Antidiarrhoeal activity of aqueous and methanolic extracts of Oxalis corniculata Klotzsch. in rats

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ABSTRACT
The antidiarrhoeal activity of the aqueous and methanolic extracts of Oxalis corniculata (Oxalidaceae) was evaluated on castor oil-induced diarrhoea in rats and on small muscle intestinal transit. At orally administrated doses of 160, 320 and 640 mg/kg of body weight, the two plant extracts significantly (p<0.05) prolonged the time of onset of diarrhoea and inhibited the frequency of defecation. These extracts also reduced the wetness of faecal droppings in castor oil-induced diarrhoea, and decreased the propulsion of charcoal meal through the small intestine. At all doses, the aqueous extract appeared to be more effective than the methanolic extract. It is suggested that the extracts of O. corniculata may contain biologically active components that may be useful against diarrhoea, thereby justifying its use in ethnomedical practice as an antidiarrhoeal agent.

Keywords: Oxalis corniculata, antidiarrhoeal activity, antipropulsive effect, rat.

RESUME
L’activité antidiarrhéique des extraits aqueux et méthanolique de Oxalis corniculata (Oxalidaceae) a été évaluée sur la diarrhée induite expérimentalement par huile de castor et sur le transit intestinal de rat. Administrés par voie orale aux doses de 160, 320 et 640 mg/kg de poids corporel, les extraits de O. corniculata ont significativement (p<0.05) prolongé le temps d’induction de la diarrhée et inhibé la fréquence de défécation. Ces extraits ont également réduit le pourcentage de selles diarrhéiques et ralenti la propulsion du charbon à travers le tractus intestinal. A toutes les doses, l’extrait aqueux était plus efficace que l’extrait au méthanol. Ces observations laissent supposer l’existence dans ces extraits de plante, de composés bioactifs responsables de l’effet antidiarrhéique ; ce qui justifierait alors son emploi en médecine traditionnelle.

Mots clés: Oxalis corniculata; activité antidiarrhéique; effet antipropulsive; rat.

INTRODUCTION
In developing countries, diarrhoea represents a major cause of children’s morbidity and mortality. Faced with this problem, the World Health Organization (WHO) has set up a diarrhoeal disease control program (CDD) which includes studies of traditional medicinal practices [1]. Many plants have been screened for their antidiarrhoeal potential [2,3,4].

Oxalis corniculata Klotzsch. of the Oxalidaceae family, is one of such plants. In India, this plant has been used to treat aphthae [5]. Traditional healers in Cameroon used the mixture of leaf or whole plant powder and kernel oil for the treatment of several diseases including fever with obstruction of the throat, vomiting of blood, swelling and diarrhoea [6]. It is known under various vernacular names according to tribes: “Hangenangle/nangnagne” (Bekossi), “Nieugue”(BaMileke) or “Obelessi, Onyang” (Bulu) [7,8]. The present study was carried out using the aqueous and methanolic extracts of Oxalis corniculata to test their antidiarrhoeal effects.

MATERIALS AND METHODS
Plant material
The whole plant of Oxalis corniculata was collected in the locality of Dschang (West Cameroon) between April and October 2003. Botanical authentication was done by Dr Focho Derek of the Department of Plant Biology, University of Dschang, Cameroon. The plant specimen is deposited at the National Herbarium in Yaoundé, under the voucher number 19799/SRF/CAM. The plant was shade-dried and pulverised in an electric grinder (Moulineux).

Preparation of the aqueous extract
The powdered plant material (400 g) was dissolved in 4 L of distilled water at room temperature for 72 hours and occasionally stirred. After filtration, the solution obtained was evaporated in an oven (50° C) for 48-72 h to give 81 g of brownish residue corresponding to an extraction yield of 20.25%. The residue was kept at 4 °C until use. The aqueous extract used in our study was extemporaneous prepared by dissolving 1 g of the brownish residue in 10 mL of distilled water. The doses used in our study were 160, 320 and 640 mg/kg of body weight.

