



Evaluation of Anti-diarrhoeal Activity of Coconut Water on Castor Oil Induced-Diarrhoea in Adult Wistar Rats

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Authors' contributions

This work was carried out in collaboration between all authors. Author GDE designed the study, wrote the protocol, and wrote the first draft of the manuscript. Authors EBE and AOI managed the literature searches, analyses of the study and performed the spectroscopy analysis and author GDE managed the experimental process and authors EBE and AOI identified the species of plant. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/BJMMR/2016/28378

Editor(s):

(1) Alex Xiucheng Fan, Department of Biochemistry and Molecular Biology, University of Florida, USA.

Reviewers:

(1) I. U. Muhammad, Bayero University Kano, Nigeria.

(2) T. Pullaiah, Sri Krishnadevaraya University, India.

Complete Peer review History: <http://www.sciencedomain.org/review-history/16511>

Original Research Article

Received 16th July 2016
Accepted 9th September 2016
Published 11th October 2016

ABSTRACT

This study was designed to evaluate the anti-diarrhoeal activity of coconut water on castor oil induced-diarrhoea in adult albino Wistar rats. Sixty five adult male Albino Wistar rats weighing 150 – 250 g were used in this study. The study was carried out in three experiments (A, B and C). Experiment A was the castor oil induced-diarrhoea test consisting of 25 animals randomly divided into 5 test groups. Experiment B was the gastrointestinal test consisting of 20 animals randomly divided into 4 test groups and experiment C was the castor oil induced enteropooling test consisting of 20 animals randomly divided into 5 test groups. Animals in experiment A were treated with water (control). 2 ml of castor oil/kg bd.wt, 30 ml of coconut water/kg bd.wt + 2 ml of castor oil/kg bd.wt orally, 40 ml of coconut water/kg bd.wt + 2 ml of castor oil/kg bd.wt and 4 mg of loperamide/kg bd.wt + 2 ml of castor oil/kg bd.wt in group 2, 3 4 and 5 respectively. Light

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microscopic observations were made on formalin-fixed, 5 µm thick paraffin sections of stomach stained with haematoxylin stains. Animals in experiment B were tested for anti-motility effect of coconut water using charcoal meal and were treated with water (control), 30 ml of coconut water/kg bd.wt, 40 ml of coconut water/kg bd.wt and 4 mg of loperamide/kg bd.wt in group 2, 3 and 4 respectively. Animals in experiment C were tested for intestinal fluid accumulation and were treated with 2 ml/kg bd.wt of castor oil (control), 30 ml of coconut water/kg bd.wt + 2 ml of castor oil/kg bd.wt, 40 ml of coconut water/kg bd.wt + 2 ml of castor oil/kg bd.wt and 4 mg of loperamide/kg bd.wt + 2 ml of castor oil/kg bd.wt in group 2, 3 and 4 respectively. Result from this study revealed that coconut water given orally to rats in all the experimental test groups significantly reduced copious diarrhoea, inhibit charcoal transit in the small intestine, inhibit intestinal fluid accumulation which was comparable to loperamide, the standard anti-diarrhoeal drug. Also protects the gastric mucosa and other components of the stomach from the irritating stimuli produced by castor oil induced-diarrhoea in a dose dependent manner. Based on these results, we concluded that coconut water may be useful in the treatment of diarrhoea especially in areas where conventional anti-diarrhoeal drugs are not readily accessible.

Keywords: Enteropooling; diarrhoea; castor oil; loperamide; coconut water.

1. INTRODUCTION

Coconut water have been reported to have a very low amount of carbohydrates, calories and sodium. Coconut water, as a tropical fruit juice, is highly valued and consumed in tropical areas since it is tasty and has desirable nutritional and therapeutic properties. The total world coconut cultivation area was estimated in 1996 at 11 million hectares, and around 93 percent (%) was found in the Asian and Pacific regions [1]. Indonesia, the Philippines, and India are the largest producers of coconut in the world. Coconut (*Cocos nucifera* Linn.) fruit is filled with the sweet clear liquid “coconut water” when the coconut is about five to six months old. Coconut water has been called the “fluid of life” due to its medicinal benefits such as oral rehydration, gastroenteritis and cholera [2,3].

It is high in electrolyte content and has been reported as an isotonic beverage due to its balanced electrolytes like sodium and potassium that help restore losses of electrolytes through skin and urinary pathways. Coconut water was claimed as a natural contender in the sports drink market with its delicate aroma, taste and nutritional characteristics together with the functional characteristics required in a sports drink [4].

The constituents of coconut water are water 94% (w/v), sugars such as glucose, fructose and sucrose around 5% (w/v), proteins around 0.02% (w/v) and lipids only about 0.01% (w/v). It is rich in minerals such as potassium, calcium, magnesium and manganese and low in sodium. Most coconut water is consumed fresh in tropical coastal areas due to its short shelf-life. Once

exposed to air, it loses most of its sensory and nutritional characteristics and deteriorates. Commercially, coconut juice production is carried out mostly in Indonesia, the Philippines, and Thailand using ultra high temperature (UHT) sterilization while some of coconut water's nutrients and its delicate flavour are lost during this thermal processing which limits the product's marketability [4].



Fig. 1. Coconut fruit

There have been cases where coconut water is being used as intravenous hydration fluid in some developing countries where medical saline was unavailable [5]. The stomach is the food storage organ in the human gastrointestinal system. It is located in the left upper quadrant of the abdomen below the diaphragm. The organ somewhat resembles a J-shape and has two openings at the either end. At the top it is connected to the oesophagus and to the bottom it opens to the duodenum. Diarrhoea indicates abnormally increased frequency of bowel movements and looseness of stools [6]. Healthy individuals experience two - three bowel movements per day.

More than three bowel movements can be called diarrhoea. Liquid stools indicate dysfunction of the digestive system [7]. Diarrhoea lasting from a few days up to a week is called acute diarrhoea and that which lasts for more than three weeks is called chronic diarrhoea [8]. Dysfunction of the digestive system leads to increased water in the stools which results in looseness of stool [9]. Coconut water contains some antioxidants which are molecule that inhibits the oxidation of other molecules. Antioxidants are widely used in dietary supplements and have been investigated for the prevention of diseases such as cancer, coronary heart disease and even altitude sickness [10]. Zymogenic cells (Zcs), acid-producing parietal cells and mucus secreting pit cells are the principal epithelial lineages in the stomach of adult mice and humans [11]. Each lineage is derived from the multipotent gastric stem cell and undergoes perpetual renewal within discrete mucosal invaginations (gastric units).

The human gastric epithelium is renewed continuously throughout life with an estimated five hundred thousand (500,000) cells shed per minute in adults [12]. Dysregulated renewal results in a number of diseases states including diarrhoea, gastric adenocarcinoma, the second most common cause of cancer-related deaths in the world [13]. The proximal third of the rat stomach is lined with a squamous epithelium while the distal two-thirds is lined with a glandular epithelium. The glandular epithelium contains thousands of tubular mucosal invaginations known as gastric units. In the central region of the stomach, each unit has a steady state of population of approximately two hundred (200) epithelial cells representing three principal lineages (pit, parietal and zymogenic) and two minor lineages (enteroendocrine and caveolated) [14] All of the lineages are derived from multipotent stem cells located in the midportion of each unit [15]. Continuous regeneration of digestive enzymes in zymogen-secreting chief cells is a normal aspect of the stomach function that is disrupted in precancerous lesions (example, Metaplasia, chronic atrophy). The cellular and genetic pathways that underlie zymogenic cell differentiation are poorly understood [16]. In mammalian gastrointestinal tract, the cell fate decisions that specify the development of multiple, diverse lineages are governed in large part by interactions of stem and early lineage progenitor cells with their microenvironment or niche [17]. The secretions of the mammalian

stomach are produced by cells present in invaginations of the epithelium. The stem cells are undifferentiated granule free cells which undergo division to maintain their own number and produce several differently oriented progenitors. The phenotype of prezymogenic cells gradually changes from mucous to serous. These cells eventually lose the ability to produce mucous and thus become the typical zymogenic cells that populate the base region [15].

2. MATERIALS AND METHODS

2.1 Materials

The following materials were used for the experiment: sixty five adult albino Wistar rats, clean cages, feeding bottles, weighing balance, dissecting board, forceps, surgical blade, paraffin wax, embedding mold, oven, wooden block, microtome and blade, camel hair brush, fuming cupboard, castor oil, water bath and clean slides. Others were microscope, staining rack, cover slips, egg albumin, glass funnel, bunsen burner, pipette, thermometer, stirrer, Pyrex glass, loperamide and refrigerator.

2.2 Reagents

The reagents used were 10% neutral buffered formalin, alcohol of different grades, xylene, 1% acid alcohol, 0.5% dilute ammonia, Eosin and Haematoxylin stain, hydrochloric acid, mounting medium, activated charcoal suspended in 5% gum acacia, coconut water and castor oil.

2.3 Drug Preparation and Calculation of Coconut Water Concentration

Loperamide was purchased from Tibest pharmacy located at 7 Calabar- Itu road in Uyo, Akwa Ibom State, Nigeria. 4 mg/kg body weight of loperamide was dissolved in 100 ml of water. This yielded a stock solution of 0.04 ml/kg. 2 ml of castor oil was administered orally to the rats. 30 ml/kg and 40 ml/kg bd.wt of coconut water were the estimated volumes selected. Based on 30 ml/kg volume selection, required dose volume for a 150 g rat was calculated as follows:

$150 \text{ g}/1000 \text{ g} \times 30 \text{ ml} = 4.5 \text{ ml/kg}$. Similarly, based on 40 ml/kg volume selection, required dose volume for a 250 g rat was calculated as follows:

$250 \text{ g}/1000 \text{ g} \times 40 \text{ ml} = 10 \text{ ml/kg}$. Each rat received coconut water based on calculated

doses with respect to body weights as shown above.

