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FRACTIONATION OF *MAERUA ANGOLENSIS* STEM BARK EXTