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Experimental Studies on Gastro-Intestinal Nematode Infection; The Effects of Age on Clinical Observations and Haematological Changes Following *Haemonchus contortus* Infection in West African Dwarf (WAD) Goats

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Abstract: The effects of age on clinical observations and haematological changes following gastro-intestinal worm *Haemonchus contortus* was investigated experimentally in West African Dwarf (WAD) goats. 10 apparently healthy adults and 10 kid goats were experimentally infected orally with 750 infective larvae of *Haemonchus contortus* for a period of 5 weeks. The adults and kids were weighed, the blood and faecal samples were collected before and after infection. Student's t-test statistical analysis was used. The age had no effects on clinical observations on the infected WAD adults and kids. There were normal rectal temperature, normal respiratory rate, normal heart rate with muscular weakness, anorexia, intestinal distension by worms, weight loss, reduced carcass quality, reduced feed intake, diarrhea and anaemia. The observable anaemia was characterized by responsive normocytic-nomochromic anaemia. The age had effects on weight and erythrocytic values which were significantly different at $P<0.05$ and $P<0.01$ compared with the control. The age had no effects on erythrocytic indices, the total WBC and differential leucocyte counts were not significantly different at $P>0.05$ and $P>0.01$. The weight and erythrocytic values are directly proportional to the level of helminthes eggs in the faeces.

Key words: Goats · *Haemonchus contortus* Clinical · Haematological changes

INTRODUCTION

Africa is believed to posses a variety of parasites due to favourable climatic condition and availability of suitable hosts. The existence of these parasites has caused a lot of damage to livestock industry in term of attaining market weight and the cost of treatment [1]. Animal production in Nigeria is the best by a number of problems amongst which pasturage, diseases and other parasitic problems form a bulk; of all these parasitic problems, helminthosis and ticks infestation ranked high and lead to anaemia [2]. Anaemia is not a disease entity but commonly seen as a result of generalized disease process. This term is defined as an absolute decrease in the packed cell volume (PCV); red blood cell (RBC); and haemoglobin (HB) concentration below the normal values [3]. Haemonchosis is an important disease of sheep, goats and cattle in Africa leading to high mortality and high morbidity in lambs and kids [2] Considerable damage is caused by fourth-stage larvae (L_4) of abomasal parasites leading to reduced appetite, haematological changes and reduced digestive capability of the abomasun as reported by Hansen and Perry [4].

There are various types and classification of anaemia due to pathophysiology, morphology and bone marrow response that affect livestock [5] leading to undesirable clinical signs. These changes may result in mediated haemolytic anaemia, metabolic disease, depression or hypoproliferative, anaemia associated with organ or tissue disorders, Aplastic or hypoplstic anaemia, myeloproliferative disorder and anaemia associated with parasitic disease haemorrhagic and haemolytic anaemia [6] the most common ones which affect (WAD) goats in this environment, leading to high mortality and high morbidity.

Anosa [7] and Otesile *et al.* [8] reported that most important endoparasites (helminthes) include *Haemonchus contortus, Ostertagia ostergi,*

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Trichostrongylus spp, Galgeria Pachycelis, Fasciola gigantica and *Monienza expansa*. However, *Haemonchus contortus* and *Monienzia expansa* appear to be the most common in this environment.

Anosa [7] and Otesile *et al*. [8] reported highly significant correlation between parasite egg per gram faeces (egg) and haemoglobin levels in *Haemonchus contortus* infection in sheep. Hematological changes include eosinophilia and terminal death is usually preceded by weakness especially of the hindquarter. While the comprehensives aid the relevance of this symptomatology are never in doubt. The chronological sequence of the same would be more useful to the clinician in following the progress, prognosticating and counseling on gastro-intestinal nematode infection. This study aimed at determine the effects of age on clinical observations and haematological changes following *Haemonchus contortus* infection in West African Dwarf (WAD) adult and kid goats.

MATERIALS AND METHODS

Experimental Animals: 10 kids and 10 adult WAD goats were bought from Samanda Area of Ibadan. The age of kids ranged between 1½ –6 months while the adult ranged between 2-3 years as determined by dentition. The kids and adult goats were acclimatized and treated with Thiabendazole at 66mg/kg body weight and 20% L. A oxytetracycline at dosage rate of 1ml/10kg body weight before the commencement of the experiment at ward 1, Veterinary Teaching Hospital, University of Ibadan, Nigeria. The blood and faecal samples were taken and analyzed. The faecal samples were negative for faecal egg counts. Haematological values recorded to serve as control. The kids and adult goats were experimentally infected with *Haemonchus contortus* for a period of 5 week (5-week post infection). 1st reading served as control while 2nd and 3rd and 4th readings at 3rd, 4th and 5th week served as experimental period.

Experimental Design

Assessment of Live Weight Changes: The kids and adult were weighed before and after experimental period with Salter (Butcher) scale machine, model 253 of sensitivity 0.05 kg.

Blood Samples: Blood samples were collected 5 ml of the blood was dispense inside the bijou bottle with etylenediamine tetra acetate (EDTA) to prevent blood clotting and samples were taken to the laboratory inside the ice pack.

Faecal Samples: Fresh rectal fecal samples were collected with bijou bottle from kids and adults for faeceal egg counts as described by MAFF [9] and Hansen and Perry [4].

Larval Culture: The faeces obtained from adult female goats confirmed positive haemenchus was cultured to produce third stage (L_3) Larvae of *Haemonchus contortus* using Baermann apparatus as described by Hansen and Perry [4].

Larval Dose and Oral Infection: 5ml of suspension containing infective larvae (L_3) were suspended in distilled water equivalent to 750 larvae of *Haemonchus contortus* were administered orally into 10 kids and adult goats (each) at moderate infection described by Hansen and Perry [4].

Monitoring of the Clinical Signs: The Routine clinical examinations of the kids and adult were done, at the morning. These examinations included respiratory rate, pulse rate, rectal temperature, alertness, general demeanor nature of hair coat and changes in live weight. Physical examinations were carried out also on all the animals for other clinical signs and lesions.

Haematology: The blood was analyzed for packed cell volumes (PCV) red blood cell (RBC), Haemoglobin concentration (Hb) total and differential leucocytes counts erythrocytic indices: Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin Concentration (MCHC) and Mean Corpuscular Haemoglobin (MCH) as described by Schalm [10] and Jain [6].

Blood Staining Technique: It was done to determine the presence of haemoprotozoan parasites as described by Jain [6].

Determination of Helminthes Eggs: Modified Mc-master floatation method was used as described by MAFF [9].

Statistical Analysis: Data were analysed using student's t-test.

RESULTS

The results of clinical parameters were indicated in Table 1 and 2 while Table 3 and 4 showed the weight and haematological changes following *Haemonchus contortus* infection. The clinical observations include mean rectal temperature, mean respiratory rate and mean heart rate for

Table 1: Clinical parameters of kids following experimental infection with *Haemonchus contortus*

		Experimental period		
Clinical parameter	Control normal values	3rd week post infection	4th week post infection	5th week post infection
Mean rectal temperature	38.15±1.24	38.15±0.24	38.20±0.76	38.25±0.24
Mean respiratory rate	15.40±0.52	15.60±0.52	15.40±0.52	15.01±0.32
Mean heart rate (minutes)	92.50±2.64	91.40±0.974	91.00±1.05	90.70±0.82

Mean prepatent period observed: 14.60±0.07 Mean and standard deviation±shown

Table 2: Clinical parameters of adult goats following experimental infection with *Haemonchus contortus*

		Experimental period		
Clinical parameter	Control normal values	3rd week post infection	4th week post infection	5th week post infection
Mean rectal temperature	38.72±0.21	38.98±0.37	38.87±0.16	38.9±0.50
Mean respiratory rate	12.39±0.22	12.61±0.16	12.56±0.22	12.80±0.79
Mean heart rate (minutes)	77±1.61	76.6±1.20	77.40±1.28	77.70±1.19

Mean prepatent period observed: 14.20±0.4 Mean and standard deviation ± shown

Table 3: Weight and Haematological changes following *Haemonchus contortus* infection in WAD kids

