stages is essential for the effective control and correct treatment of animal diseases (Grillo *et al*., 2007).

It is therefore important to collect data on species composition of helminth parasites from apparently healthy cattle not slaughtered at communal abattoirs in the High Guinea Savannah of Adamawa Region in Cameroon. This will enable us to fill the information gap regarding helminth parasites in both trypano-susceptible and trypano-resistant breeds of cattle in such a high beef production area. Development of cost-effective and sustainable control programme of helminth infections requires thorough knowledge of species of parasites present.

MATERIALS AND METHODS

Study area: The study was conducted within Adamawa Region of Cameroon, which is a vast plateau situated between latitudes 6° N and 8°N and longitude 10°E and 16°E and stretches for about 62000 km at altitude varying between 900 and 1500m. The rainy season is about twice longer than the dry season; and is very humid (80% with an annual average 65%) (IRZ\GTZ, 1989). Temperatures are moderated by altitude with maxima in March (34°C) and 10°C during December-January (IRZ\GTZ, 1989). This main work was done in the Institute of Agricultural Research for Development (IRAD) Centre at Wakwa, situated South-East at 8-10 km of Ngaoundere, the regional capital. First research started in this centre in 1930, and in 1964 the centre associated with the French Institute for Animal Breeding and Veterinary Medicine (IEMVT). The centre covers about 2000Ha and has 500 animals. The criteria for choosing IRAD farm were that the scientific objectives of the centre include identification of suitable genotypes, crossbreeding exotics with local breeds, and dissemination of improved genetic material to farmers. Since 1974, more than 500 animals have been distributed to farmers. Cattle breeds in the centre include local Gudali and the trypano-tolerant shorthorns Namchi cattle breed. The animals graze natural pasture during the rainy season and are fed hay and supplementation rations include cotton seed cake sometimes during dry season. Although the centre has its paddocks, the animals are most of the time mixed with village animals from many farmers**.** All these animals are progenies of other animals reared since many years in this Research Centre or were purchased from the local markets for the purpose of rebuilding the cattle population of the Research Centre and were aged 2 to7 years. Some of these animals are not de-wormed on arrival but during the year, only tactical anthelmintic treatment is being applied because of economic reasons, and except for confirmed clinical cases. All the animals share the same pastures, and, in the evenings, they are kept together in the same paddock. Many of these animals are slaughtered for human consumption.

Recovery and processing of helminth parasites from study animals

Fourteen cattle consisting of eleven Gudali and three Namchi breeds belonging to IRAD were used for the study between February 2018 and January 2019. Postmortem examination technique was used for collecting parasites in the gastrointestinal tract (GIT) and for the estimation of the number of parasites in GIT since faecal egg and larvae counts do not always give a reliable indication of worm burden (Hendrix, 1998; Kassai, 1999). Each carcass was opened along the *linea alba*, and the liver was inspected for parasites. Ligatures were placed at the oesophagus and anus; and double ligatures between each part of gastrointestinal tract to avoid mix-up of the contents (Wyman, 2006). Eleven of the gastrointestinal tracts studied were from the local zebu Gudali breed, and three from the *Boss taurus* Namchi breed. Breed, age, and sex of each cattle were recorded. Necropsies were conducted on carcasses within 12 hours of slaughter. Given the variability in stomach content, the dissected stomach mass, and the rest of the gastrointestinal tract (GIT) were evacuated for immediate examination. The rumen and reticulum were opened, and their contents carefully removed. Helminth parasites present in the stomach were separated from stomach contents for subsequent identification. When these processes were not finished at the same day, the entire GIT and liver were transferred immediately to an 18°C freezer.

