

Levels of Castor Oil-Induced Diarrhoea in Rats Treated with Leaf Extract of *Corchorus olitorius* Linn and Aerial Part Extract of *Scoporia dulcis* Linn



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ARTICLE INFO

Received: 30 April 2019
Revised: 23 May 2019
Accepted: 21 Jun 2019
Available online: 22 Jun 2019

DOI: 10.33945/SAMI/AJCA.2020.1.1

KEYWORDS

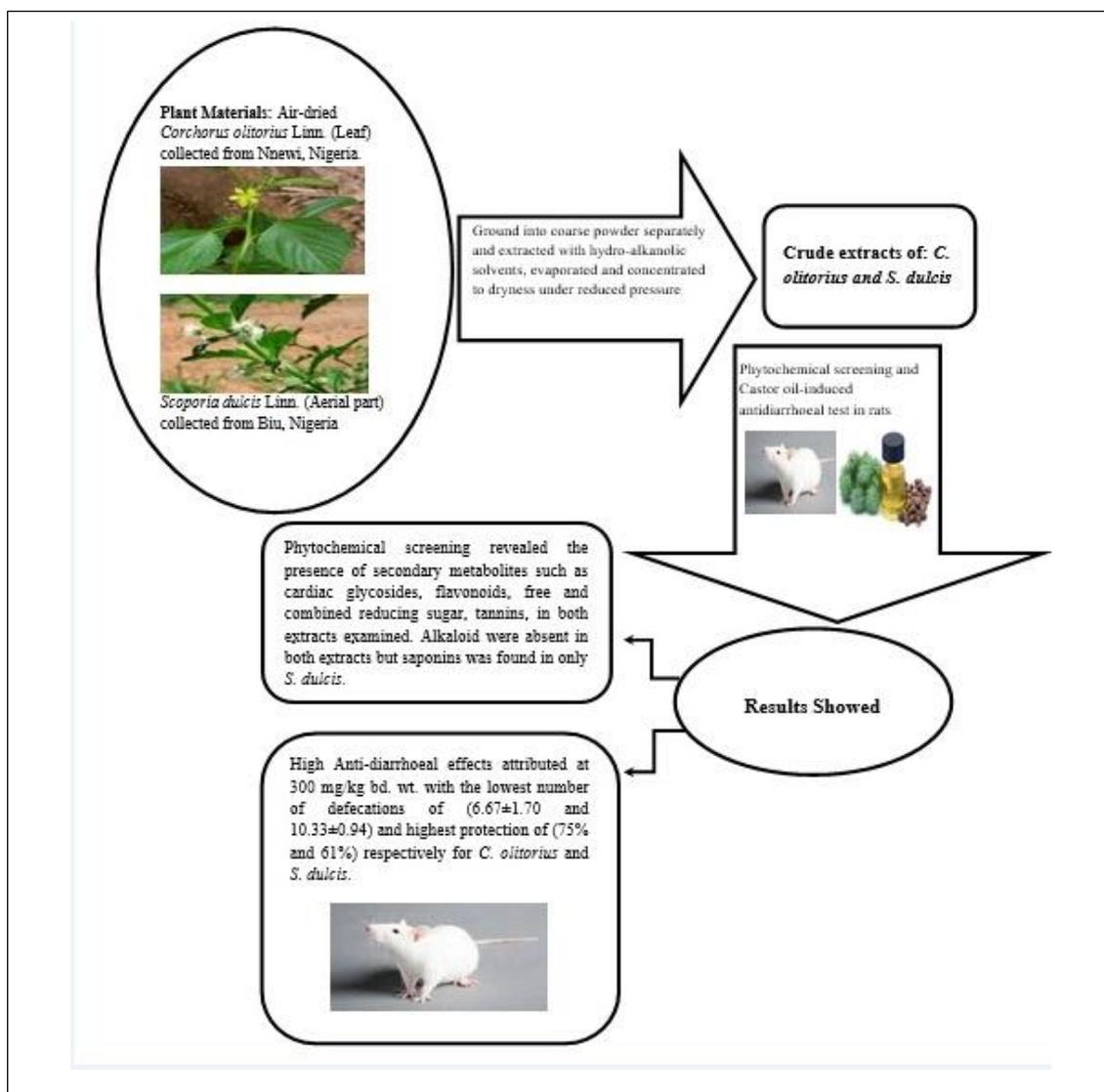
Castor oil
Corchorus olitorius
Diarrhoea
Extract
Rats
Scoporia dulcis

