

Short Communication

Phytochemical screening and anti-inflammatory evaluation of methanolic and aqueous extracts of *Euphorbia heterophylla* Linn (Euphorbiaceae)

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Euphorbia heterophylla is a local medicinal plant used in ethnomedicine for the treatment of constipation, bronchitis and asthma. The aqueous decoction and the methanolic extracts were subjected to anti-inflammatory activity using experimental animal model, in the presence of the positive control drugs. The inflammation was induced by carrageenan. From the results obtained the aqueous extract showed significant activity ($P < 0.001$) comparable to the reference drug used. At the different dose range used (50, 100 and 150 mg/kg), there was no significant differences in their anti-inflammatory activity hence they were not dose-dependent. However, the methanolic extract did not show any appreciable activity (20-24% inhibition) and were also not dose-dependent. The results of the study showed the justification of the use of the plant in the treatment of inflammatory disease conditions, and the active chemical constituents when isolated will be added to the present anti-inflammatory agents.

Key words: Anti-inflammatory activity, *Euphorbia heterophylla*, leaf extract.

INTRODUCTION

The use of medicinal herb in the treatment and prevention of diseases is attracting attention by scientists world wide (Sofowora, 1982). This is corroborated by World Health Organization in its quest to bring primary health care to the people. The plant kingdom has long serve as a prolific source of useful drugs, food, additives, flavoring agents, colourants, binders and lubricants. As a matter of fact, it has been estimated that about 25% of all prescribed medicines today are substances derived from plants (Gamaniel, 2000). The use of traditional medicine and medicinal plants in most developing countries, as a normative basis for the maintenance of good health, has been widely observed (UNESCO, 1996) Furthermore, an increasing reliance on the use of medicinal plants in the industrialized countries has been traced to the extraction and development of several drugs and chemotherapeutics from these plants as well as from traditionally used rural herbal remedies (UNESCO, 1998).

The medicinal usefulness of the plant *Euphorbia heterophylla* has been the object of numerous chemical and pharmacological studies. *E. heterophylla* is medicinal plant with the common name "spurge weed". It grows in semi-humid places especially in cassava, cowpea and Soya bean plantations. Report of previous chemical study on *E. heterophylla* is however, scanty. Recently we (Falodun et al., 2004) reported the isolation of a flavonoid quercetin from the leaves of this plant. The leaves are known to possess antibacterial activity (Falodun et al., 2003).

In the Igbo community of Nigeria, *E. heterophylla* is used as a purgative (Erden et al., 1999). The leaves are used to cook "yam poradge" and purgation ensues 3 - 4 hours after consumption. Extracts of the decoction of the leaves is also used in the treatment of respiratory tract infections and asthma by traditional medicine practitioners.

The study was therefore aimed at investigating the anti-inflammatory activity of the leaf extract with a view to Justifying the use of the plant in the treatment of inflammatory disease conditions such as asthma.

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Table 2. Anti-inflammatory activity of the aqueous and methanolic extract of the leaves of *Euphorbia heterophylla*.

Extracts	Doses (mg/kg, p.o)	Change in paw oedema mean (mm)	% Oedema inhibition relative to control at the 4 th hour
Control (normal Saline,0.9%)	0.3 ml	2.75 ± 1.35	-
Indomethacin	10	1.12 ± 0.12**	59.10
Aqueous extract	50	1.33 ± 0.48**	51.50
	100	1.17 ± 0.32**	57.60
	150	1.15 ± 0.25**	58.20
Methanolic extract	50	2.20 ± 0.35	20.10
	100	2.15 ± 2.45	21.82
	150	2.10 ± 0.58	23.64

Values are mean ± S.E.M.

*P < 0.05, ** P < 0.001, significantly different from control.

Paired t- test (n = 5), p.o = per oral.

MATERIALS AND METHODS

Collection of plant materials

Fresh leaves of *Euphorbia heterophylla* excluding the fruits and flowers were collected from the bush behind the nursing hostel at Ugbowo campus of the University of Benin, Benin City. The plant was identified and authenticated by Mr. A. Abubakar of the Department of Pharmacognosy, Faculty of Pharmacy, University of Benin, Benin City. A voucher specimen is kept in the herbarium of the same department.