Frozen GIT were later defrosted at room temperature (20°C–30°C) overnight. The various organs were separated from each other and from ligaments and were placed individually in shallow plastic trays. The rest of the digestive tract was divided into omasum, abomasum, small intestine, and large intestine. Abomasum, small intestine, and large intestine were then processed. They were first examined externally with a special focus on the mesenteric veins of the small intestine, which were inspected against a light source for Schistosomes (*Schistosoma bovis*, *S. matheei* and *S. curassoni*) (Dunn,1987). The omasum, abomasum, small and large intestines were then processed separately. Each part was opened along its length, its content washed out with tap water into bucket (Zajac and Conboy, 2012) . Intestines were laid out on a tray, the small and large intestines opened longitudinally and then the entire GI tract was cut into 25cm long sections. Additionally, intestinal walls were scraped withsoft forceps to remove mucous and food items and to enable the detection of parasites attached to the intestinal mucosa. Each section was methodically examined under a dissecting microscope (magnification varying from 1 to 4). The content of each organ washed out with tap water into a bucket was passed through a sieve with an aperture size of 200µm. The sieve residual was re-suspended in 5L of tap water. Of the thoroughly mixed suspension in 5L of tap water, an aliquot of 200ml was collected and labelled. Then formaldehyde was added to approximate 10% for preservation (v/v). All formalin-fixed samples were kept in ambient temperature until use. Before use, the aliquot was washed through a sieve with a mesh of 38µm openings to clear the samples. Using scribed glass Petri dish and a stereoscopic dissecting microscope, nematodes were recovered. Parasites imbedded within food particles were gently extracted. Parasites were counted and transferred to 70%

ethanol, then transferred later to labelled slides for subsequent identification under a photonic microscope (Zajac and Conboy, 2012 ; Taylor et *al.*, 2016).

The bile ducts in the liver were opened and viable parasites removed and placed en 70% alcohol. Five strips, each approximately 10mm thick were removed from 5 places over the entire width of the liver and placed in a plastic jar with normal saline. The strips represented one – fifth of the mass of the liver and thus represented one – fifth portion of the aliquot. All fat was removed from the abomasa and half of the abomasal mucosa scraped off. These mucosal scrapings were mixed with a pepsin solution (5g Pepsin from hog stomach (1200U/g), 8.5g NaCl, and 16ml HCI (37%), 11 H_2O $_{dest}$) in the ratio of 1 :3 and incubated for a minimum of 16 hours at 30ºC to 37ºC. The artificially digested mucosa was centrifuged, and the sediment examined for hypobiotic larvae (Zajac and Conboy, 2012 ; Taylor *et al.*, 2016). To identify the parasite species, sub-samples were further transfered by Pasteur pipette to microslides for examination under the microscope. The total number of larvae was calculated as follows: number in 10 ml sub-sample x 20 = total abomasal larval count to estimate the total burden (Wymann *et al.*,2006). The small and large intestines were not incubated (T0756e04.htm. FAOfiles).

Identification of helminthes parasites recovered

Parasites were identified based on morphological characteristics. Nematodes were cleared in lactophenol prior to identification (Zajac and Conboy, 2012; Taylor *et al.*, 2016). Identification of nematodes was made based on characteristics outlined for genus (females) and species (males) in (Zajac and Conboy, 2012; Taylor *et al.*, 2016). The keys used in the basic identification for trematodes were from (Zajac and Conboy, 2012). If parasite number in a sample was high, only 20 male specimens of each similar looking group of parasites were examined and total numbers in the sample derived by extrapolation after multiplying by 25 to estimate the total burden (Wymann *et al.*, 2006). In addition, specimens of *Cooperia punctata*, *C. spatulata* and *C. pectinata* were identified on the basis of spicule morphology and bursa (Zajac and Conboy, 2012). Specimens of *Haemonchus* (*contortus*, *placei* and *similis*) were identified on the basis of spicule length, spicule, barbs (tip to hook distance), vulval flap, cervical papilla, and gubernaculum (Zarlenga *et al*., 2016), and morphology of bursa (Zajac and Conboy, 2012). Specimens of *Nematodirus spathiger* were identified on the basis of characteristics of the spicules and bursa (Zajac and Conboy, 2012). *Haemonchus similis* were distinguished from *H. contortus* and *H. placei* on the basis of spicular structure and length, and vulvar structure and tail length. Then *H. contortus* and *H. placei* were separated on the basis of spicule lengths (Zajac and Conboy, 2012). The specimens of *Ostertagia ostertagi* were identified on the basis of spicules and cervical papillae (Zajac and Conboy, 2012). The specimens of *Trichostrongylus axei* were identified on the basis of spicules (Ghasemikhah *et al*., 2011; Zajac and Conboy, 2012). Hookworms were identified by presence of a cutting plate (Zajac and Conboy, 2012). A simple key for identifying

common gastrointestinal parasites of cattle were also adapted from FAO techniques (T0756e04.htm. FAOfiles).