ABSTRACT

This study reports the phytochemical constituents and anti-diarrhoeal potentials of leaf extracts of *Corchorus olitorius* Linn. (Malvaceae) and that of aerial part of *Scoporia dulcis* Linn. (Lantaginaceae) in rats. Samples were collected in December, 2016 from Nnewi-ichi, Nnewi, Anambra State and Mandara-Abdu, Biu-Borno State, Nigeria, respectively. The phytochemical screening of the *C. olitorius* and the aerial part of *S. dulcis* extracts were conducted using standard methods; the results revealed the presence of cardiac glycosides, flavonoids, free and combined reducing sugar and tannins. Saponins was only found in *S. dulcis*. The diarrhoea was induced in rats by castor oil-induced method. The rats were grouped into 3 rats of 5 groups for each extract; groups I and II served as negative and positive control, while groups III, IV and V as treatment groups. The results showed a non-dose dependent but significant ($p < 0.05$) effects with the lowest mean number of defecations at 6.67 ± 1.70 and 10.33 ± 0.94 ; and highest protections of 75.00% and 61.27% respectively at the same dose of 300 mg/kg bd. wt. for *C. olitorius* and *S. dulcis*. Comparatively, *C. olitorius* showed significantly ($p < 0.05$) low severity of diarrhoea relative to *S. dulcis*. Thus, it showed reduction in the faecal output and protection of the rats from diarrhoea induced by castor oil. These observed results could explain their use as anti-diarrhoeal agents in African Traditional Medicine especially in Nigeria where the plants are used.

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GRAPHICAL ABSTRACT



Introduction

Diarrhoea is usually considered as a result of altered motility and fluid defecation within the intestinal tract. Diarrhoea is one of the main causes of infant mortality in developing countries causing about 5 to 8 million deaths a year; mainly among children under the age of five [1]. Diarrhoea involves increased gastrointestinal mobility and secretion and decreased absorption of water and electrolytes [1]. The World Health Organisation (W.H.O) defines traditional medicine as the sum total of the knowledge,

skill and practices based on the theories, beliefs and experiences indigenous to different cultures, whether explicable or not, used in the maintenance of health as well as in the prevention, diagnosis, improvement or treatment of physical and mental illness [2]. Plants make many chemical compounds for biological functions including defence against insects, fungi and herbivorous mammals. A medicinal plant is a plant that has similar properties as conventional pharmaceutical drugs; they have been used throughout

history to either cure or lessen symptoms from illness (es). Typically, pharmaceutical drugs are modelled after compounds found in medicinal plants [3]. At least 12,000 such compounds have been isolated; this is estimated to be less than 10% of the total [4]. These chemicals work on the human body in exactly the same way as pharmaceutical drugs; so herbal medicines can be beneficial and have harmful side effects like conventional drugs. Furthermore, plants extracts may have multiple side effects [5]. Herbs are widely used to treat disease in non-industrialized societies, not least because they are far cheaper than modern medicines [6].

Therefore, it is important to stress the relevance of traditional medicine to the majority of Nigerians. Most Nigerians, especially those living in rural communities do not have access to orthodox medicine and therefore, it is estimated that about 75% of the populace still prefer to solve their health problems by consulting traditional healers. More so, in view of our continuous interest in search of crude drug plant of medicinal values against infectious disease; this research work was designed to evaluate two medicinal plants that are used locally in Nigeria as a remedy against diarrhoea.

Corchorus olitorius (Malvaceae) is an edible leafy vegetable which is a member of genus *Corchorus* classified under the subfamily grewioideae of the family Malvaceae. It is found in tropical and subtropical areas from Asia to Africa appreciated as food and for its strong fibre. Jute leaf, Jute mallow, Saluyot, Jews mallow, Egyptian spinach, bush okra, West African sorrel, krinkrin are some common names of Jute; and was once known as the golden fibre of Bangladesh since it was the most important cash crop for the country [7, 8]. Jute leaves are very nutritious, rich in minerals and vitamins and dietary fibres. It is also used as herbal medicine to control or prevent dysentery, worm and constipation [9].