Extraction

Aqueous extract: Fresh leaves of *E. heterophylla* were boiled with 2.5 l of distilled water for 10 min. It was filtered hot and evaporated to dryness using a rotary evaporator attached to a vacuum pump.

Methanol extract: Dried leaves of *E. heterophylla* were reduced to a fine powder with a mechanical grinder. The powder plant material (200 g) was extracted by maceration for 48 h, and concentrated to dryness using a rotary evaporator attached to a vacuum pump and stored at a temperature of -4°C until use.

Chromatography

The crude methanolic and aqueous extracts were subjected to phytochemical screening (Trease and Evans, 1970).

Anti-inflammatory activity

Wistar rats (120 - 170 g) of either sex kept at the laboratory Animal home of the Faculty of Pharmacy, University of Benin, Benin City, Nigeria were used. The animals were maintained under standard environmental conditions and had free access to standard diet and water (plant extracts were administered orally by gavage in distilled water at different dose levels).

Anti-inflammatory activity was measured using carrageenan-induced rat paw oedema assay (Winter et al., 1962; Adeyemi et al., 2002). Groups of 5 rats of both sexes (pregnant females excluded) were given a dose of the extract. After 1 h, 0.1 ml, 1% carrageenan suspension in 0.9% NaCl solution was injected into the sub-plantar tissue of the right hind paw. The linear paw circumference was

measured at hourly interval for 4 h (Bamgbose and Noamesi, 1981). Two groups of drug treated rats and one control group were used each test day, the mean paw oedema value for the test group being compared with its mean value for the control group for that day.

Anti-inflammatory activity (Duffy et al., 2001) was measured as the percentage reduction in oedema level when drug was present, relative to control as shown in Table 2.

Activity = 100 - (100 x average drug treated / average for control).

Indomethacin (10 mg/kg) was administered orally as reference drug while distilled water was used as negative control.

Statistical analysis

All data were expressed as mean ± SEM and the student's *t*-test was applied to determine the significance of the difference between the control group and mice treated with the test compounds.

Table 1. Phytochemical compositions of *Euphorbia heterophylla* extracts.

Chemical	Aqueous extract	Methanolic extract
Saponins	+	+
Alkaloids	-	-
Tannins	-	-
Flavonoids	+	-
Terpenes	+	-

+: present

-: absent

RESULTS AND DISCUSSIONS

The phytochemical studies revealed the presence of flavonoids, saponins, diterpenes and phorbol esters in the extracts (Table 1). Carrageenan-induced rat paw oedema is used widely as a working model of inflammation in the search for new anti-inflammatory drug

(Manueli et al., 1994) and appeared to be the basis of the discovery of indomethacin anti-inflammatory drug (Winter et al., 1963). The anti-inflammatory activities of the aqueous and methanolic extract of *E. heterophylla* were evaluated by carrageenan-induced rat paw oedema method (Winter et al., 1962; Adeyemi et al., 2002) and the result is shown in Table 2. The extracts were tested at three different dose levels to know if they were dose-dependent. From the results obtained the aqueous extract showed significant activity ($P < 0.001$) comparable to the reference drug used. At the different dose range used (50, 100 and 150 mg/kg), there was no significant differences in their anti-inflammatory activity hence they were not dose-dependent. However, the methanolic extract did not show any appreciable activity (20-24% inhibition) and were not dose-dependent. The results obtained complimented the earlier investigation (Falodun et al., 2003) that the aqueous leaf extract of *E. heterophylla* contains copious amount of the flavonoid quercetin, a known anti-inflammatory agent. The significant level of anti-inflammatory activity of the aqueous extract could be attributed to high amount of flavonoids present in the extract. This study also lends support to the fact that *E. heterophylla*, unlike most euphorbia, does not cause inflammation or irritation to the skin during physical handling of the material. More importantly, the research work justified the traditional use of the plant in the treatment of inflammatory disease conditions such as asthma. Further studies will be carried out to isolate and characterize other anti-inflammatory chemical constituents present in the aqueous extract of the plant.

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