Data analysis

Descriptive statistic compared worm burdens regarding sex of worms, as well as breeds and sex of cattle. Analysis of variance (ANOVA) in general linear model on raw values was summarized as arithmetic means with standard errors of means. Mixed-model ANOVA was used to establish any relationship between worm burdens of commonest species with cattle breed and sex.

RESULTS

All the necropsies' animals were old. They showed their eight teeth. Based on adult worms recovered from necropsies of 14 animals slaughtered, 100 % percent of cattle harboured helminth parasites within their GI tract**.** In nematodes, total body lengths of both sexes, cervical papillaes in both sex , spicule types and lengths in males for bursate nematodes, shape of tail in females were found to be a discriminant character to distinguish between most populations (Lichtenfels *et al*., 1988; Zajac and Conboy, 2012; Taylor *et al.,* 2016) The cattle parasites that have been identified have so far been described by many authors among which Cholet *et al*.(2000) and Ngole *et al*.(2003) in Central Africa (Cameroon), Kaufman and Pfister(1990) and Zinstag *et al*.(1997) in West Africa .

The absence of *Toxocara vitulorum, Moniezia spp and Strongyloides papillosus* may be due to the fact that samples were only collected from old animals. However, in another calf that died in another herd, *Toxocara vitulorum* was the cause with the obstruction of the small intestine, confirming its presence in Adamawa region in Cameroon. As expected, infection with *Strongyloides papillosus* is confined to calves less than eight months of age due to their immature immune status (Pandey *et al.* 1993; Vassilev, 1999). In fact healthy calves are normally not slaughtered, therefore, no data were collected from them.

Nine species of Nematodes (*Haemonchus contortus, Cooperia* species, *Trichostrongylus axei, Trichuris globulosa, Oesophagostomum radiatum, Nematodirus* species, *Ostertagia ostertagi, Dictyocaulus viviparus, Setaria labiatopapillosa*), two of trematodes and one cestode were recovered from necropsied cattle. The two Trematodes were *Calicophoron microbothrium* and *Fasciola gigantica* while the cestode was Echinococcus *granulosus* (Hydatid cyst) (Table 1). Total worm burden was 42252 but *Haemonchus contortus, Cooperia pectinata, Cooperia punctata* and *Trichostrongylus axei* were the commonest infecting the cattle. Relative percentage, average and range of worm burden of each nematode species were also presented in Table 1.

Table 1. Helminth parasites infecting cattle in the study area

					Worm	burden	
Helminth Parasites	**Phylum**	**Class.**	**Family**	**Number recovered**	**%**	**Mean**	**Range**
Haemonchus contortus	Nematoda	Secernentea	Trychostrongylidea	17320	41.00	1378.57	0-5320
***Cooperia* species**	Nematoda	Secernentea	Cooperidae	16120	38.15	1151.42	0-2380
Trichostrongylus axis	Nematoda	Secernentea	Trychostrongylidea	4000	9.46	585.71	0-800
Dictyocaulus viviparus	Nematoda	Secernentea	Dictyocaulidae	5	0.012	0.35	0-5
Oesophagostomum radiatum	Nematoda	Secernentea	Strongylidea	1420	3.36	94.28	0-1100
***Nematodirus* sp**	Nematoda	Secernentea	Molineidae	380	0.9	27.14	0-80
Ostertagia ostertagi	Nematoda	Secernentea	Trychostrongylidea	220	0.52	15.71	0-80
Setaria labiatopapillosa	Nematoda	Secernentea	Onchocercidae	2	0.004	0.14	0-2
Trichuris globulosa	Nematoda	Adenophorea	Trichuridea	2680	6.34	190	0-2760
Fasciola gigantica	Platyhelminthes	Trematoda	Fasciolidae	50	0.12	3.57	0-10
Calicophoron microbothrium	Platyhelminthes	Trematoda	Paramphistomatidae	54	0.13	3.14	0-12
Echinococcus granulosus	Platyhelminthes	Cestoda	Taeniidae	1	0.002	0.07	0-1

Total worm burden according to species and sex of worms (Table 2) indicated that female worms 28503 (67.46%) generally established more than males worms 13644 (32.29%) while hermaphroditic worms (Trematodes and Cestode) accounted for only 105(0.25%) of the total worm burden. A t-test on within within subject effects showed that female worms (χ = 2594.35 ± 3790.38 N=14) had the tendency to establish significantly($P<0.05$) more than males (χ =423.64± 1421.45 N=14) in breeds of cattle.