Preparation of the methanolic extract
The powdered material (200 g) was extracted with methanol (1 L) for 72 hours at room temperature. Following filtration and concentration under vacuum, 20.3 g of blackish residue was obtained with an extraction yield of 10.15%. The residue was kept at 4° C until use. The methanolic extract used in our study was extemporaneous prepared by dissolving 1 g of the blackish residue in 10 mL of distilled water. The doses used in our study were 160, 320 and 640 mg/kg of body weight.
Animals
Adult Wistar rats of either sex, aged 4 months and weighing 200-240 g were used in the study. They were housed in standard environmental conditions of temperature, humidity and light, and provided with standard Laboratory food and water ad libitum.

Antidiarrhoeal activity
The study was performed following the method of Amos et al [9] with minor modifications. The rats were fasted for 24 hours and randomly allocated to eight groups of 12 animals each: Group 1 serving as normal control received distilled water (10 mL/kg b.w) by gavage; Group 2 serving as positive control received orally 23.6 μg/kg b.w of loperamide (Janssen-Cilag) as standard drug; Groups 3 - 5 were treated with 100, 320 and 640 mg/kg b.w of aqueous extract respectively whereas Groups 6 - 8 were administered 160, 320 and 640 mg/kg b.w of methanolic extract respectively. After 1 h of drug administration, each rat was orally given 1 mL of castor oil. Animals were observed for defection up to 8 h. They were placed separately in an observation cage with filter paper placed at the bottom; the filter paper was changed after each defection. The following characteristic diarrhoeal parameters were calculated: diarrhoea latency, time elapsed from the administration of castor oil to when first faeces were produced; faeces frequency, total number of faeces (watery and non watery faeces) during the study (up to 8 h); wet faeces, total number of wet faeces (diarrhoeic faeces) during the study.

Effect on small muscle intestinal transit
In order to assess the effect of high dose (640 mg/kg b.w) of O. corniculata on the small intestinal propulsion, animals were fasted for 24 h and divided into four groups of five rats each. Each rat was treated orally with 1 mL of deactivated charcoal meal (Carbophos, Upsa). Animals of the first two groups received immediately after charcoal meal administration, the aqueous and methanolic extracts of O. corniculata respectively; the third group was orally treated with loperamide (29.6 μg/kg b.w) as standard drug, while the fourth group received distilled water (10 mL/kg b.w) by gavage. After 30 min, animals were killed and the movement of charcoal from pylorus to caecum was measured. The charcoal movement in the intestine was expressed in terms of percentage of control value (distilled water) [10,11].

Statistical analysis
Data were expressed as the mean ± S.E.M., 12 animals were used in each group. One way ANOVA was used to analyse data and significant means were separated using Duncan's at 5% level of significance.

RESULTS

Castor oil-Induced diarrhoea
Oral pre-treatment of rats with the aqueous and methanolic extracts of O. corniculata significantly (p<0.05) increased the castor oil-induced diarrhoea latency, and decreased the frequency of defection as well as the total number of watery faeces (wet faeces) when compared with untreated control rats. At all doses, the aqueous extract appeared to be more effective than the methanolic extract. Within the same extract, the aqueous extract at 160 mg/kg b.w gave the best result on the diarrhoea latency compared to both other doses. On the contrary, the methanol extract showed a dose-dependent action on all parameters (Table 1).

Effect on small muscle intestinal transit
The results revealed that both aqueous and methanol extracts of O. corniculata (640 mg/kg b.w) inhibited the propulsion of charcoal meal through the small intestine by 22.79% and 19.0% respectively. In the case of treatment with loperamide, the inhibitory propulsion rate was 48.18%. The aqueous extract was more effective than the methanolic extract (Table 2).

Table 1: Effect of aqueous and methanolic extracts of O. corniculata on castor oil-induced diarrhoea in rats

<table>
<thead>
<tr>
<th>Treatment groups</th>
<th>Dose (mg/kg b.w.)</th>
<th>Diarrhoea latency (min)</th>
<th>Faeces frequency (n/rat)</th>
<th>Total number of watery faeces</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (Distilled water, 10 mL/kg b.w.)</td>
<td>0.00</td>
<td>109.91 ± 19.39</td>
<td>3.0 ± 0.02</td>
<td>36</td>
</tr>
<tr>
<td>Loperamide</td>
<td>0.0296</td>
<td>224.33 ± 10.75</td>
<td>1.08 ± 0.05</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td>160</td>
<td>214.58 ± 16.82</td>
<td>0.83 ± 0.03</td>
<td>3</td>
</tr>
<tr>
<td>Aqueous extract of O. corniculata</td>
<td>320</td>
<td>138.75 ± 13.07</td>
<td>0.58 ± 0.02</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>640</td>
<td>200.16 ± 17.56</td>
<td>0.68 ± 0.03</td>
<td>1</td>
</tr>
<tr>
<td>Methanol extract of O. corniculata</td>
<td>160</td>
<td>143.44 ± 9.61</td>
<td>1.58 ± 0.04</td>
<td>8</td>
</tr>
<tr>
<td></td>
<td>320</td>
<td>179.08 ± 7.95</td>
<td>1.50 ± 0.02</td>
<td>4</td>
</tr>
<tr>
<td></td>
<td>640</td>
<td>168.50 ± 12.15</td>
<td>1.08 ± 0.04</td>
<td>3</td>
</tr>
</tbody>
</table>

