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*Full Length Research Paper*

# Antidiarrhoeal and antibacterial properties of crude aqueous stem bark extract and fractions of *Parkia biglobosa* (Jacq.) R. Br. Ex G. Don

**A. Y. Tijani[1], S. E. Okhale [2], T. A. Salawu[3]*, H. O.Onigbanjo[3], L. A Obianodo[1], J. A Akingbasote[1], O. A. Salawu[1], J. I. Okogun[2], F. O. Kunle[2] and M. Emeje[4]**

[1]Department of Pharmacology and Toxicology, National Institute for Pharmaceutical Research and Development (NIPRD), Idu, Abuja, Nigeria.

[2]Department of Medicinal Plants Research and Traditional Medicine, National Institute for Pharmaceutical Research and Development (NIPRD), Idu, Abuja, Nigeria.

[3]Department of Microbiology, Human Virology and Biotechnology, National Institute for Pharmaceutical Research and Development (NIPRD), Idu, Abuja, Nigeria.

[4]Department of pharmaceutical Technology and Raw material Development, National Institute for Pharmaceutical Research and Development (NIPRD), Idu, Abuja, Nigeria.

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**Stem bark of *Parkia biglobosa* (Jacq.) R. Br. Ex G. Don (Fabaceae) is used in African traditional medicine for management of diarrhoea- related disorders. The anti-diarrhoeal and anti-microbial activities of the aqueous stem bark extract of *P. biglobosa* and its fractions designated PF1-PF4 were investigated in mice and against selected diarrhoea-causing micro-organisms. The oral median lethal dose ($LD_{50}$) of the extract in mice was estimated to be greater than 5000 mg/kg B.W. The extract and its column chromatographic fraction $F_3$ significantly ($p < 0.05$) and dose-dependently reduced frequency of stooling in castor- oil-induced diarrhoea, castor-oil-induced intestinal fluid accumulation and intestinal transit. The crude extract as well as fractions $F_3$ and $F_4$ strongly inhibited growth of selected micro-organisms. The study showed that the aqueous extract possess both anti-diarrhoeal and anti-microbial activities. The anti-diarrhoeal action may be linked partly to direct inhibitory effect of the extract on the propulsive movement of the gastrointestinal tract smooth muscle, and the anti-microbial effect on the diarrhoea-causing pathogenic organisms.**

**Key words:** Antidiarrhoeal, antimicrobial, *Parkia biglobosa*, fractions, castor oil.

## INTRODUCTION

Diarrhoea refers to an increase in the frequency, fluidity and/or volume of faeces, greater than normal for an indi-vidual, resulting from an imbalance between the secre-tory and absorptive forces in the intestines. It is an impor-tant symptom and complications of many diseases, and of great public health importance, as diarrhoeal diseases still account for up to 16% of under -5 mortality, and 7% of total mortality in Nigeria (World Health Organization, 2006), with similar figures in other developing countries Diarrhoea has many consequences ranging from the so-cial inconvenience of increased faecal frequency (trave-ler's diarrhoea, caused by toxins of *Escherichia coli*), to complications such as diarrhoea with blood, severe dehydration and electrolyte imbalance which can rapidly lead to death as seen in cholera, or diarrhoea in young children (Syder and Merson, 1982).

Being a common symptom of disease, many traditional remedies such as *Piliostigma reticulatum* have been used in various societies for treatment of diarrhea (Sala-wu et al., 2007). *Parkia biglobosa* (Jacq.) R. Br. Ex G. Don (family Fabaceae formerly Leguminosae, sub-family mimosoideae), synonyms *P. clappertoniana* Keay, *P. filicoidea* Benth and popularly known as the African locust bean tree, is a perennial deciduous tree with a height ranging from 7 - 20 m, bole stout, not buttressed, low-branching, bearing a large wide-spreading crown, flowering while leafless; flowers in pendulous capitula bearing also pendulous, large fruit-pods. The pendulous capitulum on a long flexible penduncle is covered with tightly packed red flowers, striking and conspicuous. Amber gum exudes from wounds; the bark is dark-grey to

*Corresponding author. E-mail: salawuta@yahoo. com. Tel.: +447502391545.

brown, thick and fissured. The fermented seeds of *P. biglobosa* known in Yoruba as *Igba, or Irugba*, in Hausa as *Dorowa* and in Ibo as *Origili*, are used in all parts of Nigeria and indeed the West Coast of Africa for seasoning traditional soups (Burkill, 1995).