Scoporia dulcis (Scrophulariaceae), commonly known as sweet broom-weed, is a perennial herb widely distributed in tropical and subtropical region. In these regions fresh or dried *S. dulcis* plants have been traditionally used as remedies for stomach troubles, hypertension, diabetes, and bronchitis as analgesic and antipyretic agents [10]. The plant is seen as an ant bilious, antibiotic, antidote, aphrodisiac, blood purifier, febrifuge, hypoglycaemic and stomachic [11-12]. The roots, leaves and tops are traditionally used in India, Indo-China and South-East Asia as an analgesic, diuretic and antipyretic to treat gastric disorders such as diarrhoea and dysentery and also for cough, bronchitis, hypertension, haemorrhoids and insect bites [13]. The plant is rich in fibre and its slimy consistency when cooked is used to treat various digestive problems such as diarrhoea, stomach ache, dysentery, constipation and ulcers. It is also claimed that together with other herbs, it can cure cancer [14].

Experimental

Sample collection and identification

The *Corchorus olitorius* leaf sample was collected from Nnewi-ichi, Nnewi, Anambra State, Nigeria. Sample of *S. dulcis* (aerial part) were collected from Mandara-Abdu Village, Biu, Borno State, Nigeria. The plant sample were collected in December, 2016, identified and authenticated by a Taxonomist from Biological Science Department, University of Maiduguri, Nigeria.

Sample preparation

Corchorus olitorius leaf sample were hand-picked and separated from the twigs and debris; while *Scoporia dulcis* were also collected, carefully separated from the root then air-dried in the laboratory at room temperature. The plant materials were

ground into coarse powdered using wooden mortar and pestle and then kept air-tight until extraction.

Extraction protocol

Two hundred and fifty grams (250 g) each of the sample was used for the extraction. The extraction of *Corchorus olitorius* was carried out using reflux apparatus for 4 hours employing 70% aqueous ethanol as solvent while that of *Scoporia dulcis* using 70% aqueous methanol adopting maceration technique for 4 days. The crude extracts were filtered differently using Whatman No. 1 filter paper, poured on an evaporating dish to dryness and extent of complete dryness confirmed by weight difference technique.

Phytochemical screening

The crude ethanolic leaf extract of *Corchorus olitorius* and that of aerial part crude methanolic extract of *Scoporia dulcis* were subjected to a phytochemical screening test to determine the presence or absence of the phytochemical constituent using standard procedure as described by several authors [15-21].

Evaluation of anti-diarrhoeal activities of castor oil-induced diarrhoea in rat

Experimental animals

Albino Wister rats of both sex weighing 130-260 g were used. Institutional Animal Ethics Committee approved the experimental protocol; animals were housed under room temperature of Maiduguri in December 2016 (23±2 °C), relative humidity (30-50%) and with a 12/12 light/dark cycle. The animals were given standard diet (Vital Feeds Ltd., Jos, Nigeria) and water provided *ad libitum*. Animal handling was performed according to Good Laboratory Practice (GLP).

Induction of diarrhoea using castor oil

The rats were fasted for 18 hours prior the experiment but allowed water *ad libitum*. The rats of either sex were divided into five groups of three rats each. Group I which served as control was administered with distilled water. Group II was administered a standard drug Diphenoxylate hydrochloride orally as suspension (5 mg/kg bd. wt.). After preparation of stock concentration by dissolving known concentration of crude extracts in distilled water. From the stock concentration, working dosages were prepared thus: Each of the extract was administered orally at 300 mg/kg bd. wt. dose to Group III, 600 mg/kg bd. wt. dose to Group IV and 1200 mg/kg bd. wt. to Group V rats as suspension. After 60 minutes of pre-treatment with the extract, the animals of each group received 2 mL of castor oil orally and were individually observed in a separate cage for about 6 hours. The watery faecal material and number of defecation was allowed to drop on a plain sheet of paper laid on the base of the cage [22-24].

