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Anti-diarrhoeal Effects of Acacia *nilotica* Leaf Extract on Castor Oil Induced Diarrhoea in Wistar Strain Albino Rats

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ABSTRACT: This study was undertaken to evaluate the effects of aqueous and methanolic leaf extract of *Acacia nilotica* on castor oil-induced diarrhoea in rats. Antibacterial studies were carried out on *Escherichia coli, Salmonella typhimurium,* and *Staphylococcus aureus*. All treatments were administered orally for effect on intestinal motility, gastrointestinal transit of charcoal meal, fluid accumulation, and electrolyte secretion. Phytochemical screening of the aqueous and methanolic leaf extracts showed the presence of tannins, saponins, alkaloids, flavonoids, terpenoids, glycosides, and phenols. Quantitative estimation of the phytochemicals showed that the methanolic extract contained a higher amount of the phytochemicals with 38.46±0.41 phenolics content compared to 29.46±1.12 as the phenolics content for the aqueous extract. The methanolic extract exhibited a higher zone of inhibition of 14.34±1.54 for the growth of *Staphylococcus aureus* against 11.23±0.67 exhibited by the aqueous extract for the same organism. It was observed that 400mg/kg methanolic extract significantly ($p<0.05$) inhibited intestinal motility by 45%, fluid accumulation was reduced by 56% while animals treated with 400mg/kg of aqueous extract intestinal motility was inhibited by 35%, and fluid accumulation reduced by 47%. It was observed that the inhibitions of intestinal motility, number of wet faeces, volume, and weight of intestinal content as well as electrolytes secretion were dose-dependent. The higher the dosage, the higher the percentage of inhibition. The phytochemicals detected in these extracts may be responsible for observed effects and can attest to its uses as an anti-diarrhoeal agent in traditional medicine.

Keywords: *Acacia nilotica*, castor oil, intestinal motility, aqueous and methanolic extract, anti-diarrhoeal agent

INTRODUCTION

Diarrhoea account for more than 5 to 8 million deaths worldwide each year especially in developing countries (WHO, 2006).To combat this problem, the world health organization has initiated a diarrhoea diseases control programme to study traditional medicinal practice and other related aspects, together with the evaluation and prevention approaches. Plants have been a valuable source of natural product for maintaining human health for many years. More recently there has been a greater search for natural therapies (Mukherjee *et al*., 1995).The use of herbal medicine in the treatment of diarrhoea is a common practice in many African countries. The WHO suggested that medicinal plants would be the best source from which to develop a variety of medications.

Herbal medicine derived from medicinal plant materials and organic matter is still the main say of about 75 – 80% of the world population for health care, marketed and gaining popularity in developed and developing countries. Herbs have medicinal properties due to the presence of different active principles like alkaloids, volatile essential oils, glycosides, resins, oleoresins, steroids, tannins, flavonoids, terpenoids and phenols (Sekar *et al*., 2010).

In the last few years there has been an exponential growth in the field of herbal medicine because of their natural origin, easy availability, efficacy, safety and less side effects with efficient cure to age related disorders like memory lost and immune disorders for which no modern cure is available (Kamboj, 2000). Diarrhoea as disease or symptom of some disease conditions is associated with viral, bacterial and fungal infections, food poisoning and other disease conditions. Uncontrolled diarrhoea is dangerous as it can lead to loss of body fluids and electrolyte imbalance. Excessive loss of body fluid results in severe dehydration and death. Maintenance of electrolyte balance and replacement of body fluid is therefore crucial in the management of diarrhoea. Search for potent and cheap drug with anti-diarrhoeal properties is on the increase with the hope of discovering a well-tolerated anti-diarrhoeal drug with fewer side effects. Presently, the management of diarrhoea involves the use of anti-motility agents, antisecretory agents, antibacterial and antifungal agents, as well as oral rehydration therapy (ORT). Diarrhoea has been ranked fourth among the leading causes of death in the world and its incidence is more than any other disease condition worldwide, (WHO, 1995). Childhood mortality due to diarrhoea is frequent in developing countries. In those countries it has been estimated that about half of the cases are related to malnutrition, (Yoon, 1997).

Objectives of the study

The objectives of the study are to qualitatively and quantitatively determine the presence of some phytochemicals in the aqueous and methanolic leaf extracts of *Acacia nilotica*, to determine the antimicrobial susceptibility and zones of inhibition of the growth of three test organisms (*Escherichia coli, Salmonella typhimurium* and *Staphylococcus aureus*) by the aqueous and methanolic leaf extracts, to determine the anti-diarrhoeal effect of aqueous and methanolic leaf extracts of *Acacia nilotica* by inhibition of fluid accumulation, electrolyte secretion and intestinal propulsion in castor oil induced diarrhoea in experimental rats.

MATERIALS AND METHODS

The leaves of *Acacia nilotica* were collected in Jimeta, Yola North Local Government Area. It was identified and authenticated by the Department of Forestry School of Agriculture and Agricultural Technology, Modibbo Adama University of Technology Yola. The plant material was dried at room temperature, processed into powdered form and stored in an air tight container.

Experimental animals

Adult Wistar strain albino rats both males and females weighing between 100 – 120g were purchased from the animal house National Veterinary Research Institute Vom Jos, Plateau State Nigeria. The animals were kept in well ventilated plastic cages and stabilized using standard laboratory chaw and fed *ad libitum* with water.

Source of test organisms

The clinical isolates of enteropathogenic *Escherichia coli, Salmonella typhimurium,* and *Staphylococcus aureus w*ere obtained from the microbiology department, bacteriology unit Modibbo Adama University of Technology Yola, Adamawa State Nigeria. The organisms were maintained on nutrient agar slants and kept in a refrigerator at 4°C until required for use.

Preparation of nutrient agar

Twenty eight grammes of nutrient base powder was put into a conical flask containing one litre of deionised water and mixed thoroughly. The conical flask containing the mixture was capped with aluminium foil and autoclaved for 15min at 121° C. The mixture was allowed to cool at room temperature and stored in a refrigerator until required.

Preparation of aqueous and methanolic extracts

An aqueous extract was obtained by boiling 250g of powdered leaf of *Acacia nilotica* in 100ml of distilled water for 1h. The suspension obtained was allowed to cool and filtered through whatman No. 1 filter paper to obtain a clear solution. The methanolic extract was obtained by dissolving 250g of powdered leaf of *Acacia nilotica* in 100ml of methanol and the mixture was put into a Soxhlet extractor for extraction. The extract was concentrated using a rotary evaporator at 60° C.

Phytochemical screening

Aqueous and methanolic leaf extracts of *Acacia nilotica* were tested for the presence of alkaloids, tannins, saponins, flavonoids, steroids and glycosides using the procedures described by Sofowora, (1993).

Test for alkaloids

For alkaloids, 2ml of picric acid was added to 0.5g of the extract, the appearance of orange colour was observed indicating the presence of alkaloids.

Test for saponins

For saponins, 0.5g of the plant extract was shaken with

water in a test tube and heated to boil. Frothing was observed, indicating the presence of saponins.

Test for tannins

For tannins, 0.5g of the plant extract was added to10ml of water in a test tube and filtered. A few drops of 0.1% ferric chloride were added and a green precipitate was observed indicating the presence of tannins.

Test for steroids

For steroids, 2ml of acetic anhydride was added to 0.5g of extract and 2 ml sulphuric acid added. Violet to blue colour was not observed indicating the absence of steroids.

Test for flavonoids

For flavonoids, 10ml of ethylacetate was added to small portion of the extract and it was held over steam bath for 3min. The mixture was filtered and 1ml of dilute ammonia solution was added to 4ml of the filtrate and then shaken. Yellow colour was observed indicating the presence of flavonoids (Sofowora, 1993).

Test for phenols

For phenols, 0.5g of the extract material was boiled with 10ml of sulphuric acid and filtered while hot, and the filtrate was shaken with 5ml of chloroform. The chloroform layer was pipetted into another test tube and 1ml of diluted ammonia was added to it. A deep blue-green colour observed indicated the presence of phenols.

Test for terpenoids (Salkowskis's test).

Two millilitres of leaf extract were dissolved in 2ml of chloroform and evaporated to dryness. Then 2ml of concentrated sulphuric acid was added and heated for about 2min. A greyish colur observed indicated the presence of terpenoids (Harborne, 1989).

Test for cardiac glycosides

One hundred milligrammes of leaf extract was treated with 2ml of glacial acetic acid containing one drop of ferric chloride solution. This was under-layered with 1ml of concentrated H_2 SO_4. A brown ring observed at the interface indicated the presence of deoxy sugar; characteristic of cardenolides (Ngbede *et al.,* 2008).

Determination of antimicrobial activity of *Acacia nilotica* leaf extracts

Agar diffusion method described by Edward, (1983) was used.

Principle

An agar diffusion method operates on the principle that antibiotics will diffuse from a paper disc or small cylinder into an agar medium that contains test organism. The inhibition is observed as a failure of the organism to grow in the region of the antibiotic. It may depend on such factors as the diffusion rate of the antibiotic, the medium used, the sensitivity of the test organism, and incubation condition.

Procedure

The paper discs were put into the extracts whose concentration 20mg/ml and allowed to soak for some time. The discs were then removed and dried. The dried paper discs containing the absorbed extract were impregnated onto the surface of dried agar medium that has already been inoculated with the test organism. These were left on the work bench for prediffusion and later incubated at 37^{o} C for 24h. The plates were then observed and the diameter of zone of inhibition of growth was measured. Standard drug (loperamide) discs of 20mg/ml concentration were also used in order to compare the zone of inhibition produced by *Acacia nilotica* leaf extracts and standard drug (loperamide).

Experimental design

Thirty young adult male and female rats weighing100 – 120g were used for this study. The animals were grouped into six with five rats per group.

Group 1: Diarrhoetic control
Group 2: Standard (loperamide) control
Group 3: Aqueous extract 200mg/kg body weight
Group 4: Aqueous extract 400mg/kg body weight
Group 5: Methanolic extract 200mg/kg body weight
Group 6: Methanolic extract 400mg/kg body weight

Induction of diarrhoea using castor oil

The experimental animals were allowed to fast for 18h prior to the test but allowed free access to water. Group 1 was treated with 0.2ml of normal saline and 2ml of castor oil which served as the diarrhoetic control. Groups 2, 3, 4, 5 and 6 were challenged with 2ml of castor oil orally. The animals were then housed singly in cages lined with transparent paper to observe uniform stool formation. After two hours of diarrhoea induction, group 2 received 2mg/kg body weight standard drug, loperamide. Groups 3 and 4 received 200 and 400mg/kg body weight of aqueous extract respectively; groups 5 and 6 received 200 and 400mg/kg body weight of methanolic extract respectively. All the doses were administered orally. The animals were observed for the presence of formed stool

or absence of diarrhoea.

Effect of aqueous and methanolic extracts *Acacia nilotica* on castor oil induced intestinal propulsion

The effect of aqueous and methanolic leaf extract of *Acacia nilotica* on small intestine propulsion was tested using charcoal meal as described by Di-Carlo *et al*. (1994). The rats weighing 100 – 120g were fasted for 18h prior to the test but allowed free access to water. The rats were grouped into four with five rats per group. Group1 was administered with distilled water which served as normal control, groups II and III were administered with 200 and 400mg/kg body weight of the extracts and group IV received 2mg/kg body weight of loperamide. After 30min each rat was administered with 1ml of castor oil orally. After 30min of castor oil administration, each rat was administered with 1ml of charcoal meal (marker meal). After 30 min the rats were sacrificed, abdomen carefully opened and the intestine carefully removed. The length of the intestine measured and the distance travelled by the marker meal was measured from the pylorus to the caecum of each animal. The mean percentage movement of the charcoal meal in ratio to the intestinal length and the percentage of transit inhibition were measured. Effect of aqueous and methanolic leaf extract of *Acacia nilotica* on castor oil induced fluid accumulation and electrolyte secretion. Fluid accumulation and electrolyte secretion was determined by the method described by Di-Carlo *et al.* (1994). The grouping and number of animals per group as well as the treatments are the same as described above. The rats were sacrificed and the intestine carefully removed. A thread was tied at the pyloric and the ileocaecal junction, the intestine with its content weighed. The intestinal content was emptied into a graduated tube weighed and the volume measured. The empty intestine was weighed and the difference in weight between full and empty intestine was determined.

Estimation of potassium ions

Principle

Potassium ion was estimated by turbidometric method described by Fogh-Anderson *et al.* (1984). The extent of turbidity is proportional to the potassium ion concentration in the stool and was measured photometrically after subjecting the stool to centrifugation using desktop centrifuge at the speed of 1000rpm for 10 min.

Procedure

Two test tubes labelled standard and test. Ten millilitres (10ml) of potassium reagent was put into each test tube. To the test tube labelled standard, 2.5ml of standard solution was added and the test tube labelled test, 2.5ml of sample was added and the content of each test tube was mixed and incubated at 37°C for 5min. The absorbance of the standard and test were read against distilled water blank at a wavelength of 578nm.

$$\text{Concentration of sample} = \frac{\text{Absorbance of sample x concentration of standard.}}{\text{Absorbance of standard.}}$$

Estimation of chloride ions

Principle: In an acid medium, chloride ion and mercury form thiocyanate ions. These ions react with HNO_3 and $FeCl_3$. The intensity of the colour is directly proportional to the concentration of chloride ions.