Table 2. Total worm burdens according to species and sex of worms recovered from cattle

Species	Sex of the worm				
	Hermaphrodite	Male	Female	Total	%
Haemonchus contortus	-	7010	10310	17320	40.99
Cooperia spp.	-	-	12072	12072	28.57
Cooperia pectinate	-	2874	-	2874	2.78
Cooperia punctata	-	1174	-	1174	6.80
Trichostrongylus axis	-	1 486	2514	4000	9.47
Trichurius globulosa	-	860	1820	2680	6.34
Oesophagostomum radiatum	-	140	1280	1420	3.36
Nematodirus spp.	-	100	280	380	0.90
Ostertagia ostertagi	-	-	220	220	0.52
Calicophoron microbothrium	54	-	-	54	0.13
Fasciola gigantica	50	-	-	50	0.118
Dictyocaulus viviparus	-	-	5	5	0.012
Setaria labiatipapillosa	-	-	2	2	0.005
Echinococcus granulolus	1	-	-	1	0.002
Total (No.)	105	13644	28503	42252	
Percentage (%)	0.25	32.29	67.46	100.0	100.0

***t* = 2.166, P= 0.049**

Total worm burdens (42252) recovered from cattle necropsied show that the 11 Gudali breed accounted for 32280 or average of 2935 worm burden per animal, while the 3 Namchi breed harboured a total of 9992 worms or an average *worm* burden of 3331 per animal of that breed. (Table 3 ,4). The average worm burden per cattle necropsied was 3018.

Tests of within subjects effects showed that worms had the tendency to establish in the Gudali breed (2304.28± 3365.53 N=14) than in the Namchi breed (χ = 713.71± 2497.12 N=14) (Table 3 ,4)**.** However, analysis of variance using t-test shows that there was no significant (P> 0.05) difference of establishment of worms with respect to breed

Table 3: Total worm burdens according to breed of the cattle

	Breed	
Cattle Id	**Namchi**	**Gudali**
1	600	-
2	20	-
3	9372	-
4	-	5331
5	-	5301
6	-	90
7	-	600
8	-	1400
9	-	201
10	-	1445
11	-	3468
12	-	1950
13	-	12024
14	-	450
Sub-total	9992	32260
%	23.64	76.36
Sub-average worm burden	3331	2935
Total		42252
Total average		3018

(χ = 1590.57 ± 4593.98; t=1.295 df=13 P= 0.218)

Table 4: Total worm burdens according to species and breed of the cattle

Species	**Breed of the cattle**		
	Namchi	Gudali	Total
Haemonchus contortus		17320	17320
Cooperia spp.	9372	4000	13372
Cooperia pectinata	600	1500	2100
Cooperia punctata	20	668	668
Trichostrongylus axis	-	4000	4000
Trichurius globulosa	-	2660	2660
Oesophagostomum radiatum	-	1420	1420
Nematodirus spp.	-	380	380
Ostertagia ostertagi	-	220	220
Calicophoron microbothrium	-	54	54
Fasciola gigantica	-	50	50
Dictyocaulus viviparus		5	5
Setaria labiatopapillosa		2	2
Echinococcus granulolus		1	1

Total (No.)	9992	32280	42252
Percentage (%)	23.61	76.39	100
Average worm burden	3331	2935	3018

(χ = 1590.57 ± 4593.98; t=1.295 df=13 P= 0.218)

Total worm burden of 42252 according to sex of cattle (Table 5,6) indicated that the average worm burden per cattle necropsied was 3018.. The 12 females yielded a total of 36321 worms or an average burden of 3027 worms per female. On the other hand, the 2 necropsied bulls (male) contributed 5931 worm burden or an average of 2966 worms per bull(male). For the breeds, average worm burden was 3331 per Namchi cattle breed, with 600 per Namchi bull(male) and 4696 per Namchi cow (female). Similarly, average worm burden was 2933 per Gudali cattle breed, with 5331 per Gudali bull (male) and 2693 per Gudali cow(female). There was no significant (P>0.05) difference between males and females' animals in the two breeds, although this was at cut-off of significance (P= 0.083) (Table 5,6)

Table 5: Total worm burdens according to sex of the cattle

Cattle Id	**Sex**	
	Bull(male)	**Cow(female)**
1		
2		20
3		9372
4	5331	
5		5301
6		90
7		600
8		1400
9		201
10		1445
11		3468
12	600	1950
13		12024
14		450
Sub-total	5931	36321
%	14.037	85.96
Sub-average worm burden	2966	3027
Total		42252
Total average		3018

Effect of sex (χ = −2170.71±4330.68, t = −1875, df = 13, P= 0.083)

Table 6**:** Total worm burdens according to species and sex of the cattle

Species	**Sex of the cattle**		
	Male	Female	Total
Haemonchus contortus	5331	11989	17320
Cooperia spp.		13372	13372
Cooperia pectinate	600	1460	2O60
Cooperia punctata	-	688	688
Trichostrongylus axis	-	4000	4000
Trichurius globulosa	-	2680	2680
Oesophagostomum radiatum	-	1420	1420
Nematodirus spp.	-	380	380
Ostertagia ostertagi	-	220	220
Calicophoron microbothrium	-	54	54
Fasciola gigantica	-	50	50
Dictyocaulus viviparus		5	5
Setaria labiatopapillosa	-	2	2
Echinococcus granulolus	-	1	1
Total (No.)	5931	36321	42252
Percentage (%)	14.037	85.96	100
Average worm	2966	3027	3018

Effect of sex (χ = −2170.71±4330.68, t = −1875, df = 13, P= 0.083)

DISCUSSION

Agricultural ruminants usually harbour mixed infections of gastrointestinal nematodes. A specific diagnosis is important because distinct species can differ significantly in their fecundity and pathogenicity. The presence of criptic species is assumed especially many within the family Trichostrongylid (Leiper, 1912), which contains many gastrointestinal parasites of ruminants. Comparative morphological approaches provided an initial pathway for clear identification of *H. contortus* (and other strongylids) and continue to constitute relatively efficient means to explore species and faunal diversity that are at the foundations of managing and mitigating impacts associated with parasites and parasitism(Zarlenga *et al* ., 2016). Precise designation of these parasites is based primarily on morphological and morphometric features (Lichtenfels and Khal, 1993; Drotz, 1995) although the increasing use of molecular morphological analysis, sometimes in combination with advanced morphological

analysis, have allowed the traditionnaly regarded species to be examined more closely (Wyrobisz *et al*., 2015). Cattle in this study harbour a variety of endoparasites, particularly gastrointestinal nematodes. These worm burdens (Table 1) indicate there were favourable climatic conditions for worm establishment and proliferation in the study area. However, Belem *et al.* (2001) observed Worm burden range of 0-42290 in cattle slaughtered at an abattoir in Sahel region of Burkina-Faso, despite unfavorable conditions for parasites caused by the long dry season. It was suggested (ILRI, 1997) that these results may not be representative of the general cattle population as abattoir animals are likely to have been drawn from those showing signs of ill- health or weakness and high levels of infection. In fact, parasite burden of sick animals may differ from parasite burden of apparently healthy ones; it may either be higher if immunity is suppressed or lower if sick animals were treated (Wymann, 2006). The difference in the range of worms observed in this study (Table 1) compared with the findings in Burkina-Faso could be due to the fact that the animals in this study belonged to a Research Centre and thus relatively healthier. However, Immunity to nematodes depends upon exposure and is likely to be more intense in more heavily infected individuals (Dineen 1963). This suggests that cattle in this region of Cameroon could be more immune to gastrointestinal nematodes due to their constant exposure to larvae because of the suitable environmental condition for their development and establishment. This would tend to reduce aggregation (Galvani, 2003). However, in natural, predominantly *T. circumcincta* nematode infection, differences in immunity due to exposure are likely to be relatively small and swamped by genetic variation in immune responsiveness (Strain *et al.,* 2002).

The cattle parasites that have been identified have so far been described by many authors among which Cholet *et al*. (2000) and Ngole *et al*. (2003) in Central Africa (Cameroon), Kaufman and Pfister(1990) and Zinstag *et al*.(1997) in West Africa.

The absence of *Toxocara vitulorum, Moniezia spp* and *Strongyloides papillosus* (Table 1) may be due to the fact that samples were only collected from old animals. In another calf that died in another herd, *Toxocara vitulorum* was the cause with the obstruction of the small intestine, confirming its presence in Adamawa region in Cameroon. As expected, infection with *Strongyloides papillosus* is confined to calves less than eight months of age due to their immature immune status (Pandey *et al.*,1993; Vassilev, 1999). In fact, healthy calves are normally not slaughtered, therefore, no data were collected from them.

The number of worms is skewed and over dispersed for all major species of sheep nematode (Stear *et al.,* 1998). It could be the case in cattle. In this study, 11 nematode species, 2 trematodes and 1 cestode were present but the predominant species *Haemonchus* spp, followed by *Cooperia* spp., and *Trichostrongylus axei* are the most important gastrointestinal nematodes infecting ruminants in subtropical and tropical environments. For example, report arrangements from a similar study on cattle in Mali stated that *C. punctata* followed by

H. contortus were the most predominant species (Zinsstag, 1998). These results show only a slight difference in terms of ranking of infections but a study from the Northwest Region in another agroecological zone of Cameroon reported that *H. contortus* and *Trichostrongylus* spp were the most important species found in sheep and goats (Ndamukong and Sewell, 1992). However, in a study in prevalence of gastrointestinal parasites in bovines in and around Junagadh (Gujarat) in India **among** all the recorded GI parasitic infections, *Buxtonella sulcata* was in higher proportion in all the breeds (Maharana *et al*.,2016). This suggests that although the causes of helminth parasitism of ruminant livestock are ubiquitous in all agro-climatic zones in the world, particularly in Cameroon with prevailing weather conditions that provide favourable condition for their survival parasite species in a particular area may not be the same in number in a particular host. There is considerable variation among regions and farms in husbandry.

Haematophagous *Haemonchus* species are economically important in ruminants. *Haemonchus contortus* mostly found in sheep and goats (Lichtenfels *et al*., 1994) can also infect bovids (Hoberg *et al*., 2004). Though *H. placei* is most commonly found in cattle (Lichtenfels *et al.*, 1994), few reports have been published on apparently pure infection of *H. contortus* infection in cattle (Hogg *et al*., 2010; Akkani *et al*., 2013; Chaudhry *et al*., 2014). However, *H. contortus* is the synonym of *H. placei* and there is increasing evidence that these are the single species of *H. contortus* with only strain adaptations for cattle and sheep (Taylor et al., 2016).

Cooperia species consist of very common duodenal parasites of ruminants. Each of the species may frequently infect the same host as is the case in this study**.** In the subtropics, the epidemiology is similar to that of *Haemonchus* though *Cooperia* does not have the same high biotic potential, which might explain the lower worm burden of the genus compared to *Haemonchus* in this study. However, in some tropical and subtropical areas, some species are responsible for severe enteritis in calves (Taylor *et al.*, 2007) as could be the case in this study. *Trichostrongylus axei* whose lower prevalence is observed in this study could be due its lower biotic potential. Some nematodes such as *Haemonchus contortus* produce several thousands of eggs daily, while others like *Trichostrongylus* and *Ostertagia*, produce only a few hundred (Taylor et al., 2016). The fact that worm burdens of *H. contortus* and *Cooperia* species per cattle in this study were higher than respective permissible maximums of >1500 and >5000-10000 (T0756e04.htm. FAOfiles) suggests that anthelmintic treatments should focus on these genera according to the tables of Rommel *et al*. (2000)

All necropsied animals were adults because of the presence of their eight teeth. Worm burdens of *Oesophagostomum radiatum* has been found to be low (Pfukenyi *et al*., 2007) as in this study probably because cattle develop a good immunity, partly due to age and partly to previous exposure so that the infection becomes primarily a problem in

weaned calves. It is well-known that in the field, cattle develop strong immunity after 8-12 months of age (Taylor *et al*., 2016). From this study other minor parasite species in terms of worm burdens are from the genera *Bunostomum, Nematodirus, Ostertagia* (Tables 1 and 2). Favourable conditions are not found in the tropics for the survival and transmission of *Ostertagia* and *Nematodirus* species (Levine 1980), so both parasites are of little significance in Cameroon and particularly in the study area. Since *Ostertagia* species have been reported from Ghana (Mungube *et al*., 2015), Mali (Gasbarre and Miller, 2000; Wymann, 2006), and Cameroon (Ntonifor *et al.,* 2013) it appears that tropical climate is becoming favourable to its development and survival.

Thread-necked worms *Nematodirus* spp. found in the present study were of extremely low burden (Table 2). Unlike most Strongylids, its larvae develop to the infective stage within the egg and ruminants are infected when they ingest the hatched infective larvae. This could explain the low prevalence of this worm in cattle in this area. *Bunostomum phlebotomum,* the cattle hookworm, is one of the larger nematodes of the small intestine of ruminants and postmortem examination of infected cattle often reveals hydrothorax and fluid within the pericardium. Older livestock frequently develop sufficient immunity to limit reinfection from this worm (Taylor *et al*., 2016). This could explain the lower prevalence of this species

Hermaphrodite worms (*Calicophoron microbothrium, Fasciola gigantica* and *Echinococcus granulosus*) accounted for insignificant number. The snails, which carry the larval stages of *F. gigantica*, are primarily aquatic and as a result the disease is associated with animals grazing on naturally or on artificially flooded areas or around permanent water channels or dams. Animals in this study Research Center are given water in the crush inside the Research Center, thus explaining why they were not heavily infected. *Calicophoron microbothrium* previously known as *Paramphistomum microbothrium* infections were also extremely low in this study. Cattle develop a strong immunity which is dependent on the presence of adult flukes. The target organ for the metacestodes, *Echinococcus granulosus* is the liver and the lungs. It is commonly known as a hydatid cyst of dogs and jackal (Boomker, 2014) while cattle are considered an Intermediate host. This study was conducted in a research centre where scavengers are kept off, and this could explain the lower prevalence of this worm species. *Trichuris globulosa* being the most frequently encountered in cattle (Vassilev, 1999) was low in this study. Important feature of this species is longevity of its eggs, which may still survive after 3 or 4 years, though less likely on pasture since they tend to be washed into the soil (Taylor *et al*., 2016). Perhaps this may explain the very low worm burden of this species.

Female worms in this study were more abundant than males, (Tables 2), suggesting the high prolificacy of the species (Urquhart *et al*., 1996) and multiple generations of nematodes. The egg count is determined by the number of adult female worms and their average fecundity. There is a density-dependent effect on fecundity as the number

