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Toxicity and Anticoccidial Efficacy of Some Plants Used in the Traditional Treatment of Avian Coccidiosis in Semi-arid Northeastern Nigeria

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ABSTRACT

The aqueous stem bark extracts of *Khaya senegalensis*, *Butyrospermum paradoxum* and *Anona senegalensis* were evaluated for toxicity and antococcidial effects in *in vitro* and *in vivo* studies. Phytochemical analysis revealed that tannins, terpenes, anthraquinones, phlobotannins, alkaloids, cardiac glycosides and steroids were present in various concentrations in some of the extracts. Acute and chronic toxicity of the extracts were, respectively evaluated by administering graded single intraperitoneal (50-1600 mg kg^{-1}) and prolonged (28 days) daily oral (100-2000 mg kg^{-1}) doses of the extracts to four-week old pullet chicks. These produced varying degrees of dose-dependent clinical manifestations and lesions in all the groups with mortalities only in the groups treated with *Anona senegalensis* and *Butyrospermum paradoxum*. *Butyrospermum paradoxum* was the most toxic of the three by intraperitoneal and oral routes; *Anona senegalensis* was toxic by parenteral route but relatively safe by oral route and *Khaya senegalensis* was relatively safe by both parenteral and oral routes. Extracts of *Khaya senegalensis* had no *in vitro* anti-sporulation effect while those of *Butyrospermum paradoxum* and *Anona senegalensis* produced significant effects that were concentration dependent and comparable to amprolium at 0.6 and 1.2 mg mL^{-1} concentrations. Oral treatment of pullet chicks infected with 120,000 sporulated *Eimeria* oocysts/chick with either the extracts or amprolium significantly reduced or eliminated faecal oocyst output and improved packed cell volume and live weight of the birds. These effects were highest with *Anona senegalensis* and *Khaya senegalensis*. The high toxicity of *Butyrospermum paradoxum* precludes its possible usefulness as an anticoccidial agent.

Key words: Plants, extracts, treatment, coccidiosis, avian

INTRODUCTION

Coccidiosis is a major disease problem in the poultry industry. Outbreaks usually result in enormous economic losses as a result of the associated morbidity and mortality (Oluyemi and Roberts, 2000). The disease is controlled mainly by hygiene and the use of chemical anticoccidial agents (Soulsby, 1982). However, the development of drug resistance by the causative parasites and the escalating cost of drug development have greatly reduced the commercial incentive to develop new chemical anticoccidials (Gordon and Jordan, 1982; Permin, 1998; Oluyemi and

Roberts, 2000). Consequently, the development of alternative, safer and environmentally friendly anticoccidial agents have become priority in most parts of the world (Youn and Noh, 2001).

In Nigeria several plants have been claimed traditionally to have medicinal value for the treatment of various ailments in both man and animals (Nwude and Ibrahim, 1980; Akinniyi and Sultanbawa, 1983). However, their efficacy and safety remain doubtful as only a few of these have been properly identified and documented (Mbaya *et al*., 2007; Nwosu *et al*., 2004, 2008). Previous studies have shown that many plants said to be traditionally useful in the treatment of human and animal ailments have scientifically been shown to be either very toxic or have no therapeutic effect. In most cases, their toxic actions have been attributed to the contained active principles or over dosage due to the absence of standard dosage system in herbal medicine (Onyeyili *et al*., 2000; Hashemi *et al*., 2008).

The semi-arid zone of Northeastern Nigeria is known to account for about 30% of the domesticated livestock and poultry in the country (Nwosu *et al*., 2007). In this zone, several plant extracts including the stem barks of *Butyrospermum paradoxum, Khaya senegalensis* and *Annona senegalensis* are used traditionally by the natives in the treatment of several ailments including avian coccidiosis. Although previous experimental studies have reported the therapeutic efficacy of the stem bark extract of *B. paradoxum* against trypanosome species (Mbaya *et al*., 2007) and *K. senegalensis* against nematode species (Onyeyili *et al*., 2000) the extracts also produced toxic effects manifested in various degrees of behavioural changes, morbidity and sometimes mortalities when administered intraperitoneally to rats. However, no previous studies have been conducted to assess their toxicity and anticoccidial efficacy in chickens. This study was therefore designed to evaluate the toxicity and anticocidial efficacy of these three plants (*Anona senegalensis*, *Butyrospermum paradoxum* and *Khaya senegalensis*) said to be traditionally useful in the control of avian coccidiosis in the region.

MATERIALS AND METHODS

Collection and processing of plant materials: The plants used for this study were *Khaya senegalensis, Butyrospermum paradoxum* and *Annona senegalensis* also, respectively known as *Kadanya, Madaci* and *Gwandan daji* in Hausa, the predominant language in the study area. The stem barks of the plants were collected in February 2004 from the respective trees in Maiduguri, the capital of Borno state and the largest urban centre in semi-arid Northeastern Nigeria. The identity of the plants was confirmed by a botanist in the Department of Biological Sciences of the University of Maiduguri, Nigeria, where voucher specimens of the plants were deposited.

The stem barks of the plants were dried under shade for 10 days at 8 h per day and then ground into powder using a pestle and mortar. The powdered extracts were sieved to remove excess coarse plant materials and then individually exhaustively soxhlet extracted with water for 8 h at 60°C (WHO, 1992; Onyeyili *et al*., 2001). The soluble extract was then concentrated in a conical flask placed in a water bath maintained overnight at 60°C. Thereafter, the concentrated extract now in a gel form was collected, weighed and stored at 4°C for later use in the study.

Experimental animals: A total of two hundred and twenty-five pullet chicks were used in this study. They were purchased at day-old (ECWA Rural Development Ltd., Jos, Nigeria) housed at the Experimental Animals House in the Parasitology Unit of the Department of Veterinary Microbiology and Parasitology, University of Maiduguri, Nigeria and maintained on standard feed (Vital Feed, Grand Cereals and Oil Mills, Bukuru, Nigeria) containing 14.5% crude protein, 7% fat,

7.2% crude fibre, 1% calcium and 68.1% carbohydrate. Water was provided *ad libitum*. The birds were four weeks old when they were used for the experimental studies.

Experimental drug: Amprolium (Amprolium 200 NTCOX 20%, Sam Pharmaceuticals Ltd., Nigeria) a commercially available anticoccidial drug for the routine treatment of avian coccidiosis (due to *Eimeria* species) in Nigeria was used to compare the anticoccidial effects of the plant extracts.

Collection and culture of *Eimeria* oocysts: The *Eimeria* oocysts used in this study were derived from a rural chicken suffering from natural clinical coccidiosis. Following evisceration at post mortem, the caeca and intestines were separated, sliced open longitudinally and their contents washed into a beaker using tap water. The washings were centrifuged and the sediment re-mixed with saturated sodium chlorine solution (NaCl) to float the oocysts (Anonymous, 1977). Thereafter, traces of salt and colouring matter were removed by washing the sediments several times with water through the process of centrifugation. The harvested oocysts were multiplied in three chicks following oral infection. The chicks were routinely monitored daily for the development of clinical coccidiosis and the presence of *Eimeria* oocysts in their faeces. Thereafter, pure cultures of *Eimeria* oocysts were obtained through floatation as earlier described. Oocysts were sporulated for 72 h in 2% potassium dichromate solution and the species identified using their morphology, sporulation time, predilection site and the pathological lesions produced in chicken (Anonymous, 1977; Soulsby, 1982). Faecal oocyst counts were determined by the modified McMaster technique using saturated sodium chloride solution as the floating medium (Anonymous, 1977). The oocysts were stored in 2% potassium dichromate solution that was changed weekly until used. The infecting oocysts consisted of a mixture of *Eimeria tenella* (26.85%), *E. acervulina* (61.54%) and *E. mivati* (11.61%).

Experimental procedures

Phytochemical analysis of the aqueous stem bark extracts: The stem bark of each plant was analysed for phytochemical constituents including tannins, saponins, alkaloids, anthraquinone derivatives, terpenes, steroids and cardiac glycosides as described by Sofowora (1984).

Determination of acute toxicity of the extracts: Thirty-five pullet chicks, separated into seven groups (A-H) of five chicks each were used to determine the acute toxicity of the aqueous stem bark extract of *K. senegalensis*. The birds in groups A-G were, respectively treated intraperitoneally with graded doses (50, 100, 200, 400, 800, 1200 and 1600 mg kg^{-1}) of the extract in distilled water while chicks in group H (Control) were given only distilled water equivalent to the largest volume of the extract by the same route. Two new sets of thirty-five pullet chicks each were used to repeat the same procedure using the extracts of *B. paradoxum* and *A. senegalensis*, respectively.

The birds were monitored during a 24 h period for signs of toxicity and death. Dead birds or those sacrificed after 24 h were subjected to necropsy for lesions. The median lethal dose (LD_{50}) was determined using the arithmetic method of Karber as modified by Aliu and Nwude (1982).

Determination of chronic toxicity of the extracts: Thirty pullet chicks divided into six groups (A-F) of five chicks each were used to determine the chronic toxicity of *K. senegalensis* in chicks. The birds in groups A-E were orally treated, respectively with various concentrations (100, 500, 1000, 1500, 2000 mg kg^{-1}) of the water extract of *K. senegalensis* daily for 28 days. Group F served

as control and was orally treated with distilled water equivalent to the largest volume of the extract administered. Two other sets of thirty pullet chicks each were used to repeat the experiment using *B. paradoxum* and *A. senegalensis*, respectively. Clinical signs, mortality and gross and histopathological findings were used to assess the effects of the various extracts on the chicks.

***In-vitro* anti-coccidial effect of the extracts:** The *in-vitro* anti-coccidial efficacy trials were conducted by observing the effect of the plant extracts on the sporulation of *Eimeria* oocysts. Fresh faecal samples were collected from infected birds and the oocyst counts determined (Anonymous, 1977). Various dilutions of the respective extracts of *K. senegalensis*, *B. paradoxum* or *A. senegalensis* (100, 250, 500, 1000, 1500 mg mL^{-1}) in distilled water were placed in separate Petri dishes labelled appropriately. A known number of oocysts (100 oocysts) were added to each Petri dish and the set up was left at ambient temperature and monitored every 6 h to observe sporulation of the oocysts over a 72 h period. The number and percentage sporulation of the oocysts were then determined and recorded. These were used to determine the percentage inhibition of oocyst sporulation. Similar observations were made for oocysts sporulated in Amprolium and potassium dichromate solutions which served as controls.

***In-vivo* anti-coccidial effect of the extracts:** A total of 30 chicks divided into 6 equal groups (A-F) were used for this study. Groups A-E were each orally infected with 120,000 sporulated oocysts (the infection dose was based on the results of an earlier pilot study). The birds were monitored daily for the presence of oocysts in the faeces to determine patency and oocyst counts. On the first day that oocysts were detected in the faeces, the birds in groups A, B and C were, respectively treated with 100 mg kg^{-1} body weight of the respective extracts of *K. senegalensis*, *B. paradoxum* and *A. senegalensis*. The birds in Group D were at the same time treated with amprolium at 10 mg kg^{-1} while group E remained untreated. All treatments were given orally. Birds in group F were uninfected and untreated and used as healthy controls.

All the birds in the various experimental groups were monitored daily for signs of disease including inappetance/anorexia, diarrhoea, bloody faeces, anaemia and death during a 28 days period. Faecal oocyst counts and live body weights of the birds were determined daily while the PCV was determined every 2 days for each experimental group. Dead birds and those humanely sacrificed at the end of the study were subjected to necropsy for lesions. The microhaematocrit method was used to determine the packed cell volume of the experimental birds (Coles, 1974) while the laboratory triple beam balance was used to determine live body weights.

Histopathology: In each case, tissue samples of the heart, lungs, lymph nodes, kidney, intestine, liver and spleen were obtained at necropsy, fixed in 10% buffered formalin, embedded in paraffin wax, sectioned at 5 μm thick and stained with Haematoxylin and Eosin as described by Drury and Wallington (1976). Stained tissue sections were examined under the light microscope for presence of lesions.

Data analysis: Data obtained from the study were summarized as Means±standard deviations and statistical differences between the means determined by the analysis of variance (ANOVA) and the paired students' 't' test at 5% level of significance (GraphPad Software Inc., 1998).

RESULTS

Phytochemical constituents: The results revealed that the three plants contained varying concentrations of tannins, terpenes and steroids (Table 1). However, only *A. senegalensis* contained saponins while *K. senegalensis* and *B. paradoxum* contained cardiac glycosides and alkaloids. Anthraquinones were present in only *A. senegalensis* and *B. paradoxum*.

Acute toxicity: Following the administration of graded doses of the extracts of *K. senegalensis* to the birds, no clinical signs of toxicity were observed in the groups treated with the 50-400 mg kg^{-1} concentrations of the extract; slight depression in the group given 800 mg kg^{-1} and depression, inappetance and limping in those treated with higher concentrations (1200 mg kg^{-1} and 1600 mg kg^{-1}). No deaths were noted in any of the treated groups (Table 2). Lesions were not observed in any of the treated groups except those given the 1,600 mg kg^{-1} concentration showed had areas of epithelial necrosis and haemorrhages of the intestinal mucosa, slight hepatic and splenic congestion. The LD_{50} of the extract could not be determined due to the absence of mortalities among the treated groups.

Table 1: Phytochemical constituents of *Khaya senegalensis*, *Annona senegalensis* and *Butyrospermum paradoxum*

Phtochemical constituems	*K. senegalensis*	*B. paradoxum*	*A. senegalensis*
Anthraquinones	-	+++	+++
Tannins	++	+	++
Phlobatanins	+	+	+
Carbohydrate	+	+	+
Saponins	-	-	++
Cardiac glycosides	+	++++	-
Terpenes	+	+	++
Steroids	+	++	++
Alkaloids	++	+++	-

-: Negative, +: Mildly positive, ++: Moderately positive, +++: Copiously positive

Table 2: Mortality pattern of four weeks old chicks following single or prolonged (28 days) administration of *K. senegalensis*, *B. paradoxum* and *A. senegalensis*

		Mortality pattern [(No. (%)]		
Group*	Dose (mg kg^{-1})	*K. senegalensis*	*B. paradoxum*	*A. senegalensis*
Single intraperitoneal administration				
A	50	0	0	0
B	100	0	0	0
C	200	0	0	3 (60)
D	400	0	0	4 (80)
E	800	0	1 (20)	5 (100)
F	1200	0	3 (60)	5 (100)
G	1600	0	4 (80)	5 (100)
Prolonged (28 days) oral administration				
A	100	0	0	0
B	500	0	1 (20)	0
C	1000	0	2 (40)	0
D	1500	0	2 (40)	0
E	2000	0	5 (100)	0

*Number of birds per group = 5

No signs of toxicity were observed in the birds given 50-200 mg kg^{-1} of *B. paradoxum*. However, depression, limping, drooping and anorexia were noted in the groups given 400 mg kg^{-1} or higher concentrations of the extract; the severity increasing with concentration of the extract. Deaths occurred only in the groups given 800 mg kg^{-1} or higher concentrations of the extract (Table 2). Lesions were noted only in the groups given 1200 and 1600 mg kg^{-1} and these included haemorrhages, epithelial and glandular tissue necrosis and villous atrophy in the intestine. The intraperitoneal LD_{50} of the extract was 1260 mg kg^{-1}.

Depression and weakness were noted in the birds given 50 and 100 mg kg^{-1} concentrations of *A. senegalensis*. At higher concentrations (200 mg kg^{-1} and above) there were varying degrees of depression, weakness, anorexia, droopiness and ruffled feathers; the severity increasing with concentration of the extract. Deaths were recorded only among the birds given 200 mg kg^{-1} or higher concentrations of the extract. The lesions observed were serosal haemorrhages and epithelial necrosis of the intestine in the birds given the 200 mg kg^{-1} concentration of the extract. At the 400 mg kg^{-1} and higher concentrations, there were varying degrees of muscular and hepatic congestion and the intestines had areas of haemorrhages, epithelial necrosis and villous atrophy. The intraperitoneal LD_{50} was 270 mg kg^{-1}.

Chronic toxicity: The groups treated with *K. senegalensis* did not manifest any morbidity or mortality except in the group treated with 2,000 mg kg^{-1} concentration of the extract that were slightly depressed. However, lesions of varying degrees of severity were recorded in the various groups treated with the extract. There was myocarditis and the intestines showed villous atrophy with epithelial and glandular necrosis. The kidneys showed interstitial nephritis with mononuclear cellular infiltration and renal coagulative and tubular necrosis. The liver had focal areas of necrosis and mononuclear cellular infiltration (Fig. 1) while the lungs were congested with some degree of serous pneumonia. The severity of the lesions was concentration dependent being most severe in those chicks given the highest concentration of the extract.

No clinical manifestations of toxicity were noted in the birds treated with various concentrations of *B. paradoxum* except in the groups given 1,500 and 2,000 mg kg^{-1} concentrations of the extract that became depressed, anorectic and reluctant to move 6-8 h after the first treatment. Mortalities (20-100%) and lesions were noted only in those groups treated with 500 mg kg^{-1} or higher concentrations of the extract (Table 2). The lesions in the heart included superficial mononuclear cellular infiltration of the epicardium (Fig. 2) while in the liver there were focal areas of hepatic congestion and necrosis with mononuclear cellular infiltration. In the intestine, there was glandular and epithelial necrosis with villous atrophy and mononuclear cellular infiltration of the mucosa (Fig. 3). The clinical signs and lesions were concentration dependent being most severe in the birds that received the highest concentration of the extract.

The extract of *A. senegalensis* did not produce any signs of toxicity in the chicks except for slight depression and inappetence noted 2-3 h after the first treatment with the 1,500 and 2,000 mg kg^{-1} concentrations of the extract. There were no deaths in any of the treated groups (Table 2). However, lesions were evident in those chicks treated with 1,000 mg kg^{-1} and higher concentrations of the extract and these included focal areas of hepatic and intestinal epithelial necrosis and haemorrhages as well as mononuclear cellular infiltration of the intestinal submucosa. There was tubular necrosis and mononuclear cellular infiltration into the interstitial tissues of the kidney (Fig. 4). These lesions were concentration dependent being most severe in the group given the highest concentration of the extract.

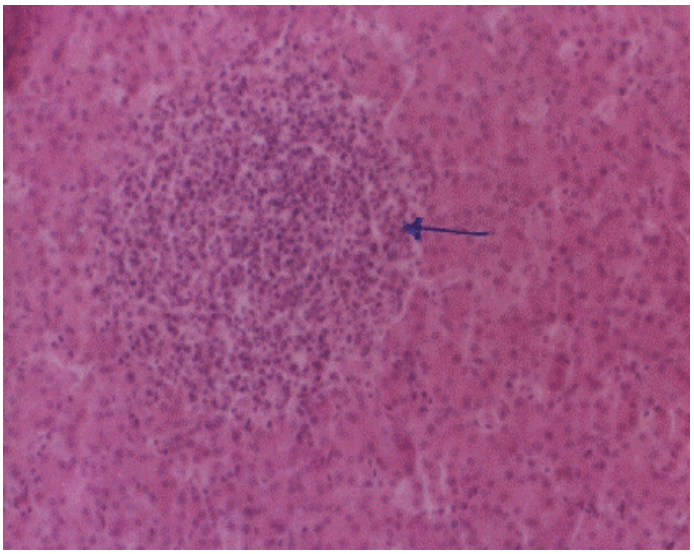

Fig. 1: Photomicrograph of the liver showing focal necrosis and mononuclear leucocytic infiltration in a chick treated with aqueous stem bark crude extract of *Khaya senegalensis* at 1000 mg kg^{-1} for 28 days. H and E x400

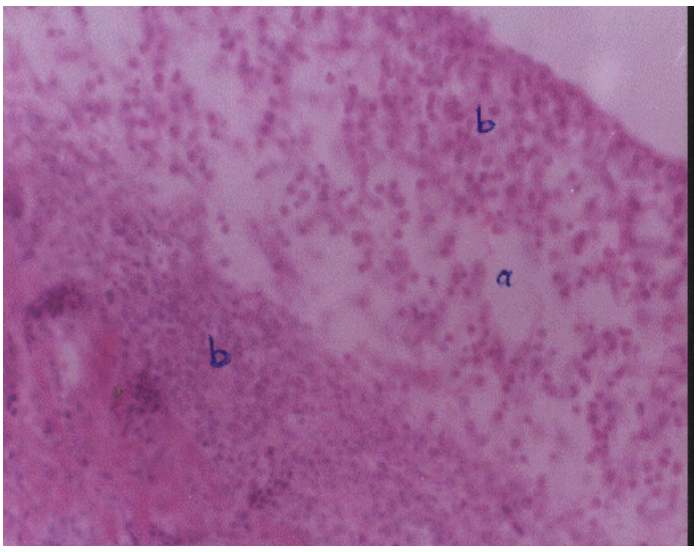

Fig. 2: Photomicrograph of the heart showing thickening by fluid exudation (a) and mononuclear cellular infiltrates (b) in a chick treated with aqueous stem bark crude extract of *Butyrospermum paradoxum* at 2000 mg kg^{-1} for 28 days. H and E x400

***In-vitro* anticoccidal effects:** Compared to the untreated control group (incubation in 2% potassium dichromate solution) oocyst sporulation was not inhibited in any way by incubation in *K. senegalensis* stem bark extract (Table 3). However, the various concentrations of *B. paradoxum* and *A. senegalensis* produced varying degrees of inhibition in oocyst sporulation; the former plant extract producing the greater effect of the two. The percentage inhibition was concentration