2.4 Coconut Water, Phytochemical Screening and Elemental Analysis (Method of Clarke [17] and Odebiyi and Sofowara [18])

Coconut water was obtained from fresh Coconut from a local village in Itu local government area of Akwa Ibom State. The water was extracted and preserved in the refrigerator. Phytochemical and elemental analysis of coconut water was done according to the methods of Clarke [17] and Odebiyi and Sofowara [18]. Test for electrolytes, carbohydrates and other sugars, dietary fibre and vitamins were carried out.

2.5 Animal Care

Sixty five adult male albino rats (weighing 150-250 g) of wistar strain were used for this research. The animals were housed in standardized environmental conditions, maintained at 12 hr light and dark cycles and kept at room temperature of 27- 30°C in clean cages in a well ventilated room at the animal house of the Faculty of Basic Medical Sciences, University of Uyo. They were fed with standard rodent diet (Growers feed) and clean drinking water. All rats were handled according to standard guidelines for the care and use of laboratory animals (Association for Assessment and Accreditation of Laboratory Animal Care, 2009). The animals were divided into three test groups namely castor oil induced-diarrhoea test group (A), Gastrointestinal test group (B) and castor oil induced-intestinal fluid accumulation test group (C).

2.6 Castor Oil – Induced Diarrhoea Test (Test Group A)

A modification of the method according to Awouters [19] was adopted. Rats were fasted for 24 h but allowed free access to water and were randomly divided into five groups of five animals each as follows:

Group A₁: was the control and were administered drinking water alone.

Group A₂: rats were administered with water.

Group A₃: rats were administered with 30 ml/kg body weight of coconut water.

Group A₄: rats were administered with 40 ml/kg body weight of coconut water.

Group A₅: was the positive control and were administered with 4 mg/kg body weight of loperamide orally.

One hour after treatment, each rat from groups A₂-A₅ were administered with 2 ml/kg body weight of castor oil orally and were kept under a glass funnel, the floor of which was lined by a blotting paper and observed for 4 hr. After each hour the blotting paper with stools was changed and frequency of defecation and total weight of stools were noted. Immediately the rats were anaesthetised with chloroform and the stomach was rapidly dissected out and preserved in 10% buffered formalin.

2.7 Gastrointestinal Test (Test Group B)

The effect of coconut water on intestinal propulsion was determined using the charcoal method employed by Ruwart et al. [20]. The rats were fasted for 24 h but allowed free access to water and were randomly divided into four groups of five animals each as follows:

Group B₁: was the control

Group B₂: rats were administered with 30 ml/kg body weight of coconut water orally

Group B₃: rats were administered with 40 ml/kg body weight of coconut water orally

Group B₄: rats were administered with 4 mg/kg body weight of loperamide orally.

Thirty minutes later, each rat was administered with 0.2 ml of standard charcoal meal (10% activated charcoal suspended in 5% gum acacia) orally. The rats were anaesthetised using chloroform 30 min after administration of charcoal meal. The small intestine of each rat was carefully identified and the distance travelled by the charcoal meal from the pylorus to the ileocecal junction (length of the small intestine) was measured and expressed as percentage using this relation.

$$Ip\% = (DT/TL)100$$

Where

IP = Intestinal propulsion.

DT= Distance travelled by the charcoal meal.

TL = Total length of the small intestine.

Inhibition of propulsion was calculated relative to the control as follows

$$\text{Inhibition of } p\% = 100[1-(a/b)]$$

Where

a = Intestinal propulsion of treated animals.

b = Intestinal propulsion of control animals.

2.8 Castor Oil– Induced Intestinal Fluid Accumulation (Enteropooling) (Test Group C)

This was determined according to the method employed by Robert et al. [21] modified by Dicarolo et al. [22]. 20 rats were used in test group C. The rats were fasted for 24 hr but allowed free access to water. They were randomly allotted in 4 groups with 5 animals each shown as follows:

Group C₁: was the control

Group C₂: rats were administered with 30 ml/kg body weight of coconut water

Group C₃: rats were administered with 40 ml/kg body weight of coconut water

Group C₄: rats were administered with 4 mg/kg body weight of Loperamide as positive control

After 1 hr, each rat was administered with 2 ml of castor oil orally respectively, 30 minutes later, the rats were anaesthetised with chloroform and the small intestine was ligated at both the pyloric and the ileocecal junctions and the intestinal contents were expelled into a graduated measuring cylinder and the volume of the contents was recorded.

2.9 Tissue Processing (Routine Method)

Tissue processing was carried out in the castor oil –induced diarrhoea test group. A unique identification number was assigned to the tissue sample accessioned in the laboratory. Tissues were fixed in 10% buffered formalin and passed through graded series of alcohol, cleared in xylene, infiltrated, embedded and then sectioned.

2.10 Staining Technique

Haematoxylin staining method according to Llewelly [23] was employed.

2.11 Data Analysis

Data were analysed using analysis of variance (ANOVA) followed by post-hoc test. All data were expressed as means ± standard error of

mean. Data were analysed using window SPSS (version 15.0).

3. RESULTS

3.1 Phytochemical Constituents of Coconut Water

Preliminary phytochemical analysis of coconut water showed the presence of the following constituents; potassium, sodium, magnesium, manganese, vitamins (B₁, B₂, B₃, B₅ and B₆), protein, dietary fibre, fat, calcium, iron, magnesium, folate, sugars, carbohydrates, phosphorus, potassium, zinc and water.

3.2 Effect of Coconut Water on Castor Oil – Induced Diarrhea

Four hours after castor oil administration, all the rats in the group administered with castor oil alone produced copious diarrhoea. Pretreatment with coconut water elicited significant ($p < 0.05$) and dose related delay in the onset of diarrhoea and reduction in the frequency of defecation (Table 1). The activity was similar to that of Loperamide (4 mg/kg), the standard anti-diarrhoeal agent.

3.3 Effect of Coconut Water on Gastrointestinal Propulsion

Coconut water elicited significant ($p < 0.05$) dose related reduction in charcoal meal transit. 30 ml/kg and 40 ml/kg body weight of coconut water reduced the distance covered by the charcoal meal in the gastrointestinal tract of rats by 40.73% and 34.70% respectively. These values were higher than 33.7% produced by loperamide (4 mg/kg), that means a similar reduction in the gastrointestinal transit of charcoal meal in rats was achieved with the standard drug, loperamide (Table 2).

3.4 Effect of Coconut Water on Castor Oil Induced Enteropooling

Pretreatment with coconut water (30- 40 ml/kg) dose dependently inhibited (45.95-67.57%) castor oil-induced fluid accumulation (enteropooling) when compared to the control (Table 3). The standard drug Loperamide also inhibited enteropooling and this inhibition (71.89%) is significantly ($P < 0.05$) greater than that of the highest dose of coconut water (40 ml/kg, 67.57%).

3.5 Histological Observation

The Haematoxylin and Eosin (H&E) sections of the stomach of control rats given water showed the presence of well defined/distinct mucosa with epithelial lining (EL), the gastric pit (GP), gastric glands (GGS), goblet cells (GC), muscularis mucosa (MM), submucosa (SM) and its component while the H&E sections of the stomach in the treatment group administered with 2 ml of castor oil alone showed surface epithelial erosion (SEE), degeneration of connective tissue component (DCT) in the submucosa. The Haematoxylin and Eosin sections of the stomach of animals administered with 30 ml/kg b.w of coconut water with 2 ml of castor oil showed mild epithelial erosion (MEE).

4. DISCUSSION

Over the years medicinal plants have been shown to contain substances of therapeutic significance [24]. One of such plant is the coconut plant, coconut fruit is an important medicinal fruit as it is a well known source of different phytochemicals, it is distributed throughout most of the part of the world including Africa. Findings through phytochemical screening shows the presence of proteins, dietary fibre, carbohydrates, fat, antioxidants and some electrolytes in coconut water. Administration of antioxidants ameliorate castor oil induced-diarrhoea in rats, coconut water possesses a broad spectrum of biological activities and the water have been reported to have large quantity of antioxidant compounds [25] and it is speculated that the gastroprotective potentials exerted by coconut water could be attributed to its antioxidant property [26]. The

presence of potassium, sodium, magnesium, manganese in coconut water could be attributed to the anti-diarrhoeal activity of coconut water.

This study was designed to evaluate the anti-diarrhoeal potentials of varied doses (30 ml/kg bd.wt and 40 ml/kg bd.wt) of coconut water on castor oil induced-diarrhoea in adult Wistar rats compared to the standard anti-diarrhoeal drug, loperamide. The stomach receives, mixes, stores and digest ingested food products and secretes different hormones that regulate digestive functions [27]. Histological examination reveal that this organ has well organised cytoarchitectural plans which when modified or altered can lead to alterations in the normal chemical, hormonal and mechanical functions of the organ. Cells present in the stomach have the potentials to secrete hydrochloric acid, gastric intrinsic factor, pepsin and hormones [27]. This implies that interference in the normal functioning of these cells can impair digestion, secretion, absorption and hormonal imbalance which can result in hyper- or hypo- activity of the entire organ. Gastric cells exposed to pernicious condition like diarrhoea indicated severe cellular distortions like epithelial cell atrophy [28]. Coconut water have been reported to express anti-inflammatory effects, anti-oxidant effect, antiviral and antibacterial effects [29]. The anti-oxidant activity of coconut water have been attributed to its protein and vitamin C contents and the antiviral/ antibacterial effects of coconut water have been attributed to the presence of saturated fatty acid, once in the human stomach partial digestion of these fatty acids produce substances that can kill certain bacteria and viruses [29].