		Experimental period		
Haematological values	Control normal values	3rd week post infection	4th week post infection	5th week post infection
Weight	7.45±1.21	5.10±1.30**	4.63±1.09**	3.79±0.97**
PCV (%)	26.30±1.12	22.85±3.64*	19.40±3.54**	17.45±4.40**
RBC (10^6/µl)	12.02±0.76	10.50±4.26*	8.42±1.98**	8.10±2.08**
Hb (g/dl)	9.48±0.97	8.31±1.30*	6.88±1.37*	6.65±1.31*
Total WBC/mm^3	1860±2993	10830±3069	14540±4968	17345±8899
MCHC (%)	35.83±3.39	36.43±1.54*	35.40±3.46*	38.72±3.06*
MCV (fl)	21.97±1.72	25.98±13.07*	23.83±5.26*	22.18±5.87*
MCH pg	7.87±0.98	9.48±5.12	8.40±1.96	8.38±1.90
Reticulocyte (%)	-	0.07±0.48	0.30±0.31	0.04±0.46
Lymphocyte (%)	42.6±17.9	30.6±7.04	38.10±19.15	41.10±11.90
Monocyte (%)	0.40±0.70	1.2±0.4	2.4±1.2	3.1±1.4
Eosinophil (%)	-	1.2±1.3	2.1±1.1	3.8±3.2
Basophil (%)	-	-	-	-
Neutrophil (%)	36.8±10.9	28.9±11.90	42.1±14.8	48.1±16.3
Epg	-	810±36.4	5570±436.4	14010±3624**

differ significantly at p<0.05 and p<0.01 * non significant at p>0.05 and p>0.01 mean and standard deviation ± shown

Table 4: Weight and Haematological changes following *Hemonchus contortus* infection in WAD adult goats

		Experimental period		
Haematological values	Control normal values	3rd week post infection	4th week post infection	5th week post infection
Weight	21.7±1.86	19.93±1.83**	19.11±1.89**	18.19±1.80**
PCV (%)	27.6±2.15	23.2±2.3*	20.65±2.67**	19.05±2.13**
RBC (10^6/µl)	12.48±0.83	10.9±0.95*	10.2±0.93**	9.89±0.85**
Hb (g/dl)	9.42±0.33	8.05±0.77*	7.82±0.64*	7.58±0.60*
Total WBC/mm^3	14042±1339	12425±2039	13035±2282	17653±6447
MCHC (%)	34.30±2.52	34.94±4.46*	38.28±3.91*	40.04±3.04*
MCV (fl)	22.17±1.63	21.33±1.90*	20.25±2.05*	19.30±1.59*
MCH pg	7.59±0.56	7.43±0.98	7.71±0.74	7.7±0.17
Reticulocyte (%)	-	0.07±0.30	0.13±0.11	0.26±0.17
Lymphocyte (%)	45.11±1.66	37.7±7.15	41.5±5.95	44.7±9.5
Monocyte (%)	1.9±1.45	0.6±0.11	2.1±1.04	3.7±1.1
Eosinophil (%)	-	1.6±11.08	3.1±1.7	5.1±2.55
Basophil (%)	-	-	-	-
Neutrophil (%)	49.2±5.86	30.9±7.33	33.1±8.43	37.4±11.8
Epg	-	1380±183.3	4740±1397	13000±4500**

Differ significantly at p<0.05 and p<0.01 * non significant at p>0.05 and p>0.01 mean and standard deviation ± shown

both adult and kid goats. *Haematological parameters* include erythrocytic values (PCV, RBC, HB), erythrocytic indices (MCH, MCHC and MCV) total leucocyte counts (WBC) and differential leucocytes along with the weight of the of experimental animals.

DISCUSSION

Preclinical Assessment: Generally, the kids and adult were clinically healthy with no abnormalities, mucous membrane were pink, they were eating well with smooth hair coat, therefore the erythrocytic values, erythrocytic indices, total WBC values and differential leucocyte counts were normal. Both Giemsa stained thin film microscopy and haematocrit techniques did not reveal the presence of any haemoparasites. Examination by floatation method of faecal samples from the kids and adult did not reveal any appreciable number of eggs.

