JCBPS; Section B; August 2017 – October 2017, Vol. 7, No. 4; 1001-1010. E- ISSN: 2249 –1929

[DOI: 10.24214/jcbps.B.7.4.10101]

Journal of Chemical, Biological and Physical Sciences

An International Peer Review E-3 Journal of Sciences

*Available online at*www.jcbsc.org

Section B: Biological Sciences

CODEN (USA): JCBPAT Research Article

Evaluation of Methanolic Extract of *Anogeissus Leiocarpus* Stem Bark on Castor Oil- Induced Diarrhoea in Rats

Memi G.G [1*], D, Dahiru [2], Junaid, O.Q [1], A. Abubakar [1], Ogah J.J [1]

[1] Department of Biological Sciences, Federal University, Kashere P.M.B 0182 Gombe state Nigeria.

[2] Department of Biochemistry, Modibbo Adama University of Technology P.M.B 1076 Yola, Adamawa State. Nigeria.

Received: 30 June 2017**; Revised:** 14 July 2017; **Accepted:** 02 August 2017

Abstract: Diarrhoea has been a major cause of morbidity and mortality especially among children in developing countries. *Anogeissus leiocarpus* (Combretaceae) has been reported to possess a number of medicinal properties. The purpose of the present study was to evaluate effects of methanolic extract of *A. leiocarpus* stem bark on castor oil- induced diarrhoea model in rats. Rats were divided into four groups of five in each and treated. Group I (distilled water), group II (200mg/kg extract), group III (400mg/kg) and group IV Loperamide (2mg/kg). All treatments were orally administered for effects on intestinal motility, fluid accumulation and electrolytes secretion pre and post administration of extract and their respective percentages expressed relative to experimental control. Methanol 10.67%. The preliminary phytochemical screening revealed the presence of tannins, saponins, flavonoids, terpenes, alkaloids and glycosides. 200 and 400mg/kg methanolic extract significantly ($P<0.05$) inhibited motility by 41.11% and 51.32% respectively, and fluid accumulation was reduced by 39.45% and 57.87%. The study also revealed progressive diminution of Na^+ and K^+ concentrations in stool. There were significant reductions, ($P<0.05$) in frequency of wet stool, with lower curatives values of 10.00% and 24.00% than

preventive values of 20.79% and 44.99%. The presence of some of the phytochemicals in the extracts may be responsible for the observed activities and attest to its utility in a wide range of diarrhoeal states. Further research to be carried on monitored fractionation and subsequent structural elucidation with a view to understanding the compound and possible mechanism of action.

Keywords: Phytochemical, *Anogeissus leiocarpus*, Propulsion, Therapeutic, Diminution, Diarrheoa.

INTRODUCTION

Diarrhoea has long been recognized as one of the most important health problems in developing countries. It is an increase in fluidity or volume of bowel movements characterized by frequent wet stool [1]. It is a condition in which faeces are discharged from the bowels frequently and in liquid form [2] as a result of infectious microorganisms and disturbances of the normal functioning of the bowels. The primary function of gastrointestinal tract (GIT) is to provide the body with continual supply of water, electrolytes and nutrients. Digestion and absorption principally take place in the small intestine. GIT while playing its physiologic functions is subjected to some pathological states, or alterations which can affect its role [3], It is a major health problem especially for children under the age of five (5) and up to 17% of children admitted in the pediatric ward die of diarrhoea. There are large numbers of epidemiological and experimental evidence pertaining to worldwide acute diarrhoea disease [4]. According to World Health Organization (WHO) estimates for 1998, about 7.1 million deaths were caused by diarrhoea [5]. Worldwide, the disease accounts for 4-5 million deaths among humans annually. In Nigeria, diarrhoea remains the number one killer disease among children aged 1-5 years [1]. In Nigeria, an estimate of 120,000 infant and child mortality was recorded per year due to dehydration related to diarrhoea [6]. Diarrhoea prevalence among children in north eastern Nigeria was estimated to be as high as 22% [7].

To overcome the menace of diarrhoea disease in developing countries, (WHO) has included a programme for the control of diarrhoea, which involves the use of traditional herbal medicine in developing countries, the majority of people living in rural areas almost exclusively use traditional medicines in treating all sorts of disease including diarrhea. The use of herbal drugs in the treatment of diarrhea is a common practice in Africa [1]. *Anogeissus leiocarpus* names of the plant include “chew stick tree” (English), “Marke” (Hausa) “Wolom” (Tangale), “Annum’’ (Kanuri) “Atara’’ (Igbo),’’ “Aina Orin Odon’’ (Yoruba) ‘’Bambara: nagallama’’ (in Mali.) and “Kojole” (Fulfulde), belongs to the family Combretaceae. Some members of the Cambretaceae have high concentrations of flavonoids, terpenoid, tannins or polyphenolic compounds. Many Combretaceae species are widely distributed in Nigeria and are used in traditional medicine for treating respiratory diseases (asthma, catarrh, chronic bronchitis, Cough, hay fever, hemoptysis, pneumonia, pulmonary disorder and tuberculosis) and other human diseases [8]. It is therefore important to identify and evaluate common available natural drugs as alternative to currently use anti diarrhoea drugs, which are not completely free from adverse effects [9]. A range of medicinal plants with antidiarrheal properties have been used by the traditional medicine practioners. The efficacy and safety of many of these medicinal plants have been reported [10, 11]. The potential for new drugs development or discovery depend on screening, evaluation and documentation of herbal plants used in the treatment of diseases including diarrhoea [12] the stem of *A. leiocarpus* as claimed by the traditional medicine practioners is utilized in the treatment of diarrhoea.

However, there is little or no existing scientific evidence about the efficacy of the plant. It is therefore necessary to carry out the pharmacological screening of the stem bark extract with emphasis on its antidiarrheal effect on castor oil diarrhoea model in rats.

MATERIALS AND METHODS

Identification of the Plant Material: Stem bark of *A. leiocarpus* was collected from Tanduru forest at Kaltungo Local Government area of Gombe State, Nigeria. It was identified and authenticated by Mr. Bristone B. Pola of Plant science department Modibbo Adama University of Technology, Yola, Nigeria. The voucher specimen was deposited in the herbarium for reference purpose.

Experimental Animals: Young adult male and female rats weighing 80-110g were obtained from National Vetinary research institute, Vom Plateau State, Nigeria. The animals were allowed to acclimatize for 7 days in the experimental laboratory under standard condition of 12:12.hours light and dark cycle. They were fed with standard rat pellets (Pfizer feeds, Jos, Nigeria) and provided with water *ad libitum*.

Extract Preparation: The stem bark of A. *leiocarpus* was washed with clean water, air-dried, pulverized using pestle and mortar and sieved. One hundred grams of the powdered stem was packed in soxhlet column and extracted by adding 500ml of absolute methanol at temperature less than 60^0C for 12hrs.The extract was concentrated using hot water boiling bath. Water was used to reconstitute the solid extract to obtain a desired concentration for the studies. This procedure was repeated three times and percentage extract yield calculated accordingly.

Phytochemical Analysis: The stem bark of *A. leiocarpus* was subjected to qualitative analysis for various phytoconstituents like flavonoides, Tannins, Saponins and alkaloids. They were identified by characteristics colour change using standard procedures [13, 14].

Effect of Methanolic Extract on Castor il-Induced Intestinal Propulsion (transit time): The effect of A. *leiocarpus* on small intestinal propulsion was tested using the charcoal meal method of Caposso *et al* [15]. Rats weighing (80-110 grams) and fasted for 24hours but allowed free access to water. They were randomized and placed in 4 groups of 5 animals each. Group I was administered with distilled water, groups II and III were treated with 200 and 400mg/kg body weight of the extract, and group IV received 2mg/kg loperamide (standard). After 30 minutes each rat was administered with 1 ml of castor oil orally. After 30 minutes of castor oil treatment, rats were served with 1ml of marker meal (10% activated charcoal in 5% gum acacia). Rats were sacrificed 30 minutes later by inhalation of chloroform and the abdomen opened, intestine carefully removed. The intestinal length noted and the intestinal distance travelled by charcoal meal (marker) was measured (in cm) from pylorus to caecum of each animal [16]. The mean percentage movement of charcoal meal in ratio to intestinal length, and percentage of transit inhibition were determined [17].