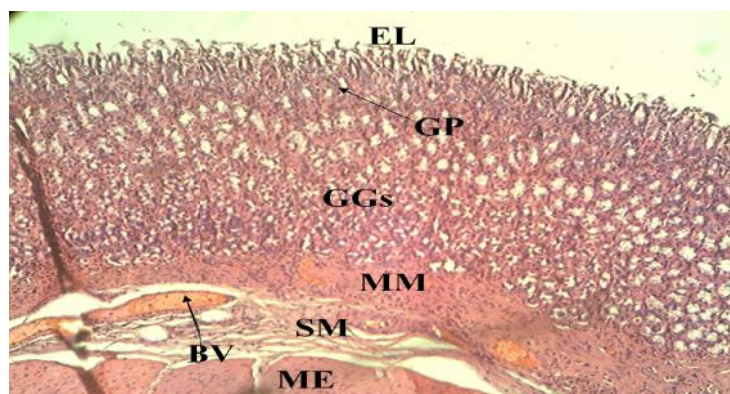


Fig. 2. Photomicrograph of the stomach from rats in control group (A₁) given water showing well defined/distinct mucosa with epithelial lining (EL), gastric glands (GGs), muscularis mucosa (MM), Submucosa (SM) and muscularis externa (ME) (H&E X100)

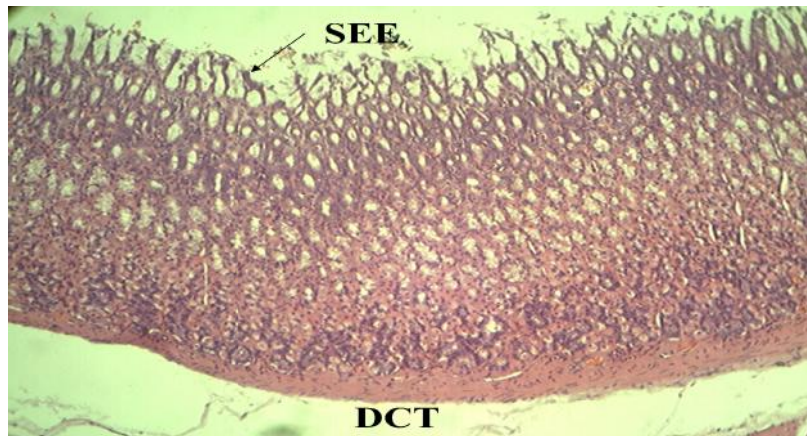


Fig. 3. Photomicrograph of the stomach from group A₂ (castor oil alone) rats showing severe epithelial erosion (SEE) and degeneration of connective tissue components (DCT) in the submucosa (H&E X 100)

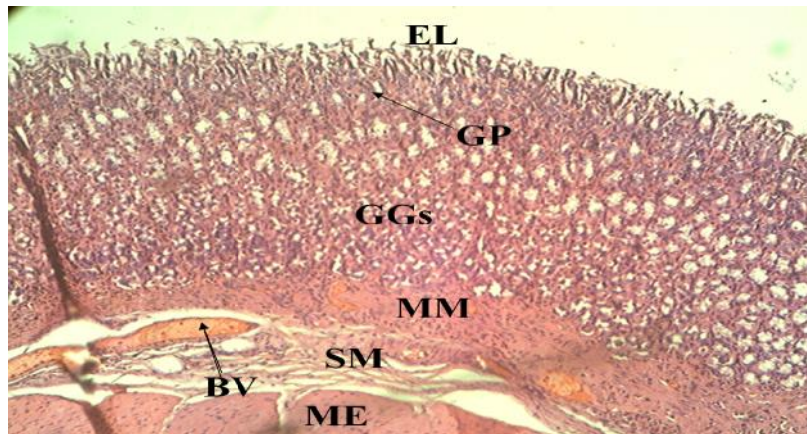


Fig. 4. Photomicrograph of the stomach from rats in control group(A₁) given water showing well defined/distinct mucosa with epithelial lining (EL), gastric glands (GGs), muscularis mucosa (MM), Submucosa (SM) and muscularis externa (ME) (H&E X100)

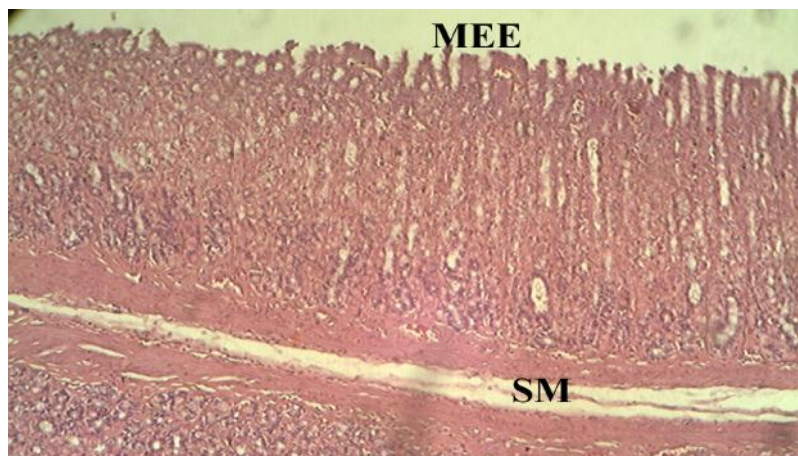


Fig. 5. Photomicrograph of the stomach from rats in group A₃ (30 ml/kg coconut water + 2 ml castor oil) showing mild epithelial erosion (MEE) (H&E X100)

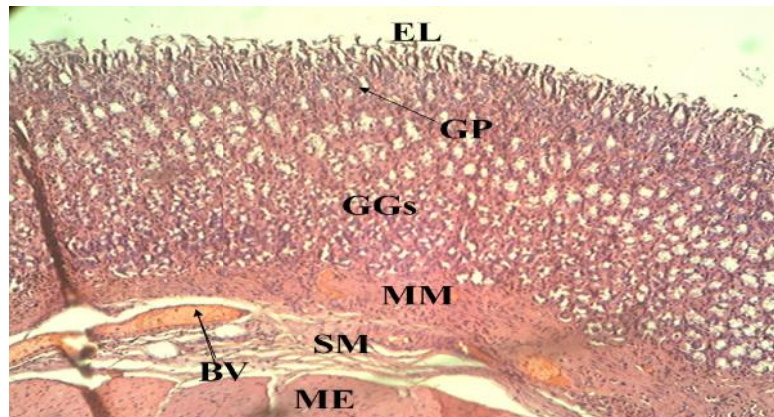


Fig. 6. Photomicrograph of the stomach from rats in control group (A₁) given water showing well defined/distinct mucosa with epithelial lining (EL), gastric glands (GGs), muscularis mucosa (MM), Submucosa (SM) and muscularis externa (ME) (H&E X100)

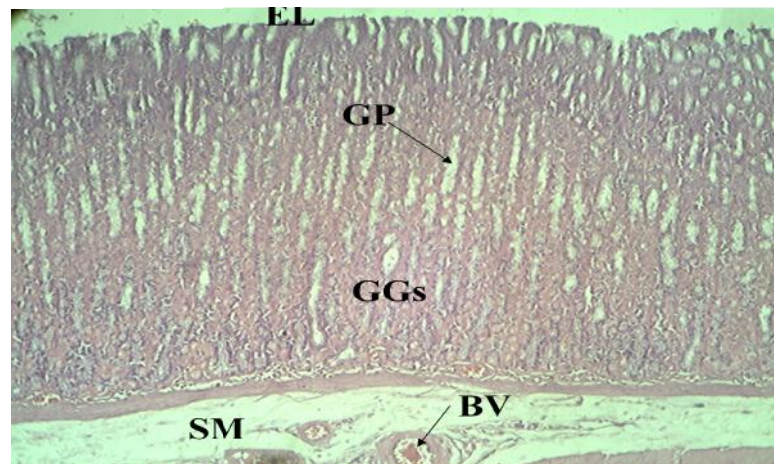


Fig. 7. Photomicrograph of the stomach from rats in group A₄ (40 ml/kg coconut water + 2 ml castor oil) showing improvement in the mucosa with epithelial lining (EL), gastric pit (GP), gastric glands (GGs), submucosa (SM) with component blood vessel (BV) (H&E X100)

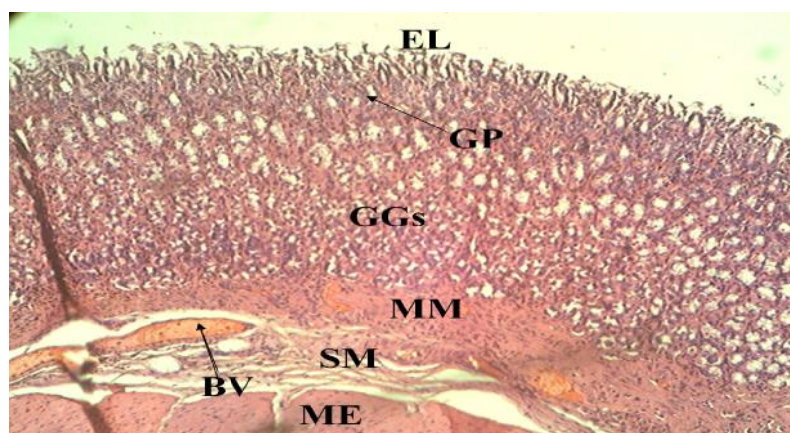


Fig. 8. Photomicrograph of the stomach from rats in control group (A₁) given water showing well defined/distinct mucosa with epithelial lining (EL), gastric glands (GGs), muscularis mucosa (MM), Submucosa (SM) and muscularis externa (ME) (H&E X100)

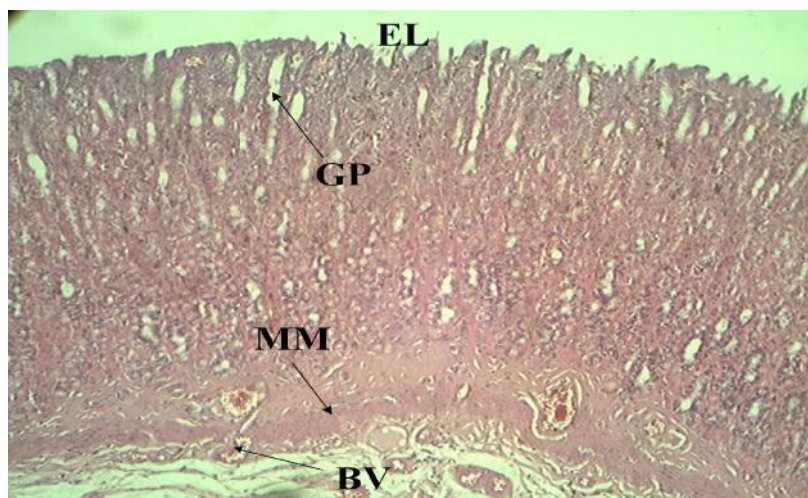


Fig. 9. Photomicrograph of the stomach from rats in group A₅ (4 mg/kg loperamide + 2 ml castor oil) showing improvement in the mucosa with epithelial lining (EL), gastric pit (GP), submucosa (SM) with component blood vessel (BV) (H&E X100)