Clinical Assessment: After infecting kids and adult with *Haemonchus contortus* for 5 weeks, the kids and adults were emanciated and lean with starring hair coat, watery faeces, inappetence with slight pale colour of mucous membrane of the eyes, mouth or vulva, the weight of the kids and adult were seriously affected with presence of helminthes eggs in the faeces. No significant differences were observed in the mean rectal temperature, mean respiratory rate and the mean heart rate of the experimental kids and adults as shown in Table 1 and 2, but there was significance differences in mean respiratory rate and mean heart rate as a result of age at $P<0.05$ and $P<0.01$ as reported by Jean [12], who reported that respiratory rate changes rapidly in a matter of minute and it is directly influenced by animal activities, the environmental condition and the size of the animal is related to the respiratory rate and it is inversely proportional to the volume of the animal. Moreover, heart rate is inversely proportional to the weight of the animal and this is observed directly by counting the heart bit through listening or by feeling with the index finger the pulse of the arteries at particular point of the body. Rectal temperature is reported by Jean [12] as the best indicator of good health of the animal and its variation above or below normal is a measure of the animal attitude to resist heart ship factors of the environment and in young animals the temperature is higher as 1-1.1% than that of adult. The observable normal temperature in both adults and kids mean that *Haemonchus contortus* did not elaborate any certain chemicals that can cause pyrexia or interfere with respiration and pulse rate. These clinical signs observed in WAD kids and adult agreed with those of previous workers [11, 12]. In the parastological aspect, Hansen and Perry [4] defined the preparent period in the helminthes infection as the time lapses between infection and the first appearance of eggs in faeces. It varies with the route of infection, sex, age and degree of acquired resistance of the host. In practical terms, it is a measure of time taken by the infective larvae (L_3) to migrate from the point of infection to the lowest intestine and reach sexual maturity and lay eggs. The mean prepatent period in this experiment is normal as describe by [4]. The haematological values revealed reduction in erythrocytic values, total WBC values, erythrocytic indices and differential leucocyte values were normal with the presence of reticulocytes as shown in Table 3 and 4.

The erythrocytic values, erythrocytic indices, total WBC and differential leucocyte counts obtained from preclinical assessment agreed with [13] in kids between 0-6 months of age and adult goats.

The mean values of PCV during 3rd week post infection did not differ significantly at $P>0.05$ and $P>0.01$ with normal, while PCV in the 4th and 5th week post infection were significantly different at $P<0.05$ and $P<0.01$ with normal in both kid and adult. This result has been confirmed with those obtained by Radostitis *et al.* [3].

The mean value of RBC during 3rd week post infection did not differ significantly at $P>0.05$ and $P>0.01$ with normal RBC while in the 4th and 5th week post infection RBC value was significantly different at $P<0.05$ and $P<0.01$ compared with normal. The 3rd, 4th and 5th week post infection RBC values were not significantly different from one another a $P>0.05$ and $P>0.01$ in both kid and adult. This result has been confirmed with those obtained by Radostitis *et al.* [3].The mean value of Hb concentration during 3rd, 4th and 5th week post infection did not significantly different at $P>0.05$ and $P>0.01$ compared with normal. The fall in erythrocytic values in an indicative of anaemia consequent to the sucking of the blood by *Haemonchus contortus*. The MCHC values during 3rd 4th and 5th week post infection did not differ significantly at $P>0.05$ and $P>0.01$. These values were normal as described by several researchers [13-16]. This is anaemia with normal MCHC value (Normocytic).The MCV values during 3rd, 4th and 5th week post infection and 5th week post infection treatment did not differ significantly at $P>0.05$ and $P>0.01$. These values were normal as described by Oduye [13] and Anosa [7] as normochromic anaemia. The weight in kg values during 3rd 4th and 5th post infection differ significantly at $P<0.05$

and P<0.01 with normal weight. This result has been confirmed with those obtained by Allonby and Urquhart [17] and Fabiyi [18] in both kids and adults. The helminthes eggs per gram faeces in 3rd week post infection indicated mild infection while in the 4th and 5th week post infection indicated moderate infection in both kid and adult as reported by Hansen and Perry [4] and Jain [7].

Eosinnophils in kids and adult in post infective period range between 1-4 and 1-5%, respectively but the normal values reported by Schalm *et al.* [19] ranged between (1-8%). But in the 5th week post infection eosinophil values ranged between (1-10%) was due to higher proportion of eosinophils in Nigeria goat due to parastitism as reported by Bamidele [11]. This experiment showed that helminthes and particularly haemonchosis infection affect the weight gain of an animal as well as erythrocytic values leading to responsive normocytic normochromic anaemia.

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