Medicines derived from *P. biglobosa* are of great value torural communities that can not afford or do not haveaccess to "modern medicine". In Nigeria, to relieve diarrhoea, including diarrhoea with blood, the dried and powdered stem bark of *P. biglobosa is* boiled to make a tea or mixed with hot pap and drunk (Burkill, 1995). For infections, wounds, and fever the stem bark is boiled and applied topically (Shao, 2002). Extracts from the leaves, flowers and seeds are also used to treat various ailments ranging from toothaches, lumbago and hemorrhoids to diabetes mellitus (Hall, 1997; Abbiw, 1990; Abo; Fred-Jayesimi, 2008). In this study we have investigated the safety, anti-diarrhoeal as well as the anti- microbial activity of *P. biglobosa* stem bark infusion and its fractions in mice, and evaluated its *in-vitro* activity against micro-organisms implicated in diarrhoea toward understanding its mechanism of anti-diarrhoeal action.

## MATERIAL AND METHODS

### Animals

Albino male mice weighing 15 - 20 g were obtained from the Animal Facility Centre, (NIPRD), Idu, Abuja. The mice were fed standard laboratory diet, given water *ad libitum* and maintained under laboratory conditions of temperature 23 ± 2°C, relative humidity 60% and 12 h light; 12 h dark cycle. Food was withheld for 24 h prior to each experiment. All animal experiments complied with the "Principles of Laboratory Animal Care" (NIH Publication No. 85 - 23, revised in 1985) and NIPRD-Standard operation procedure.

### Chemicals

Castor oil (Bell, Sons and Co. Limited, Southport, England); Loperamide (Janssen Cilag); Deactivated charcoal and Methanol (Sigma); Octadecylsilyl (ODS) silica gel (Aldrich).

### Plant material

The stem bark of *P. biglobosa* was collected from Suleja (Niger State, Nigeria) by Mallam Muazzam Ibrahim an ethnobotanist in the Department of Medicinal Plant Research and Traditional Medicine, National Institute for Pharmaceutical Research and Development (NIPRD), Idu Industrial Area, Abuja, Nigeria. The leaves, fruits and stem bark were identified and authenticated by Mrs. Grace Ugbabe a taxonomist in the same department .Voucher specimen (No. NIPRD/H/6225) was deposited in the central herbarium of NIPRD. The stem bark was cleaned, air-dried at room temperature (60 - 80°F) away from sunlight and pounded into fine powder using mortar and pestle. The powder was stored in an air-tight container and kept at 39.2°F for subsequent use.

### Extraction method

Two litres of boiled distilled water was added to 200 g of the sample in a 5 litres flat bottom flask, stirred with a glass rod, covered, shaken continuously for 6 h using GFL shaker (TUV Product Serice, Germany) and then allowed to stand for another 18 h at room temperature. The extract was then filtered using Whatmann filter paper No. 91 (18.5 cm). The filtrate was evaporated to dryness on a water bath to obtain hot water extract of *P. biglobosa* (HWEPB).

### Phytochemical screening

Phytochemical screening was carried out on HWEPB using standard methods (Odebiyi and Sofowora, 1978; Trease and Evans, 1996) for detecting the presence of secondary metabolites; alkaloids, carbohydrates, free reducing sugars, combined reducing sugars, tannins, saponins, glycosides, sterols, terpenes and flavonoids.