Procedure: After centrifugation, 10ml of working reagent was added to three test tubes labelled blank, standard and sample. To standard test tube 10ml of standard solution was added and 10ml of sample was added to sample test tube, mixed well and incubated at 37°C for 1min. The absorbance of standard and sample were read against reagent blank.

$$\text{Chloride ion concentration} = \frac{\text{Absorbance of sample x concentration of standard.}}{\text{Absorbance of standard}}$$

Estimation of sodium ions

Principle

The method described by Fogh-Anderson *et al*. (1984) was used to estimate sodium ions. Sodium ions was precipitated as the triple salt, sodium, magnesium with excess uranium being reacted with ferrocyanide producing a chromophore whose absorbance vary inversely as the concentration of sodium in the test specimen.

Procedure

To three test tubes labelled blank, test, and standard, 1ml of filtrate was added; 0.5ml of sample was added to the test and standard tubes while the blank received an equal volume of distilled water. The preparations were mixed and centrifuged at 1500rpm for 10min and the supernatant from each tube was obtained. To the second set of tubes labelled blank, test and standard, were added 1ml of methanesulfonic acid and 0.5ml of supernatant earlier obtained. To this mixture, 0.05ml of colour reagent was added to each tube, mixed and the absorbance read at 550nm.

$$\text{Concentration of sodium ions} = \frac{\text{Absorbance of sample x concentration of standard}}{\text{Absorbance of standard}}$$

Table 1: Physical characteristics of *Acacia nilotica* leaf extract.

Solvent	Weight of plant material (g)	Weight of extract (g)	Percentage yield	Colour
Aqueous extract	250	22.00	8.4	Dark brown
Methanolic extract	250	30.00	11.6	Dark brown

Table 2: Phytochemical constituents of aqueous and methanolic leaf extracts of *Acacia nilotica*.

Phytochemical constituent	Aqueous extract	Methanolic extract
Alkaloids	+	+
Tannins	+	+
Saponins	+	+
Flavonoids	+	+
Steroids	-	-
Terpenoids	-	+
Gycosides	+	+
Phenols	+	+

+ = Presence of phytochemical constituent; - = Absence of phytochemical constituent.

Table 3: Concentration of some phytochemical constituents in the aqueous and methanolic leaf extract of *Acacia nilotica* (g/100g).

Phytochemical constituent	Aqueous extract	Methanolic extract
Alkaloids	14.71±0.32	18.37±0.61
Saponins	6.93±0.64	9.08±0.62
Flavonoids	2.53±0.51	2.21±1.24
Phenols	29.46±1.12	38.46±0.41
Tannins	4.89±1.33	7.83±0.50

Key: Values are mean ± Standard error of mean (n = 3).

RESULTS AND DISCUSSION

Extraction of the plant material with methanol gave a higher yield of the bioactive components compared to aqueous extract. Since methanol is less polar than water, it can easily penetrate the cell membrane thereby permitting the extraction of large amount of phytochemicals. Methanol can dissolve some non-polar molecules as well as bioactive compounds from plants which belong to various chemical groups such as tannins, alkaloids, and saponins (Agrawal *et al*., 2010).

Acacia nilotica leaf extract contains among others alkaloids, tannins, saponins, flavonoids, terpenoids, glycosides and phenols. Some of these phytochemical components have been implicated to possess analgesic and anti-inflammatory effects (Gupta, 1994). Tannins are known to have astringent properties and therefore could be used to treat diarrhoea (Mota *et al*., 1995). Quantitative estimation of phytochemicals in *Acacia nilotica* leaf extract indicated that the plant is rich in flavonoids, tannins, saponins, alkaloids and phenols in ascending order of concentration. The concentrations of these bioactive compounds were observed to be higher in the methanolic extract compared to the aqueous extract as shown in (Tables 1 to 3). Although several constituents are present in the extract, it is possible that flavonoids acting singly or in combination with other constituents produced the anti-diarrhoeal effect of *Acacia nilotica*. The anti-diarrhoeal activity of flavonoids has been attributed to their ability to inhibit intestinal motility and hydro-eletrolytic secretion (Di-Carlo, 1994). Aqueous and methanolic extracts as well as the standard drug (Loperamide) were observed to inhibit the growth of the three test organisms (*Escherichia coli, Salmonella typhimurium* and *Staphylococcus aureus*) to a certain degree. The zones of inhibition created by the methanolic extract are higher compared to those created by the aqueous extract while those created by Loperamide are higher compared to those created by the extracts (Table 4). The diameter of zone of inhibition is determined by the initial population density of the test organism, growth rate and rate of diffusion of the antimicrobial agent. This may be the reasons behind the differences in the zones of inhibition observed. The standard drug loperamide created larger zones of inhibition against the three test organisms. This may be attributed to the fact that conventional drugs are usually prepared from synthetic materials by standard techniques and procedures while herbal medicinal products prepared from plants and animals origins are subject to contaminations and