of adult worms increases fecundity declines. Egg output is overdispersed in environment and determines the level of hosts infestation. Several different species of nematode contribute to the total egg count and the total egg count can be considered as the sum of the counts for each separate species. The species for which egg counts have been most comprehensively analysed is *T. circumcincta* (Bishop & Stear 2000). There was no significant(P> 0.05) difference of establishment of worms with respect to breed in this study. In a study in Prevalence of gastrointestinal parasites in bovines in and around Junagadh (Gujarat) in India using faecal samples, breed wise prevalence of GI parasitic infection was examined and the data was statistically non-significant ($p > 0.05$) but higher infection rates were recorded in non-descript breeds than pure breeds for both cattle and buffaloes . Among all the recorded GI parasitic infections, *Buxtonella sulcata* was in higher proportion in all the breeds . However, in a study of resistance of gastrointestinal parasites in small ruminants the evidence for both between- and within- breed genetic variation for resistance to gastrointestinal (GI) nematode parasites was reviewed. Baker(2011) observed that Red Maasai sheep and Small East African (SEA) goats are more resistant to GI parasites (predominantly *Haemonchus contortus*) than Dorper sheep and Galla goats. He concluded that much of the published research on breed characterisation for resistance suffers from poor experimental design. However, its conclusion is based on a 6-year study completed in coastal Kenya using heritability estimates with faecal samples. He concluded that this difference in heritability suggests that many centuries of natural selection have fixed most of the genes for resistance in the Red Maasai sheep. The resistant Red Maasai sheep and SEA goats were two to three times more productive than the susceptible Dorper sheep and Galla goats in the sub-humid coastal Kenya environment. The present study was a survey therefore, other studies should be carried out to determine the effect of breed to resistance to infections. However, variation in globule leucocyte numbers of accounts for about one-third of the variation in worm numbers (Stear *et al*., 1995) in sheep;for example, sheep with more globule leucocytes (discharged mast cells) have fewer adult *T. circumcincta* (Seaton *et al*.,1989; Stear *et al*. 1995) and it could also be the case with cattle. This parameter may need to be investigated to determine the difference in worm burden between Gudali and Namchi cattle breeds. However, resistance to a parasitic disease is strongly influenced by genetic variation in the host; the relatively low estimates of genetic variation in disease resistance may sometimes be a consequence of the difficulty in defining and accurately measuring resistance to disease (Stear *et al*.,2007).

Sex differences in parasite prevalence or intensity are commonly observed (Poulin, 1996). Sex wise females recorded higher infection rates than males but the difference being statistically non-significant ($P > 0.05$). These results were also found in another study in the Prevalence of gastrointestinal parasites in bovines in and around Junagadh (Gujarat) in India (Maharana *et al*., 2016)**.** Pregnant and lactating females are immunosuppressed and therefore more

susceptible to infection (Krishnan *et al.* , 1996; Lloyd, 1983). Apart from reproductive status, host immunity may also vary seasonally in relation to stress, food availability, photoperiod, etc. (Martin *et al*., 2008)

The absence of hypobiotic larvae in digesta examined in this study could mean that hypobiosis did not occur, which confirmed the report that worms in areas at altitudes of 900 to 1300m do not observe hypobiosis (Zinsstag, 2000), and Adamawa region of Cameroon has altitude varying between 900 and 2400m. However, for a better control of these worms, treatments should focus on all stages of their development. An important feature is that some larvae (i.e., *H.contortus, Ostertagia* spp) undergo hypobiosis in grazing ruminants, with the result that later development to adult worms from hypobiotic larvae will contaminate pastures with nematode eggs when environmental conditions later become favorable for development of pre-parasitic stages to infective larvae (Ikpeze, 2009). Since hypobiotic larvae are metabolically depressed, they may be less susceptible to some anthelmintic treatment. So, the choice of drugs to be used in a nematode control program will be influenced by the role of hypobiosis in specific nematode life cycles and by the susceptibility of hypobiotic larvae to the range of available anthelmintic (Ikpeze, 2009).

CONCLUSION

All the necropsies animals were old. They showed their eight teeth. These animals harbour considerable numbers of worms that may constitute an important source of infections for young animals of 0-4 years since it is known that clinical helminthosis occurs more frequently among the latter group of animals. This shows lack of immunity in these old animals compared to the same group of animals in temperate regions . The commonest helminth parasites recovered at necropsy of apparently healthy cattle were *Haemonchus*, *Cooperia* and *Trychostrongylus* species. Therefore, deworming should also take into account these species of worms in the choice of anthelmintics but also old animals to increase productivity and decrease pasture contamination but also to reduce mortality from other diseases. So, the choice of drugs to be used in a nematode control program will be influenced by the role of hypobiosis in specific nematode life cycles and by the susceptibility of hypobiotic larvae to the range of available anthelmintic. Strategic deworming and other anti-helminth treatment and grazing management will help to control parasitic helminth infections and re-infections in all ages of cattle and other domesticated ruminants in the area

Conflict of interest: The authors declare no competing interests

Authors' Contributions: ENJ designed the experiment and work plan which were approved by OIO while ENJ and MTK implemented field and bench works. IOO compiled, tabulated, and analysed data. ND and IOO interpreted results. ENJ, IOO and OIO prepared the initial manuscript. All authors read and approved final manuscript for publication.

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