### Fractionation

HWEPB was subjected to column chromatography using Octadecylsilyl (ODS) silica gel stationary phase 300 and 12.37 g of HWEPB. Gradient elution under gravity was performed with 500 ml of each mobile phase mixture in series. The mobile phase consists of distilled water and methanol, starting with distilled water (100%) and 10% increments in methanol. The final elution was performed with 100% methanol. A total of 35 fractions were obtained. The eluates were monitored with thin layer chromatography; eluates with similar Rf values were pooled to obtain four fractions (PF1 to PF4).

### Acute toxicity ($LD_{50}$) study

Acute toxicity study was carried out according to the modified method described by Lorke, 1983. The study was carried out in two phases. In the first phase, nine mice were randomized into three groups of three mice per group and given 10, 100 and 1000 mg HWEPB /kg body weight (b. w.) orally (via cannula), respectively. Animals were observed for 24 h after treatment for signs of toxicity and mortality. The results of this phase informed the choice of doses for the second phase, in which 1600, 2900 and 5000 mg HWEPB/kg b.w. were given orally to another fresh set of three mice per group. The mice were also observed for signs of toxicity and mortality. The final $LD_{50}$ value was calculated as the square root of the product of the lowest lethal dose and the highest non-lethal dose, that is, the geometric mean of the consecutive doses for which 0 and 100% survival rates were recorded in the second stage.

### Antidiarrhoeal activity of crude *P. biglobosa* extract and fractions

**Induction of diarrhoea with castor oil:** Diarrhoea was induced by single dose oral administration of castor oil. Anti-diarrhoeal activity of the extract and fractions were evaluated using the castor oil-induced diarrhoea model in mice (Capasso et al., 2007). Twenty five mice were randomized into five groups of five mice each. Group 1, which served as the negative control was given 5 ml normal saline/kg b.w. orally. Group II – IV were given 100, 200 and 400 mg HWEPB/kg b. w. orally while the positive control group received 10 mg loperamide/kg body weight. One hour after the treatment, mice in all the groups were given 0.5 ml castor oil/ 10 g b.w. orally. The mice in each group were then placed singly in cages with adsorbent paper on their floors. The diarrhoeal episodes were observed for 4 h and the cumulative frequency of wet and formed stools was noted at the end of the $4^{th}$ h. Percentage inhibition of diarrhoea was calculated using the mean stool frequency and anti-diarrhoeal activity determined in terms of percentage protection.

**Castor oil-induced enteropooling:** Intestinal fluid accumulation was determined by the method of Robert et al. (1976). Twenty-five rats were randomly divided into five groups of five mice each. Group I served as negative control and received 5 ml normal saline/kg b.w., intraperitoneally, while Groups II, III and IV received

100, 200 and 400 mg HWEPB/kg b. w., respectively. Group V received 3 mg Atropine (standard drug)/kg b. w. and served as positive control. One hour later, 0.5 ml castor oil/ 10 g b.w. or all was administered orally, and after two hours, all mice were sacrificed and their small intestines (pylorus to caecum) removed after ligating the ends. Intestinal contents were collected by milking into a graduated tube, and the volumes measured and recorded. Percentage inhibition of enteropooling was determined by calculating the mean volume of intestinal contents and comparing it with values from the negative control group.

**Intestinal Transit Test:** The effect of the extract on the gastrointestinal motility was evaluated as previously described (Salawu et al., 2007). Twenty-five mice were randomly divided into five groups of five mice each and fasted for 24 h prior to the test, but allowed free access to water. Group I served as negative control and was treated with 0.5 ml Normal saline/kg b.w. Group II served as positive control and was treated with 10 mg loperamide (standard drug)/kg b. w. orally. Groups III, IV and V received 100, 200 and 400 mg HWEPB/kg b. w. orally, respectively. After 30 min, 0.5 ml of marker - charcoal meal (deactivated charcoal suspension in 5% tragacanth) was administered orally to all animals and 30 min later, all were sacrificed. The distance traveled by the marker was then measured and expressed as a percentage of the total length of the small intestine (pylorus to caecum).