Effect of Methanolic Extract on Castor Oil-Induced Fluid Accumulation and Electrolyte Secretion: This was determined according to the method of Di-carlo *et al* [18]. Rats of either sex (80-110 g) were fasted for 24hrs, but allowed free access to water. Rats were randomized and placed in 4 groups of 5 rats per cage. All drugs were orally administered. Group I was administered with distilled water, groups II and III were pre-treated with 200 and 400 mg/kg of the extracts respectively. Thirty (30) minutes later each rat was administered with 2ml/100g b.w per rat, castor oil. The rats were anaesthetized 30 minutes later by inhalation of chloroform. Animals were sacrificed, small intestine carefully removed, tied with thread at the pyloric end and the ileocecal junction and weighed. The intestinal content was milked into a graduated tube and their

volume measured. The intestine reweighed and the difference between full and empty intestines was calculated. The Cl^-, Na^+ and K^+ concentrations in the supernatant after centrifuging the intraluminal fluid was measured by flame photometry [19, 20].

Therapeutic (Curative) Effect of Methanolic Extract on Castor Oil-Induced Diarrhoea: The method of Mukherjee *et al* [21] was adopted. Rats weighing(80-110 g) were fasted for 18hours with free access to water and randomly grouped into four groups of five animals each(n=5).Group I received distilled water (control), groups II and lll received 200 and 400mg/kg body weight of extracts respectively and group IV administered with 2mg/kg loperamide (standard). All administrations were carried out orally using orogastric cannula. Each animal was given 2ml of castor oil orally 30 minutes before treatment to induce diarrhoea .The animals were housed singly in perforate cage with a clean filter paper at the base. The characteristics of the diarrhaoel episodes were observed for a period of 4hrs. During that period the presence of wet faeces and number of wet faeces (frequency of defecation) were noted [22]. The wet faeces was counted easily at the end of the experiment, by carefully removing the filter paper at the bottom of the cage [23]. Ceasation in the production of wet stool was regarded as curative. The curative effect was determined in terms of percentage diarrhoael curative index relative to experimental control.

Preventive Effect of Methanolic Extract on Castor Oil-Induced Diarrhoea: The method of 21 Mukherjee *et al*., (1998) was adopted. Rats weighing(80-110 g) were fasted for 18hours with free access to water and randomly grouped into four groups of five animals each(n=5).Group I received distilled water (control), groups II and III received 200 and 400mg/kg body weight of extracts respectively and group IV administered with 2mg/kg loperamide (standard). All administrations were carried out orally using orogastric cannula. Each animal was given 2ml of castor oil orally after 30 minutes of pretreatments, to induce diarrhoea .The animals were housed singly in perforate cage with a clean filter paper at the base. The characteristics of the diarrhoeal episodes were observed for a period of 4hrs. During that period the presence of wet faeces and number of wet faeces (frequency of defecation) were noted [22]. The wet faeces was counted easily at the end of the experiment, by carefully removing the filter paper at the bottom of the cage [23]. Absence or delay in the production of wet stool was regarded as preventive. The preventive effect was determined in terms of percentage diarrhoael protection (inhibition) index relative to experimental control.

Statistical Analysis: The experimental results were expressed as means ± SD (Standard Deviation) and simple percentages. The statistical analysis of data was done using ANOVA (Analysis of variance) with level of statistical significance taken at $P<0.05$. T-test was used to determine the significance of deviation of treatment relative to control.

