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PHYTOCHEMICAL AND ANTI INFLAMMATORY EVALUATION OF *KHAYA GRANDIFOLIOLA* STEM BARK EXTRACT

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ABSTRACT: The ethnomedicinal application of *Khaya grandifoliola* stem bark in Africa as remedy against rheumatoid arthritis is attracting attention in view of the global focus on herbal medicine as alternatives to orthodox drugs. And this necessitated this study.The methanolic extract of the plant was subjected to phytochemical screening testing for alkaloids, tannins, saponins and flavonoids as secondary metabolites.Anti inflammatory activity of the methanolic extract was examined using carrageenan, and serotonin –induced, paw oedema and xylene induced ear oedema. The methanolic extract significantly inhibited the carrageenan-induced paw oedema from the first hour to the fifth hour at a dose of 200mg/kg while a dose of 500 mg/kg significantly inhibited the carrageenan-induce paw oedema after 3 hours of carrageenan challenge. Whereas in the serotonin induced paw oedema test, the extract significantly reduces the paw oedema at the 120th minute at the dose of 200 mg/kg and at 90th minute at the dose of 500 mg/kg. For the xylene induced oedema there was a 28.6 % inhibition at 200 mg/kg and 57.1% at 500 mg/kg. The result of the study revealed that the methanolic extract of K.grandifoliola was active at different doses tested. This gives justification to the use of the plant as anti inflammatory medicinal plant.
Keywords: *Khaya grandifoliola*, phytochemical analysis, anti inflammatory activity, carrageenan-induced paw moedema

INTRODUCTION

Acute inflammation is a short-term process which is characterized by the classic signs of inflammation - swelling, redness, pain, heat, and loss of function - due to the infiltration of the tissues by plasma and leukocytes. It occurs as long as the injurious stimulus is present and ceases once the stimulus has been removed, broken down, or walled off by scarring (fibrosis).

The process of acute inflammation is initiated by the blood vessels local to the injured tissue, which alter to allow the exudation of plasma proteins and leukocytes into the surrounding tissue. The increased flow of fluid into the tissue causes the characteristic swelling associated with inflammation since the lymphatic system does not have the capacity to compensate for it, and the increased blood flow to the area causes the reddened colour and increased heat. The blood vessels also alter to permit the extravasation of leukocytes through the endothelium and basement membrane constituting the blood vessel. Once in the tissue, the cells migrate along a chemotactic gradient to reach the site of injury, where they can attempt to remove the stimulus and repair the tissue.

Medicinal plants are of great importance to the health of individuals and the society. The medicinal value of these plants lies in some chemical substances that produce a definite physiological action on the human body [1].

Plants as sources of remedy for many diseases date back to the early century [2]. In Africa, particularly West Africa, new drugs are often beyond the reach of the poor. Hence, upto 80 % o the population uses medicinal plants as remedy against infections and diseases [3].

Treatment offered by traditional healers is primary health care that has sustained the Nigerian community till now and the medicinal plants used are selected not on the basis of their chemical constituent, but on their perceived ability to restored patients disease condition to normal [4].

Khaya grandifoliola is a medicinal plant endemic to Nigeria. It is a tall, woody tree belonging to the family Meliaceae, and commonly called African Mahogany[5]. It is widely distributed across West Africa from the Guinea coast to Cameroon and extending eastward through Congo Basin to Uganda and some parts of Sudan. It

grows up to 40 m high and 5m girth. The bark is grey in colour and yield a bitter gum when wounded.
The wood ash of K. *senegalensis* is used for storing millet seeds to preserve them for planting the following year[6]. The anti malarial, antiulcer, anti anaemic and hypoglycaemic activities of the stem bark was also reported [7-10]. Some of the chemical constituents reported include limonoids [11]. This present study was necessitated in order to justify the folkloric usage of the plant in the treatment of inflammatory diseases.

EXPERIMENTAL

Collection and Identification of Plant material

The stem bark of the tree plant *Khaya grandifoliola* was collected by Mr. Simon Peters from a Local Government Area in Benue State, Nigeria in April, 2008. The plant was identified by Prof. Idu of the Department of Botany, Faculty of Life Sciences, University of Benin, Benin City, Nigeria and a voucher specimen is deposited in our laboratory.

Preparation and Extraction of plant sample

The fresh stem bark of *K.grandifoliola* was chopped into pieces and sun dried for a period of two weeks, reduced to fine powder with the aid of a mechanical grinder. The milled powder sample was collected and stored in glass jars, tightly covered and kept for further studies in refrigerator (-4oC).