Following column chromatographic fractionation, each fraction PF1, PF2, PF3 and PF4 was evaluated separately for activity on diarrhoea frequency, enteropooling and intestinal transit as described above. For each test, nine mice were randomly grouped into three groups of three mice each. Group I, the negative control group received 5 ml Normal Saline/kg b. w. Groups II and III received 50 and 100 mg of the fraction/kg b. w., respectively. Testing was carried out as described above for HWEPB and results recorded.

#### Antimicrobial studies

**Preparation of agar for antimicrobial studies:** The procedure employed was a modification of the pour -plate technique of Hugo and Russell, 1992. Mueller-Hinton agar (260 ml) was prepared (following manufacturer's instruction) and dispensed in 13 McCartney bottles of 20 ml each and sterilized by autoclaving (at $121^{o}$C, 2 atm. for 15 min). The molten MHA was then stabilized in a water bath at 45°C until ready for use.

**Inoculation of diarrhoea-causing micro-organisms:** The inocula of five test isolates (*Candida albicans, Staphylococcus aureus, E. coli, Shigella dysenteriae and Pseudomonas aeruginosa)* were prepared directly from colonies on agar plates.

Under asepsis, colony of each isolate was transferred into sterile water until the density of the inoculums equaled the turbidity of Barium sulphate standard (0.5 MacFarland), which represents $10^5$ cfu/ml. The accuracy of the density of prepared standard was verified using a spectrophotometer. For 0.5 MacFarland, the $A_{625}$ = 0.08 – 0.10.One mililitre each of the adjusted isolate inoculums was transferred to 5 of the bottles of molten MHA, mixed, poured onto sterile Petri dishes and then allowed to gel. This was done in duplicates for each isolate. Cork borer No. 4 (8 mm diameter) was used to make five neat cups in each of the organism- containing media and the base of each cup sealed with 2 drops of molten MHA.

**Preparation of extract and fractions for antimicrobial studies:** Five different concentrations namely: 10, 5, 2.5, 1.25 and 0.625 mg/ml of HWEPB and the fractions (PF1,PF2, PF3 and PF4) were prepared by dilution in sterile distilled water and dispensed aseptically into the 5 neat cups in the organism-containing media prepared above. Organism Viability Control (OVC), Extract Sterility Control (ESC) and Medium Sterility Control (MSC) were also prepared, and along with the prepared media above, incubated for 24 h at 37°C. The diameter of the zones of Growth Inhibition were then measured and recorded.

#### STATISTICAL ANALYSIS

All numerical data are expressed as the mean ± standard error of mean (SEM). Statistical analysis was carried out using analysis of variance(ANOVA),followed by Duncan Multiple Range Test and differences between means were considered to be significant when $p < 0.05$ and $P < 0.01$ respectively.

## RESULTS AND DISCUSSION

The yields of the extracts were 9.0% w/w, 21.3% w/w, 23.2% w/w, 38.9% w/w, and 6.7% w/w for crude, PF1, PF2, PF3 and PF4 respectively.

### Phytochemical composition

Results of phytochemical screening of the hot water extract of *P. biglobosa* stem bark (HWEPB) and the PF3 is shown in Table 1.

**Acute toxicity:** There were no lethality or toxic reactions found at any of the doses of hot water extract of *P. biglobosa* used in the study. All the animals were alive, healthy and active during the observation period. The oral median lethal dose of *P. biglobosa* was therefore estimated to be greater than 5000 mg HWEPB/kg body weight in mice.

**Effect of HWEPB on diarrhoea frequency:** There was a significant and dose-dependent ($P < 0.05$) reduction in the frequency of diarrhoea among the mice in the 200 and 400 mg HWEPB/kg b.w. groups, which compares favorably with reduction in frequency of diarrhoea produced by 10 mg/kg b.w. of standard drug - loperamide .Significant ($P < 0.05$) reduction in diarrhoea frequency was seen at 100 mg PF3/ kg b.w. There was no significant difference in reduction of diarrhoea frequency caused by fraction PF3 and 10 mg Loperamide/kg b.w. (Table 2).