The results of this study shows that administration of coconut water inhibited castor oil-induced diarrhoea, gastrointestinal motility and intestinal fluid accumulation in rats when compared to the standard anti-diarrhoeal drug, loperamide. Significant increase of plasma electrolytes in rats was also evident after coconut water administration similar to that of loperamide. This implies that coconut water might have anti-diarrhoeal property therefore suggesting its use in ameliorating diarrhoea.

Coconut water significantly reduced the number of faecal boli produced after 4 h. This reduction of copious defaecation is likely due to interference with gastrointestinal peristalsis and motility [30]. The percentage inhibition by the standard drug, loperamide is greater than that of the highest dose of coconut water. Drugs affecting motility, frequency and consistency of diarrhoea also affect secretion [31]. Induction of diarrhoea by castor oil increased peristaltic activity and increased permeability changes in the mucosal membrane to electrolyte and water. There was however absence of diarrhoea in those animals treated with different doses of coconut water and loperamide respectively. This result is in line with similar work done by Ezekwesili et al. [32] who evaluated the anti-diarrhoeal effect of ethanolic extract of psidium guava leaves in the reduction of faecal boli in rats.

Apart from inhibition of castor oil induced-diarrhoea, rats pretreated with coconut water

significantly indicated reduction in the distance travelled by the charcoal meal in the small intestine and percentage of propulsion in a dose dependent manner similar to that of loperamide at a dose of 4 mg/kg body weight. These results agree with similar reports which have established reduction in the gastric motility as being the mechanism by which many anti-diarrhoeal agents act [33]. Several studies have been designed to examine the influence of substances on gastrointestinal motility. Cooke [34,35] reported that gastric emptying was unaffected by ethanol in concentrations of 6 g/100 ml but slowed at higher concentrations. On the other hand, Harichaux [36] who studied gastric emptying by a less sensitive and less accurate radiographic technique observed acceleration of emptying by ethanol in low concentration but slowing at higher concentration. Kamalraj et al. [37] reported that administration of anti-diarrhoeal agent, *Erythrina indica* Lam. Leaf extract decreased propulsion of charcoal meal through the gastrointestinal tract at the oral dose of 500 mg/kg bd.wt similar to loperamide. The result of this study also show that rats pretreated with coconut water expressed significant decrease in intestinal fluid accumulation (enteropooling), the intraluminal fluid accumulation induce by castor oil was blocked by coconut water in a dose dependent manner. Castor oil produces permeability changes in the gastric and intestinal mucosal membranes to water and electrolytes resulting in fluid and watery luminal content that flows through the small

intestine [38]. This is brought about by the irritant effect of ricinoleic acid by the pancreatic lipases which hydrolyse the oil derived from the seeds of *Ricinus communis*. Coconut water inhibited the castor oil-induced intestinal fluid accumulation without affecting the weight of the intestinal content. This result was in conformity to a study by Mbagwu and Adeyimi [39] who observed that certain anti-diarrhoeal agents like *Mezoneuron benthamianum* Bail (Caesalpiniaceae) at varied doses of 400-1600 mg/kg bd. wt dose dependently inhibited castor oil-induced fluid accumulation when compared to morphine, another standard anti-diarrhoeal drug. Similar reports were also given by Besra et al. [40], Duraid [41] and Nwafor et al. [42] who noted that certain anti-diarrhoeal agents greatly decrease enteropooling without negatively affecting intestinal contents.

Histological findings from this study reveals that that sections of stomach of rats administered with 2 ml/kg bd. wt of castor oil alone when compared with the stomach of rats in the control group showed severe epithelial erosion and degeneration of connective tissue in the submucosa, this is in line with the works by Myers and McGavin [43] who investigated cellular and tissue responses in injury, glandular stomach necrosis in a female B6C3FI mouse, loss of glandular epithelial cells and scattered pyknotic debris within the mucosa, thrombosed vessel in the submucosa resulting in an area of necrosis was investigated.

Histological findings from the sections of stomach of rats administered with 30 ml/kg bd.wt of coconut water with 2 ml/kg bd. wt of castor oil showed mild epithelial erosion, improvement of connective tissue component in the submucosa due to reversible tissue injury and muscle myopathy. Muscle hypertrophy involves an increase in the size of muscle through a growth in the component cells due to external influence [43]. The total force of contraction of a muscle is directly proportional to its physiological cross sectional area [44]. Causes of cell injury range from gross mechanical, external causes to mild endogenous causes as genetic lack of enzymes etc. Virtually all forms of tissue injuries start with molecular or structural alterations in cells. Under normal conditions, the cells are in homeostatic steady state, cells react to adverse influence by sustaining reversible injury, for example excessive work stress causes an increase in muscle mass that reflects the

increase in size of the individual muscle fibre results in higher level of metabolic activity [44].

Reversible cell injury denotes pathologic changes that can be reversed when the stimulus is removed and the cellular injury has been mild [45]. Therefore the administration of 40 ml/kg bd.wt of coconut water with 2 ml/kg bd. wt of castor oil protected the components of the stomach against diarrhoea with well defined and distinct mucosa similar to the group administered with 4 mg/kg bd.wt of loperamide, the standard anti-diarrhoeal drug. It is established that damage in gastric mucosa, disturbs in gastric secretion, alterations in permeability, gastric mucus depletion and free radical production are reversed after administration of anti-diarrhoeal agents [46]. This implies that coconut water might increase the absorption of electrolytes and enhance mucus production for gastroprotective potentials.

Coconut water produces a dose dependent and significant ($P < 0.05$) protection against castor oil induced diarrhoea from mechanisms such as direct toxin action, reduction of the secretion of bicarbonate and depletion of gastric wall mucus [47]. Anti-diarrhoeal agents are thought to increase endogenous glutathione and prostaglandin levels and decreases the release of histamine, increases the influx of calcium ion and generation of free radical [48]. Thus, the action of coconut water in this model suggests cytoprotective action possibly mediated through enhancement of mucosal defensive factors such as protection from free radicals.

Coconut water have been found to contain saturated acids, once in the human stomach partial digestion of these acids produce substances which are capable of killing bacteria and viruses [29]. In particular, the saturated fatty acids lauric, capric and caprylic, all found in coconut oil, have been identified in studies as having some anti-infective action. Some polyunsaturated fatty acids have been identified as having the same action. It seems that the saturated fatty acid digestion products destroy the wall of the virus or bacteria, which means that there is little chance of resistant types of bacteria developing (a concern with most antibiotics). These fatty acids also seem to be able to destroy *Giardia lamblia*, a gut parasite responsible for one type of severe diarrhea [29]. It may be suggested that coconut water which has anti-motility and cytoprotective effect possess anti-diarrhoeal potentials.

Table 1. Effect of coconut water on castor oil induced-diarrhoea in rats

Group	Treatment	Mean faecal matter	% reduction
Control	Water	5.00 ± 0.10	-
2	Water & castor oil	12.00 ± 0.60 ⁱ	-
3	Cw (30 ml/kg)	7.00 ± 0.20 ^{i, ii}	41.67
4	Cw (40 ml/kg)	6.50 ± 0.30 ^{i, ii, iii}	45.83
5	Loperamide (4 mg/kg)	6.00 ± 0.30 ^{i, ii, iii}	50.00

Cw - Coconut Water, n = 5, ^{i, ii, iii}p < 0.05 compared to control. I.e values bearing different superscript are significantly different from the control

Table 2. Effect of coconut water on gastrointestinal motility in rats

Group	Treatment	Length of intestine (cm)	Distance (cm)	Propulsion	% inhibition
Control	Water	45.60 ± 0.20	25.62 ± 4.10	56.20 ± 1.60	-
2	Cw (30 ml/kg)	44.68 ± 0.80	18.20 ± 1.20	40.73 ± 1.50 ⁱ	28.90 ± 0.40
3	Cw (40 ml/kg)	46.10 ± 2.60	16.00 ± 1.00	34.70 ± 0.40 ^{i, ii}	36.77 ± 0.70
4	Lp (4 mg/kg)	43.20 ± 0.30	14.54 ± 1.20	33.70 ± 0.80 ^{i, ii, iii}	43.24 ± 0.40

Cw - Coconut Water, LP -Loperamide, n = 4, ^{i, ii, iii}p < 0.05 compared to control I.e values bearing different superscript are significantly different from the control

Table 3. Effect of coconut water on castor oil- induced enteropooling

Group	Treatment	Intestinal fluid	% inhibition
Control	Water	1.85 ± 1.40	--
2	Cw (30 ml/kg)	1.00 ± 0.40	45.95
3	Cw (40 ml/kg)	0.60 ± 0.30	67.57 ⁱⁱ
4	Lp (4 mg/kg)	0.52 ± 0.20	71.89 ^{ii, iii}

Cw - Coconut Water, LP -Loperamide, n = 4, ^{i, ii, iii}p < 0.05 compared to control. I.e values bearing different superscript are significantly different from the control

5. CONCLUSION

This study has demonstrated that coconut water possess anti-diarrhoeal, anti-motility and gastroprotective potentials. These findings provide a rationale for the use of coconut water in the treatment of diarrhoea in traditional medicine practice of south-south Nigeria. Coconut water contains electrolytes that will replenish the lost electrolytes during diarrhoea.

CONSENT

It is not applicable.

ETHICAL APPROVAL

All authors hereby declare that the experiment have been examined and approved by the ethics committee of the faculty of Basic Medical Sciences, University of Uyo and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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