
Anthelmintic Activity of Ethanol Extract of *Pseudocedrela Kotschy* (Dry-Zone Cedar) against *Haemonchus Contortus* Infection in Sokoto Red Goat

Attah, D. Daniel, & Danladi Y. K

Department of Biological Sciences,
Kebbi State University of Science and Technology,
Aliero Nigeria
danielattah10@gmail.com

Adamu, Tukur

Department of Biological Sciences,
Usmanu Danfodiyo University, Sokoto
Nigeria,

Muhammad, A. Aminu

Faculty of Veterinary Medicine,
Department of Parasitology and Entomology,
Usmanu Danfodiyo University, Sokoto
Nigeria.

ABSTRACT

*The indiscriminate use of anthelmintics has resulted in the establishment of parasite resistance. Thus, this study aimed to evaluate the anthelmintic activity of *Pseudocedrela kotschi* (Meliaceae) extract on Sokoto red goat infected with *Haemonchus contortus* was evaluated. The plant material was extracted using 90% ethanol. Twenty five (25) to thirty (30) dewormed goats were artificially infected with approximately 2000 infective larvae of *H. contortus*. After confirmation of infection and determination of initial the egg per gram of faeces (EPG), the animals were divided into 6 groups (A, B, C, D, E and F) of 5 animals' each. Group A served as the positive control and was tested with a single dose (10mg/kg) of Albendazole. Graded doses of the ethanol extract of the plant (100mg/kg, 200mg/kg and 300mg/kg/body weight) were administered to groups B, C and D for four weeks which commenced on day 21 post-infection. Groups E and F served as the infected/untreated and uninfected/untreated controls respectively. Body weight (BW), EPG, and some haematological parameters (PCV, WBC, Hg and RBC) were determined in the goats prior to treatment and once a week, for a 4-week period. A significant reduction ($p<0.05$) in EPG at 200mg/kg and 400mg/kg concentrations were recorded among all the groups by Day 42 post-infection. The BW of the treated goats decreased significantly by Day 28 post infection compared with values recorded on Day 0 and elevated by Day 42 post infection following treatment with the plant extract. Similarly, a significant ($p<0.05$) decrease in values of the tested haematological parameters were recorded by Day 28 post infection and appreciated ($p<0.05$) by Day 42 post infection, which may be an indication that the phytoconstituents present in the plant have some healing effects on wounds and varicose ulcers caused by the activity of the blood sucking *H. contortus* and can improve the resistance of tissues to infection. The study has revealed that the ethanolic extract of *P.kotschy* exhibited anthelmintic potentials at the*

concentrations tested since a dose dependent reduction in EPG was observed post treatment. However, further studies on the toxicity and the establishment of adequate doses for other ruminant are required.

Keywords: *Pseudocedrela kotschy*; Anthelmintic; *Haemonchus contortus*

INTRODUCTION

Diseases caused by helminth parasites in small ruminants continue to be a major economic loss both in the tropics and subtropics (Wanzala, *et al.*, 2005). Among different types of helminths, nematodes are the most important, as far as their prevalence and adverse effects are concerned. They cause retarded growth, lowered productivity (Perry & Randolph, 1999), mortality and economic losses which adversely affect the livelihood of marginal farmers. *Haemonchus contortus* is the highly pathogenic nematode parasite of small ruminants capable of causing acute disease and high mortality (Perry & Randolph, 2002; Githiagia *et al.*, 2005).

The major control strategy adopted over the past five decades against this parasite is the repeated use of synthetic anthelmintics. However, literacy and or unfamiliarity with these drugs, resulting in incorrect usage and adaptation of the parasite to the drugs, is now strongly questioned because of the increasing development of resistance of the parasite to these compounds (Mwamaci *et al.*, 1995; Wayangu *et al.*, 1996). A practical solution to this is to develop effective drugs that are more cost-effective, efficient and available than the conventional medicines. This can rationally be approached through the study of plants with antiparasitic potentials.

For many decades, plants have been used for the treatment of parasitism and in many parts of the world they are still used in this purpose. The used of medicinal plants for prevention and treatment of gastro-intestinal parasitism has its origin in ethnoveterinary medicine (Athanasiadou *et al.*, 2007). In ethnoveterinary medicine, there seems to be a range of plants or plant extract suitable for treating almost every parasitic disease of livestock (Sawleha 2010). Considering the vast potentials of plants as source of anthelmintic drugs, the present study was undertaking to evaluate the potentials of ethanol extract of *Pseudocedrela kotschy* for anthelmintic activity in Sokoto red goat experimentally infected with *Haemonchus contortus* parasite.