The extract's efficacies were expressed as a percentage of diarrhoeal inhibition as:

$$\text{Percentage of wet faeces inhibition} = \frac{(T_0 - T_1)}{T_0} \times 100\% \quad [\text{Eq. 1}]$$

T_0 = number of wet faeces in control group, T_1 = number of wet faeces in test group.

$$\text{Severity of diarrhoea} = \frac{\text{Diarrhoeal faeces}}{\text{Total faeces}} \times 100\% \quad [\text{Eq. 2}]$$

Results and Discussion

The results of the phytochemical constituents of the 70% leaf extract of *Corchorus olitorius* and that of the aerial parts extracts of *Scoporia dulcis* is shown in Table 1. The Anti-diarrhoeal effects of the extracts are presented in Table 2.

Phytochemical evaluation

The phytochemical screening of both the leaf ethanol extract of *Corchorus olitorius* and the aerial part extract of *Scoporia dulcis* revealed the presence of cardiac glycoside,

tannins, flavonoids, free and combined reducing sugar. Alkaloid was absent in both extracts but saponins was present in only *S. dulcis* as shown in Table 1.

Table 1. Phytochemical of constituents of the leaf extract of *Corchorus olitorius* and the aerial parts of *Scoporia dulcis*

S/No	Phytochemical Constituent	<i>C. olitorius</i> (leaf)	<i>S. dulcis</i> (aerial part)
1	Test of alkaloids		
	a. Dragendorff's reagent	-	-
	b. Mayer's reagent	-	-
	c. Wagner's reagent	-	-
2	Test for Anthraquinones		
	a. Free anthraquinones	-	-
	b. Combined anthraquinones	-	-
3	Test of carbohydrate		
	a. Molisch's test	+	+
	b. Combined reducing sugar	+	-
	c. Free reducing sugar	+	-
4	Test for cardiac glycoside		
	a. Liebermann-Burchard's test	+	+
	b. Salkowski's test	+	+
	c. Keller-Killiani's test		
5	Test for flavonoids		
	a. Ferric chloride test	+	+
	b. Shinoda's test	+	+
	c. Lead ethanoate test	+	+
	d. Sodium hydroxide test	-	-
6	Test for phlobatannins	-	-
7	Test of saponins glycosides (frothing) test	-	+
8	Test for tannins		
	a. 1% ferric chloride	+	+
	b. 10% lead ethanoate	+	+
9	Test of terpenoids	-	-

Key: + = positive, - = negative

Table 2. Castor-oil induced anti-diarrhoeal effects of the leaf extract of *Corchorus olitorius* and aerial parts extract of *Scoporia dulcis* in rats

S/No	Extract/drugs	Dosage (mg/kg bd wt)	No. of defecation (Mean ± SEM)	Protection (%)	Severity of diarrhoea (%)
1	<i>Corchorus olitorius</i>	300	6.67±1.70 ^a	75.00	7.55
2	<i>Scoporia dulcis</i>	300	10.33±0.94 ^b	61.27	11.69
3	<i>Corchorus olitorius</i>	600	12.00±4.24 ^c	55.00 ^c	13.59
4	<i>Scoporia dulcis</i>	600	12.00±4.24 ^c	55.00 ^c	13.59
5	<i>Corchorus olitorius</i>	1200	10.33±5.31 ^b	61.27 ^b	11.69
6	<i>Scoporia dulcis</i>	1200	10.33±5.31 ^b	61.27	11.69
7	Normal saline (Control)	5 mL	26.67±2.05 ^d	0.00	30.19
8	Diphenoxylate HCl	5	0.00±0.00 ^e	100.00	0.00

n=3 animals per group; mean number of defecations with different superscripted letters are significantly different (p<0.05)

The presence of these phytoconstituents may be the reason why the plants are considered as medicinal plants and also responsible for the *in vivo* anti-diarrhoeal activity of the plants [26]. Report has it that flavonoid is an antioxidant and have been ascribed to their ability to inhibit intestinal motility and hydro-electrolytic secretions which are known to change diarrhoeal conditions. Tannins is astringent and has anti-diarrhoeal activity through denaturation of proteins in the intestinal mucosa by forming protein-tannins complex which may reduce secretions; research on role of tannins have also revealed that they could bring about similar functions by reducing the intracellular current or by activation of the calcium pumping system (which enhances muscle relaxation). Diarrhoea is usually considered as a result of altered motility and fluid defecation within the intestinal tract, it is one of the main causes of infant mortality in developing countries, causing about 5 to 8 million deaths a year, mainly among children under five years of age [1].

Anti-diarrhoeal effects

The leaf extract of *Corchorus olitorius* and aerial parts extract of *Scoporia dulcis* exhibited anti-diarrhoeal effects as presented in Table 2. The extract meets some of the criteria for acceptance of a drug as an anti-diarrhoeal [27]; these criteria include inhibition of the production of wet or unformed faeces in animals and inhibition of gastrointestinal propulsive action [28]. Both extracts produced a non-dose dependent anti-diarrhoeal effects and protection against the castor oil-induced diarrhoea with significantly (p<0.05) lowest mean number of defecations (6.67±1.70 and 10.33±0.94) and highest protection (75% and 61%) were obtained at the same test dose of 300 mg/kg respectively for *C. olitorius* and *S. dulcis* extracts; by prolonging and reducing the number of diarrhoea faeces. Although their effects are significantly (P<0.05) lower than that observed by standard drug (Diphenoxylate hydrochloride 5 mg/kg bd. wt.) which had 0.00±0.00 mean number of defecations as shown in Table 2; these findings deviate from the work of Rntnasooriya *et al.* (2005) [29], who worked on the anti-diarrhoeal effects of *S. dulcis*. The

C. olitorius extracts was more effective than the *S. dulcis* extract against castor oil-induced diarrhoea at all doses. However, same dosage of 300 mg/kg bd. wt. of both plant extract showed anti-diarrhoeal activity on castor oil induced model and this observation may suggest that the plant extract at low dose displays higher anti-diarrhoeal effects but with increasing dose (300-1200 mg/kg bd. wt.), the anti-diarrhoeal effects decreases; trend similar to this was observed by [30].

The *in vivo* anti-diarrhoeal indices revealed that the extract of *C. olitorius* had higher mean percentage protection of 63.76% compared to that of *S. dulcis* with mean percentage protection of 59.18%; consequent upon that, the severity of diarrhoea among the two extract was measured and it shows that *C. olitorius* 32.83% presented low mean percentage severity in relation to that of *S. dulcis* with high percentage severity of 36.97%. These expressions therefore, laid credence to the extract of *C. olitorius* as a better choice to that of *S. dulcis* comparatively as a remedy to diarrhoea. Anti-diarrhoeal activity was found in plants possessing tannins, flavonoids and terpenoids [31].

Conclusion

In conclusion, this study shows that *C. olitorius* and *S. dulcis* extracts were found to contain phytoconstituents such as flavonoids, tannins, terpenoids and steroids. The extracts reduced the faecal output and protected most of the rats from castor oil-induced diarrhoea in a non-dose dependent manner. These results reveal that *C. olitorius* and *S. dulcis* extracts may be use especially in rural settings for short-term symptomatic relief of non-specific acute diarrhoea and in the management of related cases. The authors wish to recommend for FT-IR, UV, HPLC and GC-MS of the crude extracts to ascertain the group(s) of phytochemicals responsible for the activity and also characterization of the

isolated compounds using spectroscopic techniques.

Acknowledgement

The authors wish to thank Mr. Fine Akawo of Research Laboratory, Department of Chemistry, University of Maiduguri, Nigeria for carrying out extraction of plant materials.

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How to cite this manuscript: Hamidu Usman*, Blessing Oluchukwu Mbonu, Faisal Abubakar Bello, Muhammad Awwal Tijjani, Kyari Abba Sanda, Halima Abdulsalam Umar, Sule Ibrahim, Levels of Castor Oil-Induced Diarrhoea in Rats Treated with Leaf Extract of *Corchorus olitorius* Linn and Aerial Part Extract of *Scoporia dulcis* Linn, *Adv. J. Chem. A*, **2020**, *3*(1), 1-8.