RESULTS

Table-1 showed that methanolic stem bark extract of *A. leiocarpus* gave positive reaction for each of the following secondary metabolites: alkaloids, flavonoids, tannins, saponins, glycosides, proteins, terpenoids but negative for Anthraquinones. **Table-2** showed the effect of methanolic extract of *A. leiocarpus* stem bark on intestinal propulsion. The mean distance traversed by charcoal meal (cm) in control group was 66.88 ± 0.45 which was the highest and the least distance (cm) 31.62 ± 0.41 was observed in standard drug treatment. It revealed that 200 mg/kg of methanolic extract reduced the intestinal transit by 41.11% while 400 mg/kg of the same caused a decrease of 51.32% of the intestinal propulsion relative to experimental control. The standard drug inhibited the movement of charcoal meal by 60.59% relative to experimental control. **Table-3** showed effect of methanolic extract on fluid accumulation and electrolytes secretion. The mean volume of

intestinal fluid was seen to be decreasing from control 2.18±0.31 to 1.32 ± 0.26 with 200mg/kg extract to 0.92 ± 0.29 with 400mg/kg of the extract and finally, loperamide 0.70 ± 0.24. All the treatments inhibited fluid accumulation 39.45%, 57.80% and 67.89 respectively. The treatment of rats with *A. leiocarpus* methanolic extract significantly ($p<0.05$) reduced the Na^+ concentration in the intestinal fluid at both doses of 200mg /kg and 400mg/kg. It also significantly ($p<0.05$) decreased the K^+ compared with experimental control. The Cl^- on the other hand, was shown it to increase dose dependently with 200mg/kg having 77.23 ± 1.87 meq/l and 400mg/kg with 89.75 ± 3.72 meq/l relative to control with value of 47.33 ± 1.12 An inverse relationship was observed between Cl^- and Na^+/K^+.

Table-1: Phytochemical Composition of Stem Bark Extract of *A. leiocarpus.*

Phytochemicals	**Methanol**
Saponins	+
Flavonoids	+
Terpenoids	+
Alkaloids	+
Anthraquinones	_
Tannins	+
Glycosides	+
Proteins	+

Key: +=Present =Absent

Table-2: Effect of Methanolic Extract on Castor Oil-induced Intestinal Propulsion (Transit time).

Treatment/Dose	**Mean length of intestine (cm)**	**Mean distance traversed by charcoal meal(cm)**	**Mean% movement of charcoal meal (cm)**	**%inhibition**
Control	88.84±1.56	64.88±0.45	73.22±0.14	_
Extract 200mg/kg	80.52±2.34	47.42±1.31	58.40±0.27*	41.11
Extract400mg/kg	84.52±1.46	41.16±0.48	48.60±0.28*	51.32
(Standard) Loperamide2mg/kg	80.22±2.32	31.62±0.41	39.51±0.11*	60.59

Results are expressed as means ± SD, n=5* Significance relative to experimental control ($p<0.05$)

Table-4, showed curative effect of methanolic extract administered 30 minutes after diarrhoea induction. All the rats in all the four groups were observed of passing watery stool after thirty minutes of diarrhoea induction. The onset of diarrhoea could not start in all the rats at the same time. The time interval observed was not up to twenty minutes. Treatment with 200mg/kg and 400mg/kg with methanolic extract reduced the frequency of stool passage with 10.00% and 24.00% respectively relative to experimental control. The standard drug (Loperamide 2mg/kg) reduced markedly the frequency of stooling with 67.00% curative index relative to experimental control. All these were observed under four hours.

Table-3: Effect of Extract on Castor Oil-induced Fluid Accumulation and Electrolytes Secretion.

Treatment	Mean volume of Intestinal fluid (ml)	%inhibition	Na^+ meq/L	K^+ meq/L	Cl^- meq/L
Control	2.18 ±0.31	_	145.57 ± 5.04	12.02 ±1.08	47.33 ±1.12
Extract 200mg/kg	1.32 ±0.26*	39.45	134.60 ± 1.28	7.57 ±0.26	77.23 ±1.87
Extract 400 mg/kg	0.92 ±0.29*	57.80	119.25 ± 3.35	7.15 ±0.23	89.75 ±3.72
(Standard) Loperamide 2mg/kg	0.70 ±0.24*	67.89	104.74 ±4.02	7.11 ±0.05	103.98 ±5.86

Results are expressed as means ± SD, n=5 *Significance relative to experimental control (p<0.05)

Table-4: Curative Effect of Methanolic Extract on Castor Oil-induced Diarrhoea.