MATERIALS AND METHODS

Plant Collection and extraction

Stem bark of *Pseudocedrela kotschy* was collected on 18th December, 2014 at Bedi in Zuru Local Government Area of Kebbi State Nigeria. The plant was identified and authenticated by a team of taxonomists of the Botany Unit, Usmanu Danfodiyo University Sokoto, Nigeria. The plant part was shade-dried at room temperature for 14 days and then powdered using mortar and pestle. Two hundred grams (200g) of the fine powder was macerated in 1.5 liters of 96% ethanol in 3 cycles using soxhlet extractor. The crude extract was filtered through a filter paper (Whatman No 1). The filtrate was concentrated and dried into semi-solid substance in a rotary vacuum evaporator at 30°C. The filtrate was weighed, labeled and kept in bottles in a refrigerator until use.

Phyto-chemical Analysis of the Candidate Plants

The methods of Trease & Evans 1989 and Soforowa 1993 were used to detect the presence of difference chemical constituents. Secondary metabolites tested for include: tannins, flavonoids, saponins, alkaloids, and carbohydrates, reducing sugars, anthroquiones and steroids/triterpenes, glycosides.

Experimental Animals

The research was conducted using Sokoto red goat of both sexes. Ages of the animals was estimated to be sure that only animals between the ages of 6-18 months were used in the study (Chibuzo 2006). The experimental animals were maintained and housed on concrete floors at the small ruminant pens of the Veterinary Teaching Hospital, Usmanu Danfodiyo University, Sokoto Nigeria where they were fed on groundnut husk, bean hay and water provided *at-libitum*. During the two weeks acclimatization period, the animals were given the following; Ivermectin (0.2mg/kg/bw) against endo and ectoparasites, Tetracycline (10mg/kg/bw) against blood rickettsial organisms and *Diminazine aceturate* (7mg/kg/bw) against piroplasms as recommended by the manufacturers of the respective drugs. At the end of this period, the animals were screened for the presence of gastrointestinal parasites as well as blood parasites. All experiments were carried out in accordance with international guidelines for the use of animals for biomedical research and welfare.

Source of Experimental Parasite Stock

H. contortus was used as the experimental parasite in this study. Gravid female *H. contortus* were harvested from the abomasums of freshly slaughtered sheep from Sokoto Metropolitan Abattoir. The abomasums were opened using a sharp knife and the contents evacuated into a bowl containing physiological saline solution. The ingesta were washed through using a sieve of 100 micrometer mesh size in several changes of normal saline (Egualé & Giday 2009). An aliquot was transferred into a Petri dish and examined under stereoscopic microscope. Gravid female *H. contortus* was identified using the identification key (Hansen & Perry 1994). The parasites were separated into a Petri dish and transferred into a mortar and gently grounded using a pestle to release the eggs. The mixture was centrifuged at 2500 rpm for ten minutes and clean eggs were harvested.

Culture and Recovery of Infective Larvae (L3)

Cattle faeces was sterilized at a temperature of 100⁰C for 30 minutes and allowed to cool before use. Clean eggs obtained from the procedure described in section 2.5 above, were mixed with the sterilized cattle faeces adding water until the mixture was homogenized. The mixture (sterilized faeces and parasite eggs) was placed in clean universal bottles. The faeces were pressed using pestle to exclude air spaces. The bottle covers were perforated to allow air in the mixtures and closed tightly and kept in dark closet under room temperature (35-40⁰C) for a period of 14 days (Nwosu *et al.*, 2006). The bottles were checked every two days to be sure that the faeces remained moist. At the end of the 14 day period, the third stage larvae (L3) were harvested using Baermans technique (Soulsby 1982).

Experimental Infection of donor goats

Ten dewormed goats were infected orally with *H. contortus* infective larvae (L₃) obtained from the procedure described in section 2.6, in a 10ml syringe using sterile distilled water as a vehicle

given approximately 2000 L₃ per animal to serve as donor animals (Egualé *et al.*, 2007). At the end of 21 day post-infection, faecal samples from the donor goats were examined for the demonstration of typical strongyle eggs using saturated salt flotation technique (Jørgen & Brain, 1994). The goats that were found positive served as donor goats for subsequent cultures of the *H. contortus* L₃ larvae for the *in vivo* anthelmintic experiments. At the end of the experiment, all the goats were dewormed.

Experimental infection of goats and treatment with the plant extract

Faeces and blood samples of already dewormed goats of both sexes were screened to determine their initial egg per gram of faeces (EPG) and blood parameters. Each goat was assigned a number tag and its records maintained for each goat accordingly. The experimental goats were artificially infected with an estimated dose of 2000 *H. contortus* L₃ orally in 10ml syringe. Twenty one days post-infection, faecal and blood samples were taken from each tagged goat to determine the establishment of infection and the post-infection EPG and blood parameters respectively. Only the positive animals and their records were used in the study except the uninfected/untreated control. The goats were then assigned to six groups (A, B, C, D and F) of five animals each. The concentrations of the ethanolic extract doses of the plant were prepared from the stock extract based on the individual weight of goats and administration of the extract, which commenced on day 21 post-infection, followed this order:

- Group A: received single dose of 10 mg/kg Albendazole and served as positive control,
- Group B: received 100mg/kg body weight of the extract,
- Group C: received 200mg/kg body weight,
- Group D: received 300mg/kg body weight,
- Group E: served as the infected/untreated control,
- Group F: remained the un-infected/untreated control.