**Effects of HWEPB and PF3 on castor oil- Induced enteropooling:** Results in Table 3 showed that HWEPB and PF3 dose-dependently inhibits intestinal secretion significantly ($P < 0.05$), an effect that favorably compares with inhibition caused by 3 mg atropine/kg body weight at 400 mg HWEPB/kg bodyweight. This reduction was both dose-dependent and compared favorably with reduction seen in atropine.

**Effects of HWEPB and PF3 on intestinal transit in mice:** HWEPB caused significant ($P < 0.05$) reduction in distance traveled in the intestine by the marker in a dose-dependent manner. Loperamide at 10 mg/kg body weight, however, highly- significantly ($P < 0.01$) reduced the distance traveled by the marker and thus the inte-stinal transit time. At 50 mg/kg b. w., PF3 caused a significant ($P < 0.05$) reduction in intestinal transit time while at 100 mg/kg b. w., it caused a highly significant ($P< 0.01$)

**Table 1.** Results of phytochemical screening of the hot water extract of *Parkia biglobosa* stem bark (HWEPB) and PF3.

| Secondary metabolites | HWEPB | PF3 |
|---|---|---|
| Carbohydrates | + | - |
| Free reducing sugars | + | + |
| Combined reducing sugars | ++ | ++ |
| Tannins | +++ | +++ |
| Alkaloids | + | - |
| Saponins | + | + |
| Terpenes | - | - |
| Sterols | - | - |
| Anthraquinones | - | - |
| Cardiac glycosides | - | - |
| Flavonoids | - | - |

+ Slightly present; ++ present; +++ highly present; - Absent. PF3 is the most antidiarrhoeal-active column chromatographic fraction of HWEPB.

**Table 2.** Effect of hot water extract of P. biglobosa (HWEPB) and PF3 on castor oil-induced diarrhoea in mice.

| Treatment | Mean ± SEM (hours) Frequency of diarrhoea in 4 h | % Inhibition |
|---|---|---|
| 5 ml/kg Normal Saline | 4.00 ± 0.28 | 00.00 |
| 100 mg HWEPB/kg | 1.80 ± 0.52 | 55.00 |
| 200 mg HWEPB/kg | 0.80 ± 0.18* | 80.00 |
| 400 mg HWEPB/kg | 0.60 ± 0.36* | 90.00 |
| PF3 | | |
| 50 mg PF3/kg | 1.20 ± 0.54 | 70.00 |
| 100 mg PF3/kg | 0.80 ± 0.33* | 80.00 |
| 10 mg Loperamide | 0.80 ± 0.33* | 80.00 |

*- Significantly different from control at P 0.05 **Highly significantly different from the control at P 0.01.

**Table 3.** Effect of hot water extracts of Parkia biglobosa (HWEPB) and PF3 on castor oil-induced intestinal fluid accumulation.

| Treatment | Mean ± SEM Volume of Intestinal Contents | % Inhibition of Secretion |
|---|---|---|
| 5 ml/kg Normal Saline | 1.42 ± 0.05 | 66.90 |
| 100 mg HWEPB/kg | 0.47 ± 0.04 | 85.92 |
| 200 mg HWEPB/kg | 0.20 ± 0.00* | - |
| 5 ml/kg Normal Saline | 1.20 ± 0.10 | 81.67 |
| 50 mg PF3/kg | 0.22 ± 0.01* | 85.00 |
| 100 mg PF3/kg | 0.18 ± 0.02* | 90.14 |
| 3 mg Atropine/kg | 0.14 ± 0.03* | |

*Significantly different from control at P 0.05.

reduction in the distance traveled by the marker. This reduction compares favorably with loperamide at 10 mg/kg. This is shown below in Table 4.

