EFFECT OF AMPROLIUM AND CRUDE EXTRACTS OF *PROSOPIS AFRICANA* LEAVES ON HAEMOGRAM AND SERUM BIOCHEMISTRY OF WEST AFRICAN DWARF GOATS INFECTED WITH COCCIDIAL OOCYSTS

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ABSTRACT
This study was conducted at Livestock Teaching and Research Farm of the Faculty of Agriculture, Shabu-Lafia Campus, Nasarawa State University Keffi, Narasawa state. The study was conducted to evaluate the effect of amprolium and crude extracts of Prosopis africana leaf on haemogram and serum biochemistry of West African dwarf goats infected with coccidial oocysts. Coccidiosis in small ruminant animals is one of the causes of morbidity and mortality in kids and lambs and is usually treated with organic anticoccidiosis drugs. Therefore, investigation on the efficacy and effect of amprolium and the crude extracts Prosopis afracana leaf extracts in the treatement of WAD kids. A total of 20 West African dwarf goats (WAD) of both sexes were used for the study, the goats were randomly divided into five groups comprising of four goats each. Haematological and serum biochemistry parameters were determined. Data collected was subjected to one way analysis of variance procedure in SPSS software version 22. This study showed that the leaf extracts is safe and did not cause any damage to the kidney and liver functions as CR, UR, ALT, AST and ALP levels were within the normal range when compared to negative control. The crude extracts of Prosopis africana have appreciable anticoccidial activities and could be used as an alternative anticoccidiosis in goats without causing any damage to the health of the animals.
Keywords: Amprolium, Prosopis africana, Goats, Haemogram, Serum biochemistry

INTRODUCTION

Coccidiosis in small ruminant animals is one of the causes of morbidity and mortality in kids and lambs and is usually treated with organic anticoccidiosis drugs substances (Costa *et al*., 2006; Jabbar *et al*., 2007). Various chemical anticoccidials are used in the control of coccidiosis (Felici *et al.,* 2021). However, the current efficacy of these drugs has been reduced because of development of resistant coccidial strains due to frequent use (Artho *et al.,* 2007; Behnke *et al.,* 2008). Barbour *et al*. (2015) have investigated the interest of medicinal herbs and their derivatives as substitutes for organic coccidiosis in the animal sector. These herbal compounds produce bioactive phytochemicals that are effective in controlling goats' coccidiosis while being safe, nutritious, and therapeutic for goat meat consumers (Alhotan and Abudabos, 2019; El-Shall *et al*., 2022).

MATERIALS AND METHODS

Study area

The study was conducted at Livestock Teaching and Research Farm of the Faculty of Agriculture, Shabu-Lafia Campus, Nasarawa State University Keffi. The Study area falls within Guinea Savannah, particularly on sandy-clay soil of Nigeria. It lies between latitude 8.5583653^0C North and longitude 8.5513946^0C East. It has a climate typical of the tropical zone because of its location. It has a temperature of mostly between 27^0C (81.25^0f) to 31^0C. Broken clouds: wind 2.11 m/s, humidity 77%, cloudiness 78, pressure 1015.0 hpa in September.

Plant collection and authentication

The leaves of Iron tree (*Prosopis africana)* were collected within Lafia Metropolis. The leaves were dusted and washed to remove dirts. The samples were taken to the herbarium of the Federal College of Forestry Jos, Plateau State where it was identified, authenticated and voucher number was given FHJ 304.

Processing and extraction procedure of the leaves

The processing and extraction procedure of the plant leaves were carried out at the Faculty of Agriculture Research laboratory 1 (Lab 1), Nasarawa State University Keffi. Fresh leaves samples were pounded, mixed with distilled water, squeeze and sieved to obtain Fresh Leaves Aqueous Extract

(FLAE). A second batch of fresh leaves was shade dried for two weeks and the dried leaves were pounded using mortar and pestle. It was then divided into two portions. The first portion was extracted with distilled water and obtained the dried leaves aqueous extract (DLAE) while the second portion was extracted using methanol. One thousand gram (1000g) of the dried leaves powder was packed into a clean plastic rubber container. 1.5 liter of methanol was added to the sample and allowed to remain for forty-eight (48) hours for extraction. The extract was collected using Soxhlet extractor and concentrated by drying in the oven at 45°C to remove all the remaining solvent. The extracts were stored at 4°C and continued using it until the end of the experiments

Phytochemical screening of the leave extract

Qualitative phytochemical analyses of the fresh and dried leaves aqueous extract as well as methanolic leaves extract (MLE) were carried out according to established standard methods (Harborne, 1984). These screening was usually based on visual observation of colour or precipitate formation after addition of specific reagents (Ammonia NH_3, Hydrochloric acid, Iron chloride, Water and Sulphuric acid) were the solvent used. Then subjected to qualitative phytochemical screening for: Saponins, Alkaloids, Tannins, Flavonoids and Glycosides.

Sources of experimental animals and their management

Apparently healthy West Africa Dwarf Goats of both sexes weighing between 9-12kg were sourced within Lafia Local government council and used for the experiment. The pens were disinfected two weeks prior the arrival of the goats. The goats were allowed to stay for two weeks for acclimatization on concrete floored in separate pens and were fed with crop residue and water *ad-libitum*.

Experimental design

The design of the experiment was completely randomized design, while a random sampling technique was considered for the assembling and distribution of the goats.

Experimental procedure

A total of 20 West Africa Dwarf goats of both sexes were used for the experiment. The Goats were randomly divided into five groups comprising four goats each. The goats were infected with mixed species of sporolated coccidial oocyst and all goats were monitored for clinical signs and laboratory confirmation of coccidiosis was done after seven days post innoculation before treatment commenced. Goats in Treatment 1 (T_1) were infected and left untreated (received normal water) to serve as negative control. Those in Treatment 2 (T_2) were infected and treated with Aprolium (Amprolium 250 WSP, Kepro® B.V., Holland) at 100mg/kg body weight to serve as positive control, Treatment 3 (T_3) goats were infected and treated with fresh leaves aqueous extract (FLAE), Treatment 4 (T_4) goats were infected and treated with dried leaves aqueous extract (DLAE) and Treatment 5 (T_5) goats were also infected and treated with methanolic leaves extract (MLE). T_3, T_4 and T_5 were each treated at a dose of 100mg/kg body weight (B.W.). The methanolic leaves extract was dissolved in about 10ml of water for easy administration. Blood was collected from each goat at day seven (7) post treatment from the jugular vein into Ethylene diamine tetra acetic (EDTA) and non EDTA (plain) containers for haematological and serum biochemistry analysis and taken to the laboratory for analyses immediately.

Data collection

Haematological parameters

All the blood samples were collected from the jugular vein into Ethylenediaminetetracetic acid (EDTA) container to prevent the blood from coagulating and complete blood counts were analysed for:
Packed cell volume (PCV), Red blood cell (RBC), White blood cell (WBC), Haemoglobin (Hb), Platelets, mean corpuscular volume (MCV), Mean corpuscular haemoglobin concentration (MCHC) and mean corpuscular haemoglobin (MCH). Using the method described by John and Arundhati, (2007); Monica, (2000).

Serum biochemistry parameters

All the blood samples were collected from the jugular vein of the Goats into non EDTA (plain) container to enable the blood coagulate and obtain serum. The serum was used for the analysis using Spectrum and Agappe Diagnostic test kits following the manufacturer's instructions (Young, 1990; Tanganelli *et al.,* 1982). And tests for: Total protein (TP), chlorine (CL), bicarbonate (HCO), albumin (ALB),

alkaline phosphatase (ALP), alanine aminotransferase (ALT), aspartate aminotransferase (AST), potassium (K) urea (UR), creatinine (CRE), sodium (NA) bilirubin total (BILT), and bilirubin direct (BILD) were determined using Spectrum (Egypt) and Agappe (Switzerland) Diagnostic Test kits following the manufacturer's instructions.