Treatment/ Dose	Presence of watery stool	No. of watery stool	% Curative index
Castor oil 2	5(100%)	37.8±1.30	_
Extract 200	5(100%)	34.0±1.58*	10.00
Extract 400	5(100%)	28.6±1.67*	24.00
Loperamide 2	5(100%)	12.6±1.95*	67.00

Results are expressed as means ± SD, n=5*Significance relative to experimental control (p<0.05)

Table-5: Preventive Effect of Methanol Extract on Castor Oil-induced Diarrhoea.

Treatment Dose (mg/kg)	Presence of watery stool	No. of watery stool	% Preventive Index
Castor oil 2	5(100%)	35.6±2.30	_
Extract 200	5(100%)	28.2±1.52*	20.79
Extract 400	4(80%)	19.6±1.14*	44.99
Loperamide	3(60%)	14.6±8.44*	58.99

Results are expressed as Means ± SD, n=5 *Significance relative to experimental control (p<0.05)

Table-5 showed preventive effect of methanolic extract administered 30 minutes before castor oil administration to induce diarrhoea. All the rats (100%) in control (group I) and (group II) treated with 200 mg/kg body weight methanolic extract were observed passing watery stool. And in group III pretreated with 400mg/kg body weight methanolic extract, only eighty percent (80%) were observed with diarrhoea while in group IV pretreated with Loperamide 2mg/kg (standard drug), only sixty percent (60%) were noted passing semi solid stool and in a minimal number. Under four (4) hours of observation, both 200 mg/kg, 400 mg/kg and loperamide 2mg/kg body weight exhibited a significant ($p<0.05$) decrease in wet stool (diarrhoea) numerically represented by 20.79%, 44.94% and 58.99% preventive (inhibition) index respectively relative to experimental control.

DISCUSSION

The phytochemical constituents identified in the stem bark extracts of *A. leiocarpus* are flavoniods, tannins, saponins, glycosides, steroids and or terpenes. The positivity of terpenes in methanol is in agreement with the findings of Venketesan *et al* [24] reported the presence of tarpenes in methanolic extract of *Asparagus racemosus*. Previous reports have demonstrated the antidirrhoeal activity of Tannins [21], flavonoids, alkaloids, (saponins, reducing sugars and terpenes [25]. They have been established to be frequently responsible for most medicinal plants. It is well documented that castor oil produced diarrhoea due to ricinoleic action which stimulate peristaltic (contraction) activity in the small intestine leading to hyper secretory response and electrolytes permeability changes in intestinal mucosa [9, 26]. Loperamide inhibits gastrointestinal motility by effects on circular and longitudinal muscles of the intestine. Part of its antidiarrhoeal effect may be due to a reduction of gastrointestinal secretion produced by actions at opioid receptors in the intestinal mucosa. Consequently, it inhibits the release of acetylcholine and prostaglandins, thereby reducing peristalsis and increasing intestinal transit time [27]. The extract appears to acts on all parts of the intestine. Probably by relaxing intestinal smooth muscle permitting enough time for reabsorption of water and electrolytes to take place. The sesquiterpene lactones, a large group of compounds with anti-inflammatory properties have the ability to relax smooth muscles and thereby relieve gastrointestinal distress [27].