Extraction of the plant material (800g) with methanol (3L) by maceration for 48 hours and filtration of the extract was carried out at room temperature 25^{o}C. The reddish brown extract was concentrated to dryness using a rotary evaporator at 30^{o}C at reduced pressure. The dried extract was stored in a refrigerator at -4 ^{o}C until use.

Phytochemical screening

Phytochemical tests were carried out on the powdered sample using standard experimental procedures, to identify the constituents as described by [12].

RESULTS AND DISCUSSION

The phytochemical analysis revealed the presence of alkaloids, saponins, and tannins and this agreed with the work of Stephen et al[13].
The effect of *Khaya grandifoliola* on carrageenan-induced paw oedema is shown on table 1. The extract (200 mg/kg) significantly ($p<0.05$) reduced the paw oedema. The extract (500 mg/kg), at the third hour Maximum reduction of paw oedema was seen at the second hour as the post pretreatment produced a significant ($p<0.05$).The effect of the extract was more pronounced at the dose 200 mg/kg body weight. Table 2 shows the effect of *K.grandifoliola* on serotonin – induced paw oedema. The effect on serotonin – induced paw oedema was dose dependent though not significantly different from the normal saline treated animal except at the 90th and 120 minutes. Cyproheptadine (10 mg/kg), a serotonin antagonist significantly ($P<0.001$) reduced the serotonin induced paw oedema in the rats compared to the normal saline treated animal. The methanolic extract of the plant exhibited inhibition of 28 % in xylene induced ear oedema at 200 mg/kg, with a corresponding increase to 57.1% as dosage increased to 500 mg/kg. Dexamethasone (positive control) produced 42.9% inhibition at a dose of 15 mg/kg as shown in Table 3.
Indomethacin (10 mg/kg) the reference drug (positive control) significantly ($p<0.00l$) reduce the paw oedema. The effect of the extract was more pronounced at the 200 mg/kg body dose.

CONCLUSION

The present study established the anti inflammatory activity of the methanol extract of *K.grandifoliola* in a number of experimental rats and mice models. The work also demonstrated the potential of *K.grandifolila* as anti-inflammatory agent and also lends support to the use of the plant for which it is known and used for

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Table 1: Effect of methanol extract of *K. grandifoliola* on carrageenan induced paw oedema

		Paw Oedema (Paw circumference)						
Treatment	Dose mg/kg	Time						
		0	**1**	**2**	**3**	**4**	**5**	**6**
Normal saline	-	4.42±0.24	7.2±0.13	7.54±0.24	7.60 ± 0.24	7.14±0.15	7.0±0.003	6.66±0.17
K.g	200	4.36±0.16	6.7±0.13	6.24±0.25	6.82 ± 0.16	6.12±0.15	6.34±0.24	6.44±0.37
K.g	500	4.55±0.23	6.93±0.32	6.93±0.38	6.10 ± 0.34*	6.33±0.30*	6.30±0.17*	6.73±0.44
Indomethacin	10	4.64±0.36	5.60±0.29	5.74±0.15	5.48±0.15***	5.80±0.32**	5.88±0.30**	5.76±0.30*

Values are mean ± SEM, *P<0.005, **P< 0.001, **P<0.0001, significantly different from the control (n=5) Student's t-test

TABLE 2: Effect of methanol extract of *K. grandifiliola* on serotonin Induced paw oedema

Treatment	Dose (mg/kg)	Paw Oedema (Paw Circumference) mm				
		Time				
		0	30	60	90	120
N. S	-	4.88± 0.02	7.10 ±0.09	6.50 ± 0.08	6.33±0.08	6.05 ± 0.01
K.G.	200	4.46 ±0.04	6.18 ±0.63	7.06 ± 0.66	6.28±0.33	5.82 ± 0.09*
K.G	500	4.46 ± 0.07	6.88±0.13	7.18 ± 0.71	6.70±0.1*	6.48 ± 0.13*
Ind	10	4.48 ± 0.09	6.10±0.14**	4.8±0.24**	4.8±0.2**	4.42 ± 0.06**

Values are Mean ± SEM, *P < 0.05, **P < 0.001, significantly different from the control, (n = 5), Student's t-test
N. S; normal saline,
K.G; *K.grandifoliola*
Ind; indomethacin

TABLE 3: Effect of methanol extract of *K. grandifoliola* on xylene – induced ear oedema

Treatment	Dose (mg/kg)	Ear Swelling (mg)	Percentage Inhibition
Normal Saline	-	17.5 ±7.5	-
K.G	200	12.5 ± 2.5	28.6
K.G	500	7.5 ± 4.8	57.1
Dexamethasone	15	10.0 ±4.1	42.9

Values are Mean ± SEM, (n = 4), Anti-inflammatory activity is presented as percentage inhibition, K.G; *Khaya grandifoliola*

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