Clinical changes in goats

Twenty one days after infection with the larvae, the animals were monitored weekly for four weeks for the study of blood parameters, egg per gram of faeces and body weight according to standard criteria. Faecal samples were collected per rectum every week and evaluated using the Mc Master technique for demonstration of the presence of infection and to provide a quantitative estimate of the egg output. At the end of the experiment, all the goats were dewormed

Statistical Analysis

Data obtained was expressed as means standard deviation (\pm SD). Analysis of Variance (ANOVA) was used to test the significance of difference between the results of the treated, untreated and control groups using a computer software package (SPSS version 20.1). The difference was considered significant at the conventional level of significance ($p < 0.05$).

RESULT

Phytochemical Analysis

Result of the phytochemical screening of the ethanolic extract of *P. kotschyi* revealed the presence of chemical compounds such as, flavonoids, saponins, glycosides, terpenes, steroids, alkaloids, tannins, reducing sugar and carbohydrates.

In vivo Anthelmintic Activity

Table 1 showed the mean EPG of goats infected with *H. contortus* and treated with graded doses of *P. kotschy* and their controls. In Animals treated with Albendazole, a peak mean EPG of $2,476 \pm 175.52$ recorded by day 21 declined significantly ($p < 0.05$) to 125 ± 25.00 by Day 35 and was completely eliminated by Day 42 post-infection.

In animals treated with 100mg/kg, the peak EPG recorded ($2,470 \pm 180.12$) by Day 21 post-infection declined (986 ± 109.46) significantly by Day 42 post infection. In the group treated with 200mg/kg of *P. kotschy*, a peak mean EPG ($2,486 \pm 174.31$) was encountered by Day 21 post-infection. Following treatment with the extract it declined ($p < 0.05$) to 343 ± 35.98 by Day 42 post-infection. Animals treated with 400mg/kg recorded a maximum faecal egg count reduction (128 ± 13.87) by Day 42 post-infection. In the infected control, a peak mean EPG of $2,484 \pm 357.12$ increased ($p < 0.05$) without abatement to $2,692 \pm 212.03$ by Day 42 post-infection, while no eggs were seen in the faeces of the uninfected control.

This result showed a significant ($p < 0.05$) reduction in faecal egg count of treated animals compared with the infected/untreated control. This is evident by a dose depended significant decline in the mean peak egg count of all treated animals by Day 42 post infection observed in this study. The reduction in faecal egg count were found to be highest in Albendazole treated animals compared with the animals treated with the plant extract, which signified that the control drug (Albendazole) is more effective than *P. kotschy* for the concentrations tested in this study.

Effect on mean body weight (kg) of goats

Variable weight changes were observed in the experimental animals as presented in Table 2. The Albendazole treated group gained weight just like the un-infected control. However, significant mean body weight decrease (14.02 ± 4.05 , 14.20 ± 3.98) was recorded for animals treated with 200mg/kg and 400mg/kg plant extract by day 28 post infection and appreciated (14.99 ± 3.90 , 15.28 ± 4.03) significantly ($p < 0.05$) by Day 42 post infection. On the other hand, the values in the uninfected control (Group F) remained fairly constant. The changes in weight observed in treated animals in this study were dose dependent.

Effects on some haematological parameters of goats

Results of the effect on PCV, Hb, RBC and WBC of the experimental goats are presented in Tables 3, 4, 5 and 6. Animals treated with the plant extract and Albendazole had a significant ($p < 0.05$) reduction in the PCV values by Day 28 post- infection. Following treatment, the values significantly appreciated by day 42 post infection. The mean peak PCV values (31.40 ± 2.40) of animals in the untreated group declined (22.20 ± 1.01) significantly, while those of the uninfected control remained fairly constant (Table 3).

In the same manner, both the plant extract and Albendazole treated animals had a significant ($p < 0.05$) decrease in Hb concentration by day 28 post-infection, which later increased significantly ($p < 0.05$) from days 35 to Day 42 post infection (Table 4). There was no significant difference ($p < 0.05$) between the Albendazole treated group and the groups treated with the plant extract (Table 4).

Similarly, a significant ($p < 0.05$) decrease in RBC values of the infected animals was also observed by Day 28 post-infection. However after treatment with the plant extract and Albendazole, significant ($p < 0.05$) increases in the values were recorded. Animals treated with 200mg/kg and 400mg/kg of the plant extract, differed significantly with the group treated with 100mg/kg (Table 5).

The effect on WBC in the experimental goats showed significant ($p < 0.05$) increased values by day 21 post-infection compared with Day 0. However, the values decreased significantly ($p < 0.05$) by Day 42 post infection almost near to their pre-infection values of Day 0. Decline in WBC values observed in this study differed significantly between Albendazole and plant extract treated groups. Similarly, the decrease differed significantly between all the treated groups and the infected/untreated control (Table 6).

Table 1: Effect of *P. kotschy* Ethanol extract on Mean Faecal Egg Count (EPG) in Goats Infected with *H. contortus*.

Group	Concentration	Mean faecal egg count (EPG)				
		Post- administration duration (days)				
		0	21	28	35	42
A	10 (mg/kg)	00±00	2476±175.52 ^a	385±12.29 ^b	125±25.00 ^c	00±00 ^d
B	100 (mg/kg)	00±00	2470±180.12 ^a	1982±58.00 ^b	1320±93.70 ^c	986±109.46 ^d
C	200 (mg/kg)	00±00	2486±174.31 ^a	1602±147.54 ^b	987±42.76 ^c	343±35.98 ^d
D	400 (mg/kg)	00±00	2487±177.12 ^a	1204±12.83 ^b	412±70.44 ^c	128±13.87 ^d
E	Infected control	00±00	2484±35412 ^b	2596±321.09 ^a	2698±318.82 ^a	2697±212.03 ^a
F	Uninfected control	00±00	00±.00 ^a	00±.0 ^a	00±00 ^a	00±00

Values are the mean values ± standard deviation of 5 goats.

Means with different letters column wise, differ significantly (p<0.05)

Table 2: Effect on Mean Body Weight (kg) of *H. contortus* Infected Goats before and after Treatment with the Ethanol Fraction of *P. kotschy*

Group	Concentration	Mean body weight (kg)				
		Post administration duration (days)				
		0	21	28	35	42
A	10 (mg/kg)	15.40±5.89 ^{ab}	15.20±5.38 ^b	15.30±5.50 ^b	15.50±5.56 ^a	15.95±5.71 ^a
B	100 (mg/kg)	15.40±3.20 ^a	14.51±3.60 ^a	14.40±3.84 ^a	14.31±3.32 ^a	14.33±3.47 ^b
C	200(mg/kg)	15.02±3.74 ^a	14.81±3.52 ^a	14.02±4.05 ^b	14.63±4.03 ^a	14.99±3.90 ^a
D	400 (mg/kg)	15.45±3.74 ^a	14.72±3.35 ^a	14.20±3.98 ^b	14.69±4.01 ^a	15.28±4.03 ^a
E	Infected control	15.80±3.70 ^a	14.86±3.92 ^a	13.40±3.72 ^b	13.32±3.70 ^b	13.01±3.89 ^b
F	Uninfected control	14.40±3.20 ^a	14.60±2.73 ^a	14.91±2.78 ^a	15.44±2.88 ^b	15.51±3.05 ^b

Values are the mean values ± standard deviation of 5 goats.

Means with different letters column wise, differ significantly (p<0.05)

Table 3: Mean Packed Cell Volume (PCV) (%) of Treated and Untreated Goats as Affected by Ethanol Fraction of *P. kotschy*.

Group	Concentration	Mean Packed Cell volume (PCV)				
		Post- administration duration (days)				
		0	21	28	35	42
A	10 (mg/kg)	31.80±1.30 ^a	30.01±1.14 ^b	28.44±1.30 ^c	29.98±1.84 ^b	31.72±0.84 ^a
B	100 (mg/kg)	31.20±1.93 ^a	29.41±1.14 ^a	25.60±1.79 ^b	26.22±1.81 ^b	26.98±0.59 ^b
C	200 (mg/kg)	32.01±1.60 ^a	28.85±1.02 ^b	26.01±1.00 ^c	28.71±2.08 ^b	30.65±3.07 ^a
D	400 (mg/kg)	32.11±1.70 ^a	28.40±1.41 ^b	27.60±1.40 ^b	28.80±2.21 ^b	31.99±0.85 ^a
E	Infected control	31.40±2.40 ^a	30.00±1.63 ^a	27.20±1.79 ^b	24.03±1.41 ^c	22.20±1.01 ^c
F	Uninfected control	32.21±1.92 ^a	32.22±1.73 ^a	33.07±1.09 ^a	33.47±0.89 ^a	33.49±0.89 ^a

Values are the mean values ± standard deviation of 5 goats.

Means with different letters column wise, differ significantly (p<0.05)

Table 4: Mean Haemoglobin (Hb) Concentration (g/dl) of Treated and Untreated Goats as Affected by Ethanol Fraction of *P. kotschy*.

Group	Concentration	Mean Haemoglobin (Hg) Concentration (g/dl)				
		Post- administration duration (days)				
		0	21	28	35	42
A	10 (mg/kg)	10.96±1.76 ^a	10.24±1.21 ^a	9.54±1.02 ^b	9.94±1.29 ^b	10.54±1.39 ^a
B	100 (mg/kg)	11.28±0.95 ^a	10.08±1.02 ^b	9.56±0.83 ^c	10.06±1.45 ^b	10.20±1.40 ^b
C	200 (mg/kg)	11.76±0.93 ^a	10.02±1.22 ^a	9.06±0.99 ^b	9.74±1.01 ^b	10.22±0.20 ^a
D	400 (mg/kg)	11.28±1.29 ^a	10.24±1.25 ^b	9.30±2.18 ^c	9.90±1.27 ^b	10.89±0.29 ^a
E	Infected control	11.60±0.63 ^a	10.78±0.65 ^b	9.20±1.41 ^c	8.60±1.03 ^d	7.90±1.27 ^e
F	Uninfected control	11.30±0.73 ^a	11.32±0.73 ^a	11.40±0.69 ^a	11.36±1.04 ^a	11.36±0.69 ^a

Values are the mean values ± standard deviation of 5 goats.

Means with different letters column wise, differ significantly (p<0.05)

Table 5: Mean Red Blood Count (RBC) ($\times 10^6 \text{mm}^3$) of Treated and Untreated Goats as Affected by ethanol extract of *P. kotschyi*.

Group	Concentration	Mean Red Blood Count ($\times 10^6 \text{mm}^3$)				
		Post- administration duration (days)				
		0	21	28	35	42
A	10 (mg/kg)	8.32 \pm 0.75 ^a	6.24 \pm 0.29 ^a	6.40 \pm 1.02 ^a	7.48 \pm 0.38 ^a	8.20 \pm 0.79 ^a
B	100 (mg/kg)	8.28 \pm 0.36 ^a	7.30 \pm 0.65 ^a	6.10 \pm 0.41 ^b	6.74 \pm 0.91 ^a	6.94 \pm 0.30 ^a
C	200 (mg/kg)	8.50 \pm 0.43 ^a	6.18 \pm 0.45 ^b	6.26 \pm 0.40 ^b	6.60 \pm 0.45 ^b	7.37 \pm 1.01 ^a
D	400 (mg/kg)	8.50 \pm 0.36 ^a	6.19 \pm 0.45 ^b	6.90 \pm 0.53 ^b	7.58 \pm 0.43 ^a	7.65 \pm 0.37 ^a
E	Inf/cont	8.30 \pm 0.33 ^a	6.18 \pm 0.49 ^b	4.48 \pm 0.59 ^c	3.65 \pm 1.84 ^c	3.03 \pm 0.38 ^c
F	Uninf/cont	8.58 \pm 0.49 ^a	8.68 \pm 0.51 ^a	8.76 \pm 0.67 ^a	8.78 \pm 0.86 ^a	8.74 \pm 0.69 ^a

Values are the mean values \pm standard deviation of 5 goats.

Means with different letters column wise, differ significantly ($p < 0.05$)

Table 6: Mean White Blood Count (WBC) ($\times 10^3$) of treated and untreated Goats as affected by ethanol fraction of *P. kotschyi*.

Group	Concentration	Mean White Blood Count ($\times 10^3$)				
		Post- administration duration (days)				
		0	21	28	35	42
A	10 (mg/kg)	10.76 \pm 0.64 ^a	15.22 \pm 1.80 ^b	7.30 \pm 1.04 ^c	7.28 \pm 1.58 ^c	9.20 \pm 0.98 ^d
B	100 (mg/kg)	11.14 \pm 0.89 ^a	13.18 \pm 1.01 ^b	6.62 \pm 0.90 ^c	6.92 \pm 0.97 ^c	8.72 \pm 0.87 ^d
C	200 (mg/kg)	10.04 \pm 1.25 ^a	14.72 \pm 0.94 ^b	7.96 \pm ^{c0.66}	9.13 \pm 0.75 ^a	11.63 \pm 0.60 ^e
D	400 (mg/kg)	10.28 \pm 1.28 ^a	14.82 \pm 0.80 ^b	7.98 \pm 0.90 ^c	9.42 \pm 0.75 ^a	11.98 \pm 1.00 ^e
E	Inf/cont	10.46 \pm 1.09 ^a	14.86 \pm 1.43 ^b	11.74 \pm 3.07 ^c	7.90 \pm 0.14 ^d	5.75 \pm 1.06 ^e
F	Uninf/cont	10.82 \pm 1.26 ^a	10.86 \pm 1.24 ^a	10.90 \pm 1.21 ^a	10.94 \pm 1.18 ^a	10.98 \pm 1.15 ^a

Values are the mean values \pm standard deviation of 5 goats.

Means with different letters column wise, differ significantly ($p < 0.05$)

DISCUSSION

The result of the present study indicated that the ethanolic extract of *P. kotschyi* could result in the mortality of adult *H. contortus*. The significant reduction in EPG in goats treated with the extract by Day 42 post-infection gave an indication of the plant's effectiveness in killing *H. contortus* parasite, although, none of the concentration caused 100% mortality. This result agrees with a finding (Emaruk & Olila 2006) who reported the anthelmintic effect of *P. kotschyi* on *Ascaris suum* *in vitro*. The decrease in EPG recorded in this study suggests an active principle that may have anthelmintic potentials. Phytochemical screening of the ethanolic extract of the experimental plant revealed the presence of tannins, anthraquinones, triterpenoids, glycosides, saponins, flavonoids, alkaloids and resins, which agrees which agrees with similar findings of (Otimenyin *et al.*, 2004; Alhassan *et al.*, 2014). Tannins which were present in the plant were reported to have inhibitory effect on gastrointestinal nematodes (Molan *et al.*, 2002). Two hypotheses to explain the effect of tannins against parasitic gastrointestinal nematodes in ruminants have been reported:

- i. That tannins might directly affect, by pharmacological process, the biology of the worms and consequently modulate the epidemiology of gastrointestinal nematode infection;
- ii. Alternatively, tannins may act indirectly, through the improvement of the host response against the worms. Because of their binding ability, tannins protect proteins of the ruminant, consequently, favouring the increase in intestinal protein/peptide flow and amino acid absorption. It is known that any increase in the metabolizable proteins favours the two components of the host response (resistance and resilience) to nematodes (Coop & Kyriazakis 2001; Athanaiadou & Kyriazakis 2004).

Based on the first hypothesis, a study conducted by Brunet *et al.* (2008) has put some insight on the interactions between tannins and the infective larvae. The data indicated that the presence of tannins disturbed the two early steps of nematode establishment, first the larval exsheathment and second, the penetration of the exsheathed larvae in the digestive mucosae. Another phytochemical found in the plant was saponins. The antiparasitic activity of various plants that contain saponins has been described (Chapagain *et al.*, 2008). The associated mechanism of action may be due to the destabilization of membranes and increased cell permeability (Frances *et al.*, 2002), because saponins consist of a sugar moiety linked to a hydrophobic aglycone (triterpenoid or steroid) and are characterized based on their ability to reduce the surface tension of water in addition to their detergent emulsifying properties (Botura *et al.*, 2011).

The loss of weight seen in this study is not surprising because usually haemonchosis have been reported to come with weight loss which is caused by different factors. One of which is the impairment of animal productivity through reduction in food intake, efficiency of absorption of food nutrients, thus disturbing protein metabolism and availability, reduces absorption and retention of minerals in the animal, consequently leading to anaemia and hypoproteinaemia on the metabolic reserves and erythropoietic potentials of the infected animals (Githiagia *et al.*, 2005; Shakya 2007). The significant elevation in body weight of the experimental animals by Day 42 post-infection may be due to the presence of the active principles of tannins, saponins and flavonoids in the plant sample. Tannins have healing effects on wounds and varicose ulcers and improve the resistance of tissues to infection (Igboko 1983; Anthanasiadou *et al.*, 2001). Saponins, being a principle with varied pharmacological effects including aiding in the absorption of nutrient, readily increases the permeability of the mucosal cells of the small

intestine, thereby facilitating the uptake of minerals to which the gastrointestinal tract would not normally be permeable (Frances et al., 2002; Johnson et al., 1986) and this could be partly responsible for the significant appreciable gain in weight following treatment of *P. kotschy* extract.

Administration of the ethanolic extract of *P. kotschy* to the experimental goats was associated with decrease in the values of PCV, RBC, Hb and WBC by Day 28 is similar to other works (Sujon et al., 2008; Camila et al., 2012). Significant reduction of these parameters could be due to the fact that anaemia is a significant feature of haemonchosis (Maria 2006). The haematophagus effect of the parasite is capable of removing about 0.05lm of blood per day by ingestion and seepage from lesions (Soulsby 1982). A sheep with 5000 *H. contortus* worms may lose about 250ml of blood daily (Abbott et al., 2009). In this study, the PCV, Hb and RBC values were observed to improve following treatment with the plant extract and Albendazole by Days 35 and 42 post-infection respectively, which agree with other findings (Botura et al., 2011). This improvement could be as a result of the presence of flavonoid that inhibit inflammation or possibly stimulate haemopoiesis. Active principles like flavonoids have been reported to have influence on arachidonic acid metabolism thus could have anti-inflammatory, antiallergic, antithrombotic or vasoprotective effects (Nainwal et al., 2011). Other reports have also shown the beneficial effects of flavonoids on blood capillaries which include helating metals thereby sparing ascorbate from oxidation, prolongation of epinephrine action by the inhabitation of O-methyl transferase, stimulation of pituitary-adrenal axis and acting on the aggregation of erythrocytes (Robbins 1973). The effects of these active principles may be an explanation for the rise in the values of PCV, RBC and Hb values following treatment with the plant extract. The rise in WBC value following treatment with the extract may be suggestive of the extract's effect on the immune system. Saponins have been known to act as immunostimulants, thereby justifying their use as adjuvant in vaccine development (Campell 1993; James & Peace 1998).

Conclusion

Although *P. kotschy* did not eliminate *H. contortus* completely as observed with the reference drug (Albendazole), this finding has revealed that the ethanolic extract of *P. kotschy* exhibited anthelmintic potentials at the concentrations tested. It is evident from this result that the plant extract can reduce the level of worm burden, improve the state of blood parameters as well as improve the body weight status of the treated animals. It is also expected that morbidity and mortality caused by helminthiasis may reduce in treated animals compared to the untreated. Therefore, the identification of novel promising anthelmintic plant extracts such as *P. kotschy* may contribute for the development of phytotherapeutic products that could be more cost effective and more accessible and provide a lower risk of resistance than the synthetic anthelmintics currently employed. The phytochemical screening of experimental plant revealed the presence of a wide range of phyto-constituents which include; tannins, saponins and flavonoids, some of which have been reported to possess anthelmintic activity. Based on the above conclusion, further research to isolate the active component(s) contained in the plant and establish the mechanism of action, and a need to conduct detailed toxicological studies of the extract are recommended.

Acknowledgement

The authors are grateful to the financial support received from Tertiary Education Trust Fund

(TETFUND), Nigeria. We are also thankful to the Department of Parasitology and Entomology, Faculty of Veterinary Medicine, Usmanu Danfodiyo University Sokoto, Nigeria for providing the laboratory facilities for this study.

REFERENCES

- Abbott, K.A., Talor, M. and Stubbings, L.A (2009). Sustainable worm control strategies for sheep, 3rd Ed. A technical Manual for veterinary Surgeons and Advisers. *Sustainable control of parasites in sheep* (SCOPS). Pp 1-54.
- Alhassan, M. A., Ibrahim, M. and Musa, I. A. (2014). Phytochemical screening and antimicrobial evaluation of stem bark of *Pseudocedrela kotschy* (Schweinf.) Herms. *British Journal of pharmaceutical Research*, **4**(16): 1934-1944.
- Athanasiadou, S. and Kyriazakis, I. (2004). Plant secondary metabolites: antiparasitic effects and their role in ruminant production system. *Proceedings of the Nutrition Society*, **63**: 631-639.
- Athanasiadou, S., Githiori, J. and Kyriazakis, I. (2007). Medicinal plants for helminth parasite control: facts and fictions. *Animal*, **1**(9): 1392-1400.
- Athanasiadou, S., Kyriazakis, I., Jackson, F., Coop, R. I. (2001). Direct anthelmintic effects of Condensed Tannins towards different gastrointestinal nematodes of sheep: in vitro and in vivo studies. *Veterinary parasitology* **99**, 205-219.
- Botura, M.B., Silva, C.D., Lima, H.G., Oliveira, J.V.A., Souza, T.S., Santos, J.D.G., Branco, A., Moreira, E.L.T., Almeida, M.A.O. and Batatinha, M.J.M. (2011). *In vivo* anthelmintic activity of an aqueous extract from sisal wastes (*Agave sisalana* Perr.) against gastrointestinal nematodes in goats. *Veterinary Parasitology*, **177**: 104-110.
- Brunet, S., Jackson, F. and Hoste, H. (2008). Effect of sainfion (*Onobrychis viciifolia*) extract and monomers of condensed tannins on the association of abomasal nematode larvae with fundic explants. *International journal of Parasitology*. **38**: 783-790.
- Camila, O.C., Ana, C.S.C., Fernando, C., Maysa, F., Luciana, G.B., Francisco, C.M.C., Marilia, P.S., Humberto, R.B., Alessandro, F.T.A (2012). The anthelmintic effect of plant extracts on *Haemonchus contortus* and *Strongyloides venezualensis*. *Veterinary Parasitology*, **183**: 260-268
- Campbell, J. B. (1993). *Saponin-Adjuvants: Theory and Application* (Eds DES Sterwart-TULL), Heinemann Inc. Toronto, London, New York, Pp 334.
- Chapagain, B.P., Sahara, V. and Wiesman, Z. (2008). Larvicidal activity of saponins from *Balanites aegyptiaca* callus against *Aedes aegypti* mosquito. *Bioresoures and Technology*, **99**:1165-1168.
- Chibuzo, G.A. (2006). *Ruminant Dissection Guide: a regional approach in goats*. Beth-Bekka Academic publication. Maiduguri. Pp. 59-60.
- Coop, R. L. & Kyriazaakis, I. (2001): Influence of host nutrition, development, and consequences of nematode parasitism. *Trends in Parasitology*, **17**:325-330.
- Eguale, T., Tilahun, G., Debella, A., Feleke, A. & Makonnem, E. (2007). In-vito and In-vivo anthelmintic activity of crude extracts of *Canadium sativum* against *Haemonchus contortus*. *Jourbal of pharmacology*. **110**(3):428-433
- Eguale, T. and Giday, M. (2009). *In vitro* anthelmintic activity of three medicinal plants against *Haemonchus contortus*. *International Journal of Green Pharmacy*, **3**: 29-34.
- Emaruk, E. and Olila, D. (2006). Ascaricidal Activity of some medicinal plants used by the Karimojong: A nomadic Community in Uganda. *Journal of Animal and Veterinary*

Advances, **5**(9): 724-728

- Frances, G., Kerem, Z., Makkar, H.P.S., Becker, K. (2002). The biological action of saponin in animal system: a review. *British Journal of Nutrition*. **88**(6): 587-605.
- Githigia, S. M., Thamsborg, S. M., Maingi, N. & Munyua, W. K. (2005): The epidemiology of gastrointestinal nematodes in goats in low potential areas of Thika District, Kenya. *Bulletin of Animal Health Production in Africa*. **53**: 5-12
- Hansen, J. & Perry, B. (1994): *The epidemiology, diagnosis and control of health parasites of ruminants: A handbook*. International Livestock Research Institute, Nairobi, pp171. ISBN. 9290557031.
- Igboko, D.O. (1983). Phytochemical and biological studies on some constituents of vernonia amyglalina (compositae) leaves. PhD Thesis, Department of Biochemistry, University of Ibadan, Nigeria. Pp 100-123.
- James, S.L. and Peace, E.J. (1998). The influence of adjuvant on induction of protective immunity by a nonliving vaccine against Schistosomiasis. *Journal of Immunology*. **140**: 2753.
- Johnson, I.T., Gee, J.M., Price, K., Curl, C., Fenwick, G.R., (1986). Influence of saponin on gut permeability and active nutrient transport in-vitro. *Journal of Nutrition*. **116**:2270-2277.
- Jørgen, H. and Brian, P. (1994). Epidemiology, Diagnosis and Control of Helminth Parasites of ruminants. International Livestock Centre for Africa. Addis Ababa, Ethiopia. Pp. 67. ISBN 92-9055-703-1.
- Maria L. L. (2006). *Haemonchus contortus* (Barber Pole Worm) infestation in goats. Alabama Cooperative Extension System. Retrieved August, 2013 from www.aces.edu/urbam: (accessed on 23th Aug. 2015).
- Molan, A.L., Wahorn, G.C and McNabb, W. C (2002). Effect of Condensed Tannins on egg hatching and larval development of *Trychostrongylus columbriformis* in vitro. *Veterinary Records*. **150**: 65-69.
- Nainwal, P., Dhamija, K. and Tripathi, S. (2011). Study of antihyperlipidemic effect on the juice of the fresh fruits of *Lagenaria siceraria*. *International Journal of Pharmacy and Pharmaceutical Sciences* **3**(1): 88-90.
- Nwosu, C.O., Iwuoha, C.L, Torru, C. and Mohammed, A. (2006). Prevalence of caprine strongyle infection and diagnostic efficacy of some media used for faecal culture and nematode larval recovery from goat faeces. *Animal Research International*, **3**(1): 419-421.
- Otimenyin, S.O., Uguru, M.O. and Atang, B.L. (2004). Ant-inflammatoiy and Analgesic Activities of *Ficus thonningii* and *Pseudocedrela kotschy* extracts. *Nigerian Journal of Pharmaceutical Research*, **3**(1): 82-85.
- Perry, B. D. & Randolph, T. F. (1999): Improving the assessment of the economic impact of parasitic diseases and their control in production animals. *Veterinary Parasitology*, **84**:145-168.
- Perry, B. D., Randolph, T. F., Mcdermott, J. J., Sones, K. R. and Thomson, P. K. (2002). *Investing in Animal Health Research to alleviate poverty*. ILRI, Nairobi Kenya. Pp 148. ISBN. 9291461083
- Robbins, R.C. (1973). Physiological effects of plant flavonoids. *Journal Chemistry and Pharmacology*, **4**:271-278.
- Sawleha, Q., Dixit, A.K. and Pooja, D. (2010). Use of medicinal plants to control *Haemonchus*

- contortus* infection in small ruminants. *Veterinary World*, **3**(11): 515-518.
- Shakya, K.P. (2007). Evaluation of selected immune response to *Haemonchus contortus* in Gulf coast native compared to Suffolk lambs. PhD Dissertation, Louisiana State University, United States. Pp 10-11.
- Sofowora, A. (1993): *Medicinal plants and traditional medicine in Africa, 2nd Ed.* Spectrum Books Ltd. Ibadan, Nigeria pp150.
- Souslby, E. J. L. (1982): *Helminths, Arthropods and protozoa of domestic animals. 7th Ed.* W. B. Saunders, London. ISBN 070200896962.
- Sujon, M.A., Mostofa, M., Jahan, M, S., Das, R.A. and Rob, S. (2008). Studies on medicinal plants against gastrointestinal nematodes of goats. *Bangladesh Journal of Veterinary Medicine*; **6**(2): 179-183.
- Trease, G. E. & Evans, W. C. (1989): *A text book on pharmacology, 13th ed.* Bailliere Tindall Ltd, London.
- Wanzala, W., Zezzin, K. H., Kyule, N. M., Baumann, M. P. O., Mathias, E. & Hassanali, A. (2005): Ethno-veterinary medicine: A critical review of its evolution, perception, understanding and the way forward. *Livestock Research for rural Development*, **17**(11): 234-243