All values: Mean ± S.E.M.
Number of rats per group, 12; n/rat: total number of faeces per rat.
* p<0.05 when compared with control (distilled water).
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Table 2: Dose response effect of aqueous and methanolic extracts of O. corniculata on small muscle intestinal transit in rats

<table>
<thead>
<tr>
<th>Treatment groups</th>
<th>Dose (mg/kg b.w.)</th>
<th>Percentage of distance travelled by charcoal meal (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (Distilled water, 10 mL/kg b.w.)</td>
<td>0.00</td>
<td>96.04 ± 1.77</td>
</tr>
<tr>
<td>Loperamide</td>
<td>0.0296</td>
<td>51.82 ± 6.73* (48.18)</td>
</tr>
<tr>
<td>Aqueous extract of O. corniculata</td>
<td>640</td>
<td>77.21 ± 3.66 (22.79)</td>
</tr>
<tr>
<td>Methanol extract O. corniculata</td>
<td>640</td>
<td>81.00 ± 1.40 (19.00)</td>
</tr>
</tbody>
</table>

All values: Mean ± S.E.M.  
Number of rats per group: 5  
Figure in parenthesis: represents the percentage of inhibitory propulsion rate.  
* p<0.05 when compared with control (distilled water).

DISCUSSION
The present work was aimed at determining the anti-diarrhoeal potential of the aqueous and methanolic extracts of O. corniculata against castor oil-induced diarrhoea in rats. The action of castor oil as diarrhoea inducer has been largely proven [9,12,13,14] and it is well known that its most active component is the ricinoleic acid which produces an irritating action that abolishes the muscle tone of the small intestine, thus causing changes in the fluid and electrolytic permeability and forward-propelling the intestinal contents [15]. Like loperamide, a standard anti-diarrhoeic drug, the aqueous and methanolic extracts from O. corniculata significantly protected the animals against castor oil-induced diarrhoea at all doses, as evidenced by the increase in the time of onset of diarrhoea on the one hand, and the significant reduction (p<0.05) of the faecal outputs on the other hand. It has already been demonstrated that loperamide, an opiate analogue, possesses an antinotility and antisecretory properties on the small intestine [16,17]. Its antinotility effect results from the increase in the small intestinal muscle tone whereas the antisecretory activity is due to the inhibition of the colonic mucus secretion and stimulation of the absorption of fluid, electrolytes and glucose along the small intestinal muscle without affecting the adeny cyclase/cAMP pathway [18,19,20]. The results obtained showed that the aqueous and methanolic extracts of O. corniculata may contain some biologically active principles that may be active against diarrhoea through their interference with motile and probably secretory processes of the small intestine. Similar observations have also been reported with the extracts of Pentaclethra macrophylla [21], Roueopsis obliquifoliate and Epinetrum illosum [22]. Furthermore, in order to clarify the effect of O. corniculata extracts on the small muscle intestinal transit, high dose (640 mg/kg b.w.) of aqueous and methanolic extracts was given to rats after charcoal meal administration. Since these extracts reduced the propulsive movement of the small intestine with significant effect obtained with the aqueous extract, it is likely that O. corniculata possesses anti-diarrhoeic property through its antinotility activity. The inhibition of gastrointestinal motility by some plant extracts have also been reported [9,23].

In conclusion, our results establish the anti-diarrhoeal effect of O. corniculata and could account for its use in traditional medicine for treatment of diarrhoea. However, phytochemical studies are needed to identify the active principles and assess their mechanism of action.

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REFERENCES