**Antimicrobial effects of HWEPB and fractions (PF1-PF3) on microorganisms implicated in diarrhea:** HWEPB and its four column chromatographic fractions PF1, PF2, PF3 andPF4 were tested against five pathogenic organisms at concentrations of 10, 5, 2.5, 1.25 and 0.625 mg/ml. As shown in Table 5, they showed varying degrees and specificity of activity against these bacteria. Growth inhibitory activity was seen against *S. aureus, E. coli and S. dysenteriae* at 10 mg/ml concentration of the aqueous extract, with less activity against S. aureus at 5

**Table 4.** Effect of hot water extract of Parkia biglobosa (HWEPB) and PF3 on intestinal transit time in mice.

| Treatment | Mean Intestinal Length (cm) | Mean Distance traveled by marker (cm) | Intestinal Transit % | Inhibition % |
|---|---|---|---|---|
| 5 ml/kg Normal Saline | 34.20 ± 2.29 | 33.20 ± 2.18 | 97.08 | - |
| 100 mg HWEPB/kg | 37.40 ± 2.60 | 17.20 ± 3.25* | 45.99 | 51.09 |
| 200 mg HWEPB/kg | 40.90 ± 0.93 | 13.70 ± 2.00* | 33.50 | 63.58 |
| 400 mg HWEPB/kg | 36.50 ± 2.22 | 5.25 ± 1.02* | 14.38 | 82.70 |
| 50 mg PF3/kg | 29.38 ± 4.00 | 3.93 ± 0.24* | 13.37 | 76.88 |
| 100 mg PF3/kg | 34.50 ± 4.58 | 2.27 ± 0.26** | 6.58 | 86.65 |
| 10 mg Loperamide/kg | 35.25 ± 1.60 | 2.8 ± 0.60** | 7.94 | 89.14 |

* - Significant different from the control at P 0.05 ** - Highly significantly different from the control at P 0.01.

mg/ml concentration. The fractions PF3, PF4 showed activity against the three organisms above in addition to *C. albicans* at 10.0 and 5.0 mg/ml, respectively. Fraction PF3 also showed slight activity against *P. aeruginosa* at 10 mg/ml.

The extent of growth inhibition by Fraction PF3 was greater than that of HWEPB at the same dose against all the microorganisms tested. The OVC, ESC and MSC values were normal.

Qualitative phytochemical analysis of the aqueous stem bark extract of *P. biglobosa* revealed the presence of tannins, alkaloids and reducing sugars, and qualitative phytochemical analysis of the fraction PF3 revealed the presence of tannins, reducing sugar and saponins. The presence of this secondary metabolite may be responsible for the antidiarrhoeal properties of the crude extract and the fraction PF3.This result is consistent with previous studies. (Longanga-Otshudi et al., 2000; Al-rehaily et al., 2001). These authors opined that the antidiarrhoeal and anti-dysenteric properties of medicinal plants were due to tannins, alkaloids, saponins, reducing sugars, sterols and triterpenes. A possible mechanism may be by precipitation of proteins in enterocyte and production of protein tannates that lead to reduced secretion and peristaltic movement (Yu et al., 2000) The lack of death at oral treatment of over 5000 mg extract/kg body weight obtained suggests that ethanolic stem bark extract of *P. biglobosa* is practically non-toxic acutely (Corbett et al, 1984). The safety margin of the extract of *P.biglobosa* is higher than that of atropine and loperamide respectively. It is therefore safe acutely for oral use in the ethno-therapeutic management of diarrhoea. The high safety profile obtained may have been responsible for its wide spread use in different ethno-therapeutic interventions. The extract showed significant activity in reducing the frequency of castor oil-induced diarrhoea, but enteropooling and intestinal transit time in a manner that is comparable with loperamide. Loperamide is a commonly-used opioid anti-diarrhoeal agent which acts by increasing colonic phasic segmenting activities through inhibition ofpresynaptic cholinergic nerves in the submucosal and myenteric plexuses. These effects result in increased colonic transit time and feacal water absorption thus reducing the frequency of defeacation (Camilleri, 2004). In addition Loperamide has anti-secretory activity against cholera toxin and some forms of *E. coli* toxin, presumably by acting on Gi-linked receptors and countering the increase in cellular cyclic AMP generated in response to the toxin. However, atropine an anti-muscarinic agent is known to reduce both tone and propulsive movement of the gastro-intestinal tract from the stomach to the colon. Following fractionation, these effects were seen at even lower doses in the fraction PF3 of the aqueous extract.