Decrease intestinal motility (suppressed propulsion of charcoal meal) was observed in dose dependent manner compared to experimental control. The inhibitory activity of flavonoids on intestinal motility in a dose related manner was reported [18, 28]. The antidiarrhoeal activity of flavonoids have been attributed to their ability to inhibit intestinal transit and hydro-electrolytes secretion [18, 29] Sesquiterpene, diterpenes, terpenes, flavonoids and terpenoids derivatives are known for inhibiting release of autocoids and prostaglandins, thereby inhibit the motility and secretion induced by castor oil [30]. Vankatesan *et al* [24] reported that, 200mg/kg exract of *Asparagus racemosus* suppressed motility similar to 0.1mg/kg atropine. In this study, the extract demonstrated nonspecific spasmolytic activity. It is widely known that 90% ricinoleic in castor oil Mekeon *et al* [31], when metabolized induces permeability changes in mucosal fluid and electrolyte(s) transport that results in hyper secretory response and diarrhoea [24].Castor oil is also reported to induce diarrhoea by increasing the volume of intestinal content by prevention of the reabsorption of water. Liberation of ricinoleic causes irritation and inflammation of intestinal mucosa leading to release of prostaglandins, which results in stimulation of secretion. Thereby prevents the reabsorption of NaCl and H_2O [32]. 200mg/kg, 400mg/kg extract reduced fluid content by 39.45%and 57.80% respectively. The antidiarrhoeal activity may be due to presence of tannins that formed protien tannates (denatured proteins), protein tennates make the intestinal mucosa more resistant and reduce secretion [33].

This study also showed dose dependent decrease in intestinal Na^+ and k^+ concentrations. The extract may have altered the activity of intestinal Na^+k^+ATPase. Several mechanisms have been previously propose to Na^+, K^+ ATPase activity to reduce normal fluid absorption [34]. The secretory diarhhoea is associated with impaired sodium absorption by the villi, and activation of Cl^- channels in the crypts cells. Causing water and salts loss from the cell of intestinal lumen [34]. Inappropriate chloride channels may have been activated leading to increase in intestinal levels of Cl^- as observed in this study. Antidiarrheal treatment in patient is achieved through the objective of the therapy which includes increasing resistance to flow (segmental contraction and decrease propulsion) and increased mucosal absorption or decreasing secretion [34]. Post administration of the extract 200 and 400mg/kg, revealed lower curative percentages compared to their pre-administrative values under 4hrs.

Among the several mechanisms proposed to explain the diarrheal effect of castor oil are activation of adenylate cyclase or mucosal cAMP mediated active secretion [15]. The extract tested at 200mg/kg and 400mg/kg significantly inhibited (prevented) the frequency of wet stool compared to control. The extract may have acted by inhibition of (inactivation) of adenylate cyclase. This result is in agreement to the findings of Bakeri *et al* [16] who reported that, 200 and 400mg/kg aqueous leave extract of *Momordica charantia* in rats like the standard drug, morphine significantly inhibited the frequency of defecation compared to control. It also supports the previous claims that antidiarrhoeal plants are known to reduce number of wet stools as reported for *Eremomastax speciosa* and *Xylocarpus granatum* [36, 37]. The preventive capacity of this plant, makes it beneficial as preventive agent.

CONCLUSION

Based on the results of the present study, it could be said that the plant extracts contain chemical constituents of pharmacological significance. The presence of these chemical constituents in this plant is an indication that the plant, if properly screened using additional solvents other than methanol could yield drugs of pharmaceutical significance. It also justified the use of stem bark extracts of *A. leiocarpus* by traditional medicine practitioners in the treatment of both infectious and non-infectious diarrhea.