Table 4: Zones of inhibition of the growth of some bacteria by the aqueous and methanolic leaf extracts of *Acacia nilotica* (mm).

Test organism	Aqueous extract	Methanolic extract	Loperamide
E. coli	9.12±2.22	11.05±1.63	22.13±1.20
Salmonella typhie	10.21±1.34	13.02±2.12	20.25±1.13
Staphylococcus aureus	11.23±0.67	14.34±1.54	25.16±0.67

Key: Values are mean ± Standard error of mean (n = 3).

Table 5: Effect of aqueous and methanolic leaf extract of *Acacia nilotica* on castor oil induced diarrhoea in Wistar rats.

Treatment	Mean number of wet faeces or defaecation in 24h	Percentage inhibition
Normal/control	1.29±0.01	-------
Lomeramide2mg/kgbwt	0.30±0.56	76.7%
AE 200mg/kgbwt	0.80±1.36	37.5%
AE 400mg/kgbwt	0.75±0.25	41.8%
ME 200mg/kgbwt	0.65±1.32	49.6%
ME 400mg/kgbwt	0.60±0.25	53.4%

Key: Values are mean ± Standard error of mean (n = 3) ($p<0.05$). Methanolic extract exhibited significantly ($p<0.05$) higher percentage inhibition compared to aqueous extract and the percentage inhibition is dose dependent in both cases.

Table 6: Effect of aqueous and methanolic leaf extract of *Acacia nilotica* on castor oil induced fluid accumulation and electrolyte secretion.

Treatment	Mean volume of intestinal fluid (ml)	% inhibition	Na^+ meq/l	K^+ meq/l	Cl^- meq/l
Normal/control	0.76±0.49	-----	134.21±1.71	27.2±0.12	56.03±2.13
Loperamide 2mg/kgbwt	0.20±0.11	73%	105.34±2.19	5.5±0.99	105.21±1.21
AE 200mg/kgbwt	0.46±1.26	39%	122.02±1.28	10.0±0.67	65.32±1.72
AE 400mg/kgbwt	0.40±1.62	47%	125.74±4.12	9.7±1.08	77.21±2.51
ME 200mg/kgbwt	0.36±1.26	52%	120.60±3.35	8.1±0.18	75.72±2.24
ME 400mg/kgbwt	0.33±1.21	56%	102.25±2.30	8.7±0.54	85.11±0.65

Key: Values are mean ± Standard error of mean (n = 3) ($p<0.05$).

Mean volume of fluid in the intestine decreases with increase in the doses of the extract as depicted on the (Table 6). Percentage inhibition increases with increase in the doses of the extracts. The methanolic extract exhibited lower mean volume of fluid in the intestine and higher percentage inhibition compared to aqueous extract.

deterioration most of the time (Adeshina *et al*., 2012). The use of castor oil as diarrhoea inducer is well documented (Dahiru, 2006; Akter, 2013). The aqueous and methanolic leaf extracts of *Acacia nilotica* exhibited a significant anti-diarrhoeal effect against castor oil induced diarrhoea. It was observed that the mean number of semi solid faeces and number of defaecation decreases with increase in the concentration of the extracts (Table 5). In contrast to what was observed earlier in the case of mean number faeces and number of defaecations, percentage inhibition of diarrhoea by the extracts as well as the standard drug (Loperamide) increases with increase in the concentration of the extracts as shown in (Table 5). More so, methanolic extract exhibited higher percentage inhibition compared to aqueous extract. The effect of aqueous and methanolic leaf extract of *Acacia nilotica* on castor oil induced fluid accumulation and electrolytes (Na^+, K^+, and Cl^-) secretion (Table 6) showed decrease in fluid accumulation with increase in the concentration of the extracts. Percentage inhibition of fluid accumulation increases with increase in the concentration of the extracts. Sodium and potassium secretion decreases with increase in the concentration of the extracts while chloride secretion increases with increase in the concentration of the extracts. Loperamide on the other hand showed significantly ($p<0.05$) lower volume of intestinal fluid accumulation, lower Na^+ and K^+ secretion and higher Cl^- secretion compared to normal/control. In the case of castor oil induced interpooling, the volume of intestinal content as well as the weight of intestinal content significantly ($p<0.05$) decreases with increase in the concentration of extracts (Table 7). In contrast to this observation, percentage inhibition of interpooling significantly increases with increase in the concentration of the extracts.

In the case of intestinal propulsion of experimental animals induced by castor oil, loperamide exhibited higher mean intestinal length, higher mean distance

Table 7: Effect of aqueous and methanolic leaf extract of *Acacia nilotica* on castor oil induced interpooling.

Treatment	Volume of intestinal content (ml)	% inhibition	Weight of intestinal content (g)	% inhibition
Normal/control	0.67±0.21	-----	0.68±0.61	------
Loperamide 2mg/kgbwt	0.15±0.17	77.12%	0.20±1.23	70.5%
AE 200mg/kgbwt	0.45±0.34	32.83%	0.42±1.26	38.2%
AE 400mg/kgbwt	0.39±0.76	41.72%	0.41±0.61	40.3%
ME 200mg/kgbwt	0.36±1.22	46.21%	0.35±1.22	47.7%
ME 400mg/kgbwt	0.30±1.21	50.70%	0.29±1.26	57.3%

Key: Values are mean ± Standard error of mean (n = 3) ($p<0.05$).
The volume of intestinal content and weight of intestinal content decreases with increase in the doses while percentage inhibition increases with increase in the doses. Methanolic extract exhibited higher percentage inhibition as well as higher weight of intestinal content compared to aqueous extract.

Table 8: Effect of aqueous and methanolic leaf extract of *Acacia nilotica* on castor oil induced intestinal propulsion.

Treatment	Mean length of intestine (cm)	Mean of distance moved by charcoal meal (cm)	% inhibition
Normal/control	87.32±2.51	80.26±2.48	------
Loperamide 2mg/kgbwt	81.20±1.81	32.30±1.72	60.2%
AE 200mg/kgbwt	86.86±2.41	55.13±1.47	30.2%
AE 400mg/kgbwt	71.33±1.00	52.41±2.86	35.2%
ME 200mg/kgbwt	69.71±2.01	50.31±1.47	37.5%
ME 400mg/kgbwt	83.37±0.51	44.24±2.16	45.0%

Key: Values are mean ± Standard error of mean (n = 3) ($p<0.05$).

moved by charcoal meal and higher percentage inhibition of intestinal propulsion compared to both extracts but significantly lower ($p<0.05$) compared to normal/control (Table 8). Mean intestinal length, mean distance moved by charcoal meal of the extracts are significantly lower ($p<0.05$) compared to normal/control rats. The mean intestinal length for the aqueous extract decreases with increase in concentration, but for the methanolic extract mean intestinal length increases with increase in concentration. Mean distance moved by charcoal meal in both extracts decreases with increase in concentration. In all the cases observed so far the percentage inhibitions exhibited by loperamide were significantly higher ($p<0.05$) compared with those exhibited by either aqueous or methanolic extract. The methanolic extract on the other hand exhibited significantly higher percentage ($p<0.05$) inhibition when compared with the aqueous extract in all the cases (i.e. mean number of faeces, volume of intestinal content, weight of intestinal content, mean volume of intestinal fluid, electrolyte secretion, mean intestinal length, and mean distance moved by charcoal meal). Overall, these extracts have demonstrated their anti-diarrhoeal potentials although with lower percentages of inhibition as compared with the standard drug (loperamide). These observations have to some extent justified the use of *Acacia nilotica* leaf by the traditional and herbal medicinal practitioners as an anti-diarrhoeal agent.

Conclusion

Aqueous and methanolic leaf extracts of *Acacia nilotica* subjected to qualitative screening and quantitative estimation of phytochemicals revealed the presence of alkaloids, saponins, flavonoids, tannins, glycosides, terpenoids and phenols with the methanolic extract showing higher concentrations of these active components of the plant. Both extracts exhibited their antibacterial potentials by the various zones of inhibition of the growth of *Staphylococcus aureus, Salmonella typhimurium*, and *Escherichia coli* when subjected to antimicrobial susceptibility test. The plant extract at the end of the analysis has proven its anti-diarrhoeal potentials by the high percentage of inhibition of intestinal motility as compared to the standard drug (loperamide).

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