The Castor Oil model for diarrhoea was chosen for these pharmacological studies, because it increases the volume of intestinal contents by preventing water reabsorption from the intraluminal space, stimulates secretion by mucosal glands (Pierce et al., 1971; Gaginella et al., 1975). Castor Oil therefore induces diarrhoea in a manner similar to all the three broad classes of pathogenetic. This present work shows that *P.biglobosa* aqueous stem bark extracts and fractions has potentials for use in reversing diarrhoea resulting from a wide variety of causes.

Anti-microbial studies of *P.biglobosa* extracts showed significant growth inhibition of *S. aureus*, *E. coli* and *S. dysenteriae*, all of which are implicated in diarrhoea and dysentery (bloody and/or mucoid diarrhoea). In addition to this growth inhibition of greater magnitude with PF3, the fraction also had wider spectrum of anti microbial activity as it inhibited the growth of *P. aeruginosa* and *C. albicans*. Our results suggest that the anti-diarrhoeal effect of the extract may be mediated via antagonism of gastro-intestinal cholinergic receptors and growth of micro-organisms implicated in diarrhoea.

Results obtained in this study showed that the hot water extract of stem bark of *P. biglobosa* contains active principle with remarkable anti-diarrhoeal and antibacterial properties. The post-fractionation studies showed that these active principles are found predominantly in the fraction PF3.

This study thus provides plausible explanations for the observed efficacy of powdered stem bark of *P. biglobosa* taken mixed with hot pap or as infusion in the treatment of diarrhea and dysentery in the traditional system. The hot water extract thus have potentials for use in management of toxin-induced diarrhoea (for example, cholera, shigellosis, and traveler's diarrhoea), diarrhoea due to

**Table 5.** Zones (mm) of growth inhibition by hot water extract of Parkia biglobosa (HWEPB) and its four fractions PF1 to PF4.

| | Conc. (mg/ml) | Candida albicans | Staphylococcus aureus | Escherichia coli | Shigella dysenteriae | Pseudomonas aeruginosa |
|---|---|---|---|---|---|---|
| **HWEPB** | 10.0 | - | 10 | 8 | 2 | - |
| | 5.0 | - | 4 | - | - | - |
| | 2.5 | - | - | - | - | - |
| | 1.25 | - | - | - | - | - |
| | 0.625 | - | - | - | - | - |
| | PF1 | | | | | |
| | 10 | - | - | - | - | - |
| | 5.0 | - | - | - | - | - |
| | 2.5 | - | - | - | - | - |
| | 1.25 | - | - | - | - | - |
| | 0.625 | - | - | - | - | - |
| | PF2 | | | | | |
| | 10.0 | - | - | - | - | - |
| | 5.0 | - | - | - | - | - |
| | 2.5 | - | - | - | - | - |
| | 1.25 | - | - | - | - | - |
| | 0.625 | - | - | - | - | - |
| | PF3 | | | | | |
| | 10.0 | 8 | 18 | 16 | 2 | 1 |
| | 5.0 | 4 | 4 | 10 | - | - |
| | 2.5 | 3 | - | - | - | - |
| | 1.25 | 2 | - | - | - | - |
| | 0.625 | - | - | - | - | - |
| | PF4 | | | | | |
| | 10.0 | 4 | 6 | 10 | - | - |
| | 5.0 | 2 | - | 6 | - | - |
| | 2.5 | - | - | - | - | - |
| | 1.25 | - | - | - | - | - |
| | 0.625 | - | | - | - | - |

intestinal malabsorption (for example. radiation enteritis, lactose intolerance) or dysentery due to microbial infections.

Further studies are ongoing in our laboratory to isolate, purify and characterize the active principles in PF3 and determine its mechanism of action in reducing diarrhoea.

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### REFERENCES

Abbiw D (1990). Useful Plants of Ghana. Intermediate Technology Publications and the Royal Botanical Gardens, Kew.