Statistical analysis

Data collected were subjected to one-way analysis of variance (ANOVA) of the SPSS statistical package version 22. Where significant differences exist, means were separated using Duncan Multiple Range Test (DMRT) of the same package, at the confident level of 5% level of significant.

RESULTS

Effects of iron tree (*Prosopis africana)* leaves extracts on complete blood counts of WAD goats infected with natural mixed species of coccidial oocyst.

Table 1 presents the effects of *prosopis africana* leaves extracts on complete blood counts of WAD goats infected with natural mixed species of coccidial oocyst. The extracts had no significant (P>0.0.5) effects on all the parameters except on red blood cells and platelets.

Table 1: Effects of *Prosopis africana* (iron tree) leaves extracts on the haemogram of WAD goats infected with natural mixed species of coccidial oocyst.

Treatment	PCV (%)	RBC×10$^{12/L}$	HB(g/dL)	WBC×10$^{9/L}$	MCV×10$^{-9/L}$	MCH×10^{12}	MCHC(g/l)	PLAT×10$^{9/L}$
Control(-ve)	32.75±2.78	11.55±0.64^{ab}	10.88±0.92	7.03±0.41	2.78±0.08	9.35±0.30	332.08±0.50	179.75±1.65^{a}
Positive(+ve) Ampr 100mg	27.25±0.63	10.08±0.35^{ab}	9.05±0.21	6.53±0.64	2.68±0.03	8.98±0.10	332.10±0.69	192.33±2.95^{ab}
FALE 100mg	26.00±0.82	9.40±0.21^{b}	8.63±0.27	5.65±0.46	2.73±0.48	9.13±0.11	331.73±0.61	206.65±10.17^{b}
DALE 100mg	27.00±1.08	10.10±0.42^{ab}	8.98±0.35	6.63±0.46	2.70±0.41	8.95±0.13	332.43±0.55	174.13±5.58^{a}
MLE100mg	32.50±4.91	12.95±1.98^{a}	10.80±1.63	6.33±0.27	2.53±0.18	8.43±0.61	332.33±0.61	184.08±3.78^{a}
LOS	NS	*	NS	NS	NS	NS	NS	*

Key: -ve= Negative, +ve= Positive, FALE= fresh aqueous leaves extract, DALE= Dried aqueous leaves extract, LOS= Level of significant and NS= Not significant, Ampr= Amprolium. Values in the same column with different letters superscripts are significantly different.

Effects of iron tree (*Prosopis africana)* leaves extracts on the percentage distribution of WBC of WAD goats infected with natural mixed species of coccidial oocyst.

The effects of *prosopis africana* leaves extracts on the percentage distribution (differential count) of WBC of WAD goats infected with natural mixed species of coccidial oocyst is presented in Table 2. From the result obtained, the extracts had significant (P<0.05) across all the parameters except basophils.

Table 2: Effects of *Prosopis africana* leaves extracts on the percentage distribution of leucocyte differentials count of WAD goats infected with natural mixed species of coccidial oocyst.

Treatment	Neutrophils (%)	Lymphacyst (%)	Monocysts (%)	Eosinphils (%)	Basophils (%)
Control (-ve)	57.25±2.56^{ab}	38.75±2.43b^{c}	2.75±0.85^{a}	1.25±0.49^{b}	0.00±0.00
Positive(+ve) Ampr 100mg	62.75±3.20^{c}	34.75±1.93^{c}	1.50±0.29^{a}	0.75±0.25^{ab}	0.00±0.00
FALE 100mg	39.50±3.20^{a}	57.00±2.68^{a}	2.50±0.65^{a}	1.00±0.58^{ab}	0.00±0.00
DALE 100mg	51.00±2.65^{b}	45.00±3.11^{b}	2.50±0.65^{a}	1.50±0.29^{b}	0.00±0.00
MLE 100mg	56.75±2.98b^{c}	37.75±2.39b^{c}	5.50±1.04^{b}	0.00±0.00^{a}	0.00±0.00
LOS	*	*	*	*	NS

Key: -ve= Negative, +ve= Positive, FALE= fresh aqueous leaves extract, DALE= Dried aqueous leaves extract, LOS= Level of significant and NS= Not significant, Ampr= Amprolium. Values in the same column with different superscripts are significantly different.

Effects of iron tree (*Prosopis africana)* leaves extracts on serum biochemistry (LFT and RFT) of WAD goats infected with natural mixed species of coccidial oocyst.

The effects of *prosopis africana* leaves extracts on serum biochemistry (LFT and RFT) of WAD goats infected with natural mixed species of coccidial oocyst is presented in Table 3. The result indicated that the extracts had no significant (P>0.05) effects across all the parameters except on ALT, ALB and ALP respectively.

Table 3: Effects of *Prosopis africana* leaves extracts on serum biochemistry of WAD goats infected with natural mixed species of coccidial oocyst.

Treatment	ALT (u/l)	AST(u/l)	TP (g/l)	ALB (g/l)	ALP (u/l)	BIL.D (mg/dl)	BIL.T (mg/dl)	Ur (mmo/l)	Cr (mmo/l)
Control (-ve)	4.50±0.65^{a}	13.00±1.08	64.50±1.85	43.25±0.48^{b}	141.00±8.38^{b}	0.00±0.00	0.00±0.00	1.42±0.21	1.08±0.31
Positive(+ve) Ampr 100mg	5.25±0.85^{a}	13.50±1.94	68.75±2.78	46.00±1.35^{b}	161.75±19.78^{b}	0.00±0.00	0.00±0.00	1.63±0.10	1.35±0.30
FALE 100mg	7.25±1.11^{a}	16.25±1.25	85.50±20.63	37.00±3.39^{a}	77.75±17.54^{a}	0.00±0.00	0.00±0.00	1.38±0.52	1.35±0.14
DALE 100mg	12.00±1.00^{b}	16.25±1.70	67.00±0.58	44.50±2.33^{b}	144.50±12.14^{b}	0.00±0.00	0.00±0.00	1.29±0.11	1.35±0.14
MLE100mg	11.75±2.67^{b}	16.25±1.60	64.25±0.48	47.25±1.55^{b}	315.50±6.08^{c}	0.00±0.00	0.00±0.00	1.20±0.34	1.43±0.17
LOS	*	NS	NS	*	*	NS	NS	NS	NS

Key: -ve= Negative, +ve= Positive, FALE= fresh aqueous leaves extract, DALE= Dried aqueous leaves extract, LOS= Level of significant and NS= Not significant. Values in the same column with different letters superscripts are significantly different.

Effects of *Prosopis africana* (iron tree) leaves extracts on serum electrolyte of WAD goats infected with natural mixed species of coccidial oocyst.

The effects of *Prosopis africana* leaves extracts on serum electrolyte of WAD goats infected with natural mixed species of coccidial oocyst is presented in Table 4. The result showed that the extracts had no significant (P>0.05) effects across all the parameters except on potassium.

Table 4: Effects of *Prosopis africana* leaves extracts on serum electrolyte of WAD goats infected with natural mixed species of coccidial oocyst.

Treatment	Na^{+} (mmol/l)	K^{+} (mmol/l)	HCo^{-}_{3} (mmol/l)	Cl^{-} (mmol/l)
Control (-ve)	179.00±35.12	2.17±0.35^{b}	22.55±1.24	78.50±10.43
Positive(+ve) Ampr 100mg	149.25±8.70	3.95±0.45^{a}	24.40±1.24	77.00±0.82
FALE 100mg	155.50±17.24	3.25±0.42^{ab}	24.40±1.19	77.00±0.91
DALE 100mg	161.75±11.76	3.20±0.32^{ab}	25.40±2.43	122.75±29.58
MLE100mg	146.25±12.65	3.43±0.19^{a}	26.23±2.14	99.50±15.58
LOS	NS	*	NS	NS

DISCUSSION

The PCV values obtained in this study was fairly higher than 30.0±2.1 (%) and 25.3±1.2 (%) for buck-kid and doe-kid of West African Dwarf Goats reported by Daramola *et al.* (2005). Tambuwal *et al.* (2002) reported that Red Sokoto goats have the PCV value of 25.7±3.1 (%) while Azab Abdel-maksoud (1999) reported the PCV value of 27.25±0.59 (%) for Baladi goats. Patterson *et al.* (1960) attributed increase in environmental temperature as the cause of increase in PCV values. The slight increase of PCV values in this study compared to other reports could be attributed to the differences in the environmental temperature of the study areas.

Red blood cells (RBC) values in this study agreed with 11.5±0.4 obtained for WAD goats (Daramola *et al*., 2005). However, Haemoglobin (Hb) in this study fell within the range of 9.8±0.3 for WAD goat and 11.4±1.6 for Red Sokoto goat respectively (Daramola *et al.,* 2005 and Tambuwal *et al*., 2002).
Total white blood cells (WBC) differential counts is lower than 13.5±0.8 (WBC $\times 10^{3\mu/l}$) reported by Daramola *et al*. (2005). The percentage distribution of WBC in this study revealed *Prosopis afracana* leaves extracts had significant (P≤0.05) effect on the distribution of WBC with the values slightly higher than 33.5±1.7 and 36.4±2.5 for neutrophils reported by Daramola *et al*. (2005) and 73.0±2.0 for buck-kid and 67.9±1.0 doe-kid lymphocytes, 0.2±0.1 monocytes and 0.3±0.1 eosinophils reported by Tambuwal *et al.* (2002). The high mean value of monocytes in the group that was treated with methanolic extracts could be attributed to the presence of coccidial and or other infection in the goats. The significant difference observed in eosinophils was due to the presence of parasites in the different groups of goats. The significant difference observed in the distribution of WBC in this study could be because, eosinophils and neutrophils increase in number during coccidiosis infection and all the components of WBC will also increase in number due to infections. In this study the percentage

distribution of neutrophils was slightly higher than lymphocytes contrary to the reports of Olusaya *et al.* (1976) that in goat lymphocytes are more than neutrophils in circulation. This may be because neutrophils are usually released in large number during last stage of infection than lymphocytes.

Mean corpuscular haemoglobin concentration (MCHC) values obtained in this study slightly higher than MCHC values 32.1±1.4 to 32.8±0.6 (%) for buck-kid and doe-kid reported by Daramola *et al*. (2005). The non-significant differences in the values of mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and Platelet in this study implies that the test materials do not have negative effects on them.

For serum biochemistry, the values obtained in this study for Alanine transaminase (ALT) agreed with ALT values 8.3±0.5 for doe-kid (µ/l) and 10.6±0.1 for buck-kid (µ/l) reported by Daramola*et al.* (2005). The non-significant values for aspartate aminotransferase (AST) was slightly lesser than 17.3±0.3 (µ/l) for buck-kids and 19.7±0.2 (µ/l) for doe-kid of WAD goats reported by Daramola *et al.* (2005).
The values obtained for Alanine transaminase, albumin and alkaline phosphatase in this study have differed significantly. However, all the values fall within the reference range, therefore *prosopis afracana* extracts having less harm to the goats.

The non-significant values across all the parameters of serum electrolyte except on potassium when compared to negative control, the significant difference that exist in pattassium showed the group that received Amprolium and methanolic extract having the best result, however, the values obtained in potassium all falls within the normal reference range. Therefore, the leaves extracts could be used by the animals.

CONCLUSION

The result revealed apparent safety of the extracts to be used since there was no harmful effects in the animal liver, kidney, blood and serum biochemistry parameters after used and the plants can be found almost everywhere in our locality and easy to process.

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