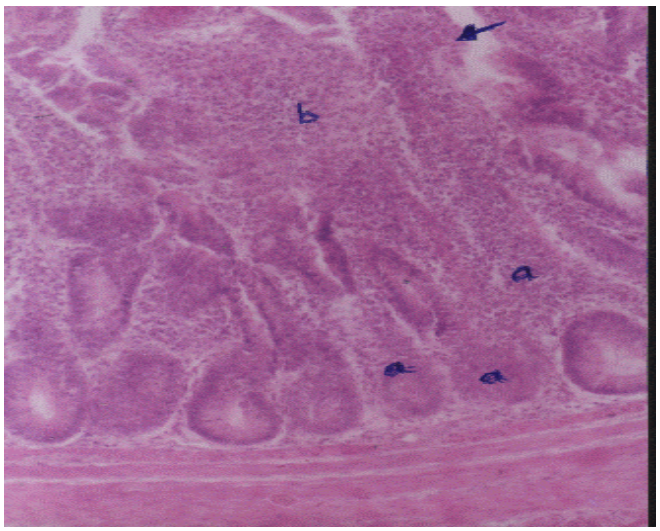

Fig. 3: Photomicrograph of duodenum showing epithelial necrosis (arrowed) leading to loss of villi, glandular necrosis (a) with mononuclear cellular infiltration (b) in a chick treated with aqueous stem bark crude extract of *Butyrospermum paradoxum* at 1500 mg kg^{-1} for 28 days. H and E x200

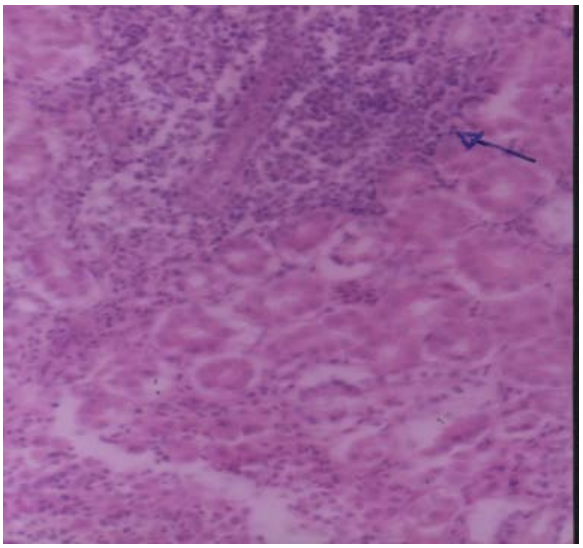

Fig. 4: Photomicrograph of kidney showing focal area of tubular necrosis with mononuclear cellular infiltration (arrowed) in a chick treated with aqueous stem bark crude extract of *Annona senegalensis* at 2000 mg kg^{-1} body weight for 28 days. H and E x400

dependent, being highest at the highest concentrations of the extracts used. The inhibition of oocyst sporulation produced by the two extracts was similar to that produced by amprolium at the 1.2 mg mL^{-1} concentration used in the study.

***In-vivo* anticoccidial effects:** Oocysts were first detected in the faeces of all the infected birds by day 4 post infection (Fig. 5). In the untreated group, oocyst numbers rose rapidly to attain peak

Table 3: *In vitro* anti-coccidial effect of the plant extracts on the sporulation of *Eimeria* oocysts

Drug/extract Concentration (mg mL^{-1})	No. of cultured (oocysts)	% Sporulation	% Inhibition (sporulation)
Potassium dichromate	100	100	0*
K. senegalensis			
100	100	100	0
250	100	100	0
500	100	100	0
1000	100	100	0
1500	100	100	0
A. senegalensis			
100	100	20	80
250	100	2	98
500	100	0	100
1000	100	0	100
1500	100	0	100
B. paradoxum			
100	100	0	100
250	100	0	100
500	100	0	100
1000	100	0	100
1500	100	0	100
Amprolium			
0.6	100	37	63
1.2	100	2	98

*Culture in potassium dichromate was used as control and standard (0% reduction in oocyst sporulation)

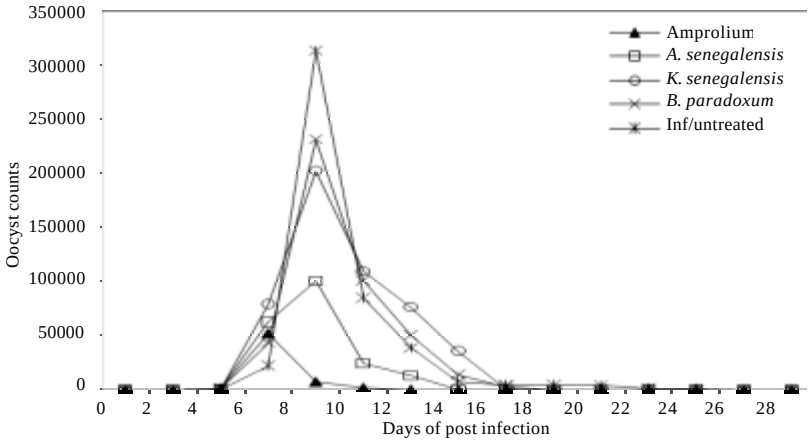

Fig. 5: Oocyst counts of chicks infected with mixed *Eimera* species and treated with the extracts or amprolium and their controls

counts by day 8 post infection before being reduced to lower numbers maintained to the end of the study. Treatment with amprolium or the three extracts checked the rise in oocyst out put so that lower peaks were recorded in these groups with an eventual elimination of the oocysts from the faeces by day 11, 14 and 18, respectively post treatment with amprolium, *A. senegalensis* and

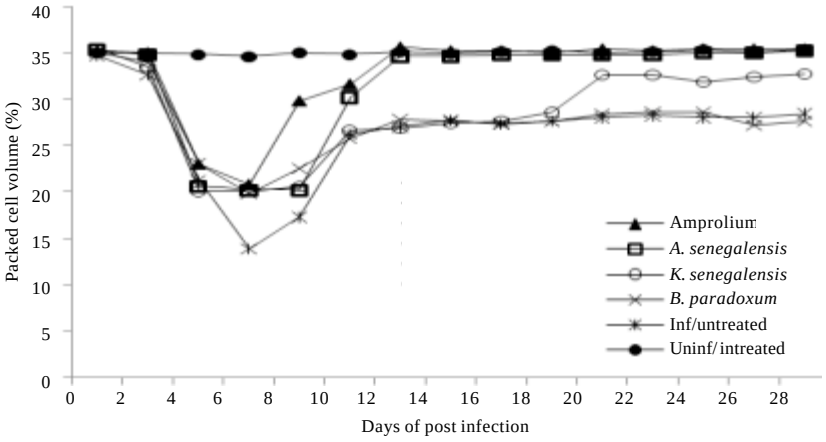

Fig. 6: Packed cell volume of chicks infected with mixed *Eimera* species and treated with the extracts or amprolium and their controls

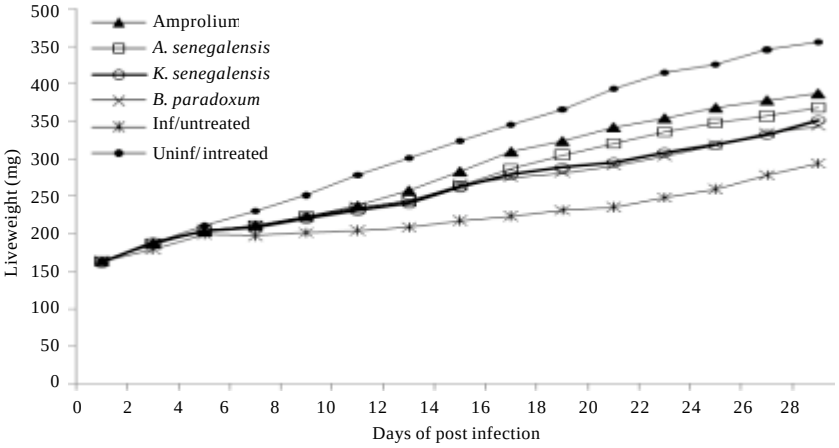

Fig. 7: Live weight of chicks infected with mixed *Eimera* species and treated with the extracts or amprolium and their controls

B. paradoxum but not with *K. senegalensis* or the untreated group during the remaining part of the study.

The Packed Cell Volume (PCV) of the healthy control birds remained within their preinfection levels without any significant changes throughout the study period (Fig. 6). On the other hand, the PCV of all the infected groups became reduced from day 4 post infection. The decline in PCV was greatest in the infected/untreated group followed respectively in descending order by those groups treated with the extracts and amprolium. Attempt at return to preinfection values in all the infected groups was successful from day 12 post infection and only in the groups treated with amprolium and *A. senegalensis*.

The chicks in all the experimental groups gained weight during the study (Fig. 7). However, weight gain was highest in the healthy control group followed, respectively in descending order by those treated with amprolium *A. senegalensis*, *K. senegalensis* and *B. paradoxum*. The untreated

group gained the least weight while the healthy control group gained significantly more weight than any of the infected but treated groups.

DISCUSSION

The results of the phytochemical analysis revealed that the aqueous extracts of the stem barks of *K. senegalensis*, *B. paradoxum* and *A. senegalensis* contain several active chemical components many of which are present in each of the three plant extracts with a few being exclusively present in only one or two of them. Many of the active chemical components obtained are known to be toxic (Humphreys, 1988) and clinical manifestations and lesions associated with their toxicity were noted in some of the birds given either the single intraperitoneal or prolonged oral treatment of the extracts. The dose-related nature of the manifestations and lesions suggest they are pharmacological and a reflection of the variations in the concentrations of the various components in each plant extract.

Among the three extracts tested *K. senegalensis* had the least number and concentration of the active chemical components and at the concentrations and duration of administration used in this study the extract was the least toxic as it produced only mild clinical signs and lesions without mortality even at very high concentrations. In an earlier study, Onyeyili *et al.* (2000) administered up to 6,400 mg kg^{-1} of the extract in rats without recording any severe toxic effects or deaths. On the other hand *B. paradoxum* was most toxic, causing up to 80-100% mortality when administered intraperitoneally or orally. Mbaya *et al.* (2007) reported essentially similar clinical signs, pathological lesions and mortality pattern in rats following intraperitoneal administration of graded doses (400-3200 mg kg^{-1} body weight) of the ethanolic stem bark extract of the plant. *Anona senegalensis* was also toxic but at concentrations in excess of 200 mg kg^{-1} when given by the intraperitoneal route. However, when administered orally, *A. senegalensis* extract became less toxic and caused no mortality among the treated birds probably due to poor rate of absorption of its toxic chemical components or their degradation to relatively safer by-products by digestive enzymes in the gastrointestinal tract during the long period of administration to the animals. Furthermore, the reduced toxicity of the extract by the oral route agrees, in part, with the observations of Hashemi *et al.* (2008) that water suspensions of herbal aqueous extracts were not usually toxic to birds when administered by the oral route even at concentrations of up to 2000 mg kg^{-1} body weight.

The susceptibility of animals to plant materials depend on the types of active principles in the plant, the concentration given to the animal, the rate of their metabolic conversion to metabolites in the liver and their subsequent excretion from the body (Amna *et al.*, 2011; Abdelgadir *et al.*, 2010). The liver is the major organ for metabolism in the body (Baggot, 1984). Consequently, the lesions observed in the liver may be as a result of damage caused in the organ during the biotransformation of these active chemical components by the organ. The excretion of some of the active components or their by-products through the kidney and their subsequent toxicity to the organ may account for the severe and widespread lesions noted in the kidney of some of the treated birds during this study. The widespread nature of the lesions in several organs and tissues including the lungs, heart, intestines and spleen suggest that the active components were widely distributed in the body where they produced toxic effects especially in the birds treated with *B. paradoxum* extracts.

Among the three extracts tested, *B. Paradoxum* exerted the greatest percentage reduction in oocyst sporulation but had the lowest *in-vivo* anti coccidial effect. These contrasting features show

that it has relatively little contribution in the treatment and control of coccidiosis through the oral route as it appears to act most effectively during the pre-infective stage. It is possible that the active components responsible for the high anti-sporulation effect produced *in vitro* were denatured, degraded or inactivated by enzymatic action in the intestine following oral treatment. On the other hand, *K. senegalensis* produced no *in vitro* anti-sporulation effect but had mild effect *in vivo* suggesting that the active components responsible for the noted *in vivo* effects may be by-products of enzymatic action on some of the chemical components in the extract. The *in vitro* anti-sporulation effect produced by *B. paradoxum* and *A. senegalensis* was concentration dependent suggesting that they were pharmacologically mediated.

Compared to the healthy control group of birds, treatment of *Eimeria* infected birds with each of the three extracts resulted in substantial reduction in faecal oocyst out put by the birds. *Annona senegalensis* produced the highest effect followed respectively *K. senegalensis* and *B. paradoxum* and these effects were manifested through the appreciation of the packed cell volume, improvement in live weight gain and the suppression of oocyst out put and sporulation.

CONCLUSION

In conclusion therefore, the results of this study showed that the crude aqueous extracts of the stem barks of *K. senegalensis*, *A. senegalensis* and *B. paradoxum* contain many chemical components that may be toxic to four-weeks old chicks when administered orally or intraperitoneally. *Khaya senegalensis* exhibited a highest margin of safety while *B. paradoxum* was the most toxic by both routes of administration. The three extracts also produced anticoccidial effects mediated through substantial reduction or complete termination of oocyst production and sporulation as well as appreciation of the PCV and weight gain of *Eimeria* infected birds treated with the extracts. These effects were highest with *A. senegalensis* and *K. senegalensis*. The high toxicity of *B. paradoxum* precludes its possible usefulness as an anticoccidial agent. There is need for farmers who use these extracts in the traditional treatment of avian coccidiosis to be mindful of their levels of toxicity to avoid unproductive effects.

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