REFERENCES

1. P. Sharma, G. Vidyasagar, S. Singh, B. Kumar. Antidiarrhoeal activity of leaf extract of Medicinal Plants. *(Phytotherapeutics)*. 2010, **10**: 375-380.
2. N. Venkatesan, V.J. Vadivu, R. Thandavarayan and B.P. James. Anti-diarrhoeal potential of *Asparagus racemosus* wild root extracts in laboratory animals. *J. Pharm. Pharmaceut. Sci*. 2005, **8** (1): 39-46.
3. A.C. Guyton. Textbook of Medical Physiological 8th edn. W.B Saunders Company. Saunders, Japan. 1991, 136-146.
4. J.D. Snyder and M.H. Merson. The magnitude of the global problem of acute diarrhea: A review of active surveillance of data. *Bull WHO*. 2002, **60**: 605-613.
5. K. Park. Park's Textbook of Preventive and Social Medicine. Banarsidas Bharat. Publishers Jabalpur. 2000, 122-175.
6. E.E. Ekanem and N.S. Benebo. Proportionate morbidity ratio and aetiology of acute diarrhoeal diseases in infants and young children in a deprived peri-urban area of Lagos, Nigeria. *Early Child Develop Care*. 2000, **54**: 1-9.
7. A.K. Das, S.C. Manda, S. Bernerel, J. Das, B.P. Saha and M. Pal. Studies on antidiarrheal activity of *punicagranatham* seed extract in rats. *J. Ethnopharm*. 2009, **69**: 205-208.
8. A. Mann, J.O. Amupitan, A.O. Oyewal, J.I. Okogun, K. Ibrahim. An ethnobotanical survey of indigenous flora for treating tuberculosis and other respiratory diseases in Niger state. *J. Phytomed. Therap*. 2007, **12**: 1-12.
9. J.G. Hardman and L.E. Limbird. *The Pharmacological Basis of Therapeutics*. 10th edition, McGraw Hill, New York. 2001, 941-932.
10. E. U. Etuk, M.B. Agale, P.A. Onyeyili and C.U. Ottah. Antidiarrhoeal effect of *Boswellia dialzeilli* stem bark extract in albino rats. *J. Pharamcol. Toxicol*. 2006, **(6)**: 211-215.

11. A.Y. Bala and T. Adamu. Antidiarrhoeal activity of *Psidium guajava* aqueous leaf extract in experimental animals. *Nig. J. Basic Appl. Sci.* 2008, **16** (2): 182-187.
12. O.P. Ajagbonna and P.A. Onyeyili. Effects of *Rhaptopetalum coriaceum* o/w (*Scypetalaceae*). *African Journal of Biotechnology*. 2002, **7** (6): 880-87.
13. G.E. Trease and W.C. Evans. *Pharmocognosy*. 12Ed. Baillieere Tindal, London. 1983, 423-442.
14. J.B. Harborne, W.C. Evans. Phytochemical Methods. A guide to Modern Techniques of plant Analysis. 3rd edition. Chapman and Hall, New York. 1998, 1-198.
15. F. Capasso, G. De–Ruggiero, M. Derosa and L. Serrenteno. RecehefarmacologicheSulla – 4-amino-5- chloro-4 (2 di-etilaminoetil) 2-methossibenamide. *Bolettinochemico fermaccnticco.* 2006, **115**: 649-657.
16. R.I. Bakare, O.A. Magbagbeola, A.I. Akinwande and O.A.T. Ebuehi. Effect of aqueous leaf extract of *Momordica charantia* on intestinal enzyme activities in diarrhoeagenic mice. *Nig. Quart. J. Hosp. Med.* 2011, **20** (1): 24-28.
17. S.S. Rajahbau, V.B. Karnakumar, V.C. Basararay and J. Shambulingaya. *In-vitro* antidiarrhoeal activity of ethanolic extract of *Delonix regia* flowers in experimental induced diarrhea in wister albino rats. *Int. J of Pharm and chemistry*. 2011, **1** (3): 443-444.
18. G.D. Di-carlo, N. Mascolo, A.A. Izzo and F. Capasso. Effect of quercetin on the gastrointestinal tract in rats and mice. *Phytother. Res*. 1994, **8**: 42-45.
19. B.S. Adzu, S. Amos, M.B. Amizan and K.S. Gamaniel. Evaluation of the antidiarrhoeal effects of *Spina-christi* stem bark in rats. *Acta Trop.* 2003, **6** (1): 1-5.
20. R. Boominathan, B.P. Devi, S. Dewanjee and S.C. Manda. Studies on antidiarrhoeal activity of *Ionodium suffruticosamging,* (violaceae) extract in rats. Recent Progress in. *Celosia argentea* in experimentally induced diarrhoea in rats. *J. Adv Pharm Tech Res.* 2005, **1**: 41-8.
21. P.K. Mukherjee, K. Saha, T. Murugesan, S.C. Pandal, M. Pal and B.P. Saha: Screening of antidiarrhoeal profile of some plant extracts of a specific region of west Bengal. *India J. of Ethnopharm*. 1998, **60**: 85-89.
22. F. Awouter, C.V.E. Neimegeers, F.M. Lenart and P.A.J. Janssen. Delay of castor oil diarrhoea in rats: A new way to evaluate inhibitors of prostaglandins biosynthesis. *J. Pharm and Pharmacol*. 1978, **30**: 41-45.
23. E.O. Julius, E. Sheila, G.A.A. Assi, F.M. David. Effect of *Eremomastax Speciosa* on experimental diarrhoea. *Afr. J. Trad. CAM*. 2006, **3** (1): 95-100.
24. N. Venkatesan, V.J. Vadivu, R. Thandavarayan and B.P. James. Anti-diarrhoeal potential of *Asparagus racemosus* wild root extracts in laboratory animals. *J. Pharm. Pharmaceut. Sci*. 2005, **8** (1): 39-46.
25. S. Gricilda and M. Thomas. Study of anti-diarrhoeal activity of four medicinal plants in castor oil-induced diarrhoea. *Journal of Ethnopharmacology*. 2001, **76**: 73-76.
26. M.A. Zavala, S. Perez, C. Perez, R. Vargal and R.M. Perez. Antidiarrhoeal activity of *Wiltheria* a*mericana, Commelins cuelestris* and *Altternanthra rapens. J. Ethnopharmacol*. 1998, **61**: 41-47.

27. M. Heinrich, M. Robles, J.E. West, B.R. Ortizde-Mentellano and E. Rodriguez. Ethnopharmacology Mexican asteraceae. *Annu.Rev.Pharmacol.Toxicol.* 1998, **38**: 539-65.
28. R. Meli, G. Autore, G. Dicarlo and F. Capasso. Inhibitory action of *quercetin* on intestinal transit in mice *J. Pytother. Research*. 1990, **4**: 201-202.
29. V.S.N. Rao, F.A. Santos and T. Sobreika. Investigation on the gastroprotective and antidiarhoeal properties of ternatin, tetramethoxyflavone from *Eaglets viscose*. *J.phytomed*. 1997, **7**: 172-187.
30. R.C. Havagiray, C. Ramesh and K. Sadhna. Studies on anti-diarrheal activity of *Calostropis gigantea* in experimental animals. *J.Phar.Pharmaceut.Sci.* 2005, **7** (1): 70-75.
31. T.A. Mekeon, J.J. Linn, A.E. Stafford. Biosynthesis of ricinoleate in castor oil. *Adv. Exp. Med. Biology.* 1999, 46437-46447.
32. R.C. Havagiray, C. Ramesh and K. Sadhna. Studies on anti-diarrheal activity of *Calostropis gigantea* in experimental animals. *J.Phar.Pharmaceut.Sci.* 2008, **7** (1): 70-75.
33. K.D. Tripathi. Essentials of medical pharmacology. Jaypee Brothers Medical Publishers New Delhi. 1994, 775.
34. J.A. Guttman and T. Finlay. Proteinase inhibitors TPCK and TLCK prevent *Entamoeba histolytica* induced disturbance of tight junctions and microvilli in enteric cell layers invitro. *Int. J. Parasitol.* 2009, **34**: 785-94.
35. World Health Organisation. *Manual for treatment of diarrhoeal diseases*. WHO/CDR: Geneva, 3. 2004.
36. J.E. Oben, S.E. Assi, G.B. Agbor and D.F. Musoro. Effect of *Eremomastax speciosa on* experimental diarrhea. *Afr. J. Tradit. Compl.Altern. Med.* 2006, **3** (1): 95-100.
37. R. Rouf, S.J. Uddin and M. Alamgir. Assessment of antidiarrhoeal activity of the methanol extract of *Xylocarpus granatum* bark in mice model. *J .Ethnopharmacol*. 2007, **109**: 539-542.

***Corresponding author: Memi G.G;**
Department of Biological Sciences, Federal University,
Kashere P.M.B 0182 Gombe state Nigeria.
On line publication Date: 02.08.2017