Abo KA, Fred-Jayesimi AEA (2008). Ethnobotanical Studies of Medicinal Plants Used in the Management of Diabetes Mellitus in Southern Nigeria. J. Ethnopharmacol. 115: 67-71.

Al-Rehaily AJ, El-Tahir KEH, Mossa JS, Rafatullah S {2001). Pharmacological studies of various extract from hexane extract of Ticlea nobilis in rodents. Nat. Prod. Sci. 7: 76-82.

Burkill HM (1995). The Useful Plants of West Tropical Africa. Royal Botanic Gardens. Kew. 3: 245-51.

Camilleri M (2004). Chronic diarrhoea: A review of pathophysiology and management for the clinical gastroenterologist.Clin. Gastroenterol. Hepatol. 2: 198.

Capasso R, Borrelli F, Cascio MG, Aviello G, Huben K, Zjawiony JK, Marini P, Romano B, Dimarzo V, Capasso F, Izzo AA (2008). Inhibitory effects of salvinorin A, from Salva divinorum on ileiti-induced hypermotility: Cross- talk between kappa-opioid and cannabinoid CB (1) receptors. Br J. Pharmacol. 18: 622-08.

Cobett JR, Wight K, Baille AC (1984). The Biochemical mode of action of Pesticides. 2nd Ed. Academic Press, London and New York.

Gaginella TS, Steward JJ, Oslo WA, Bass P (1975). Actions of ricinoleic acid and structurally related fatty acid on the gastrointestinal tract II. Effect on water and electrolyte absorption in vitro, J. Pharmacol. Exp. Ther. 195: 355-361.

Hall JB (1997). Parkia biglobosa, A Monograph, University of Wales, Bangor.

Hugo WB, Russel AD (1992). Pharmaceutical Microbiology, 5th ed., Blackwell Scientific Pub., New York. P. 119-120.

Longanga-Otshudi A, Vercruysse A, Foriers A (2000). Contribution to the Ethnobotanical, Phytochemical and Pharmacological Studies of Traditionally used Medicinal Plants in the Treatment of Dysentery and Diarrhea in Lomela area, Democratic Republic of Congo (DRC), J. Ethnopharmacol. 71(3): 411-423.

Lorke D (1983). A new approach to Practical Acute toxicity testing, Arch. Toxicol. 54: 275-287.

Odebiyi A, Sofowora AE (1978). Phytochemical Screening of Nigerian Medicinal Plants - Part III. Iloydia 41: 243-246.

Pierce NF, Carpenter CC J, Elliot HZ, Greenough WB (1971). Effect of prostaglandins, theophylline and cholera exotoxin upon transmucosal water and electrolyte movement in canine jejunum, Gastroenterol. 60: 22- 32.

NIH (1978). Guide for the care and use of Laboratory Animals (Revised). NIH Publication No. 82-83.

NIPRD-Standard operation procedures for Pharmacology and Toxicology (unpublished).
Robert A, Nezamis JE, Lancaster C, Hanchar AJ, Klepper MS (1976). Enteropooling assay; a test for diarrhoea produced by prostaglandins, Prostaglandins 11: 809-828.
Salawu OA, Tijani AY, Obidike IC, Chindo BA (2007). Evaluation of the anti-diarrhoeal properties of methanolic root extract of Pilostigma reticulatum in rats. J. Phytomedicine and Therapeutics 12: 44-50.
Shao M (2002). Parkia biglobosa: Changes In Resource Allocation in Kandiga, Ghana", Submitted in partial fulfillment of the requirements for the degree of Master of Science in Forestry Michigan Technological University.
Syder JD, Merson MH (1982). The magnitude of the global problems of acute diarrhoeal disease; A review of active surveillance data. Bulletin of the World Health Organisation 60: 605-613.
Trease GE, Evans WC (1983). Pharmacognosy, Baillere Tindall, London 12th ed. 387: 475-476.
WHO (2006). Country Health System Fact Sheet.
Yu LL, Liao JF, Chen CF (2000). Anti-diarrheal effect of water extract of Evodiae fructus in mice, J. Ethnopharmacol. 73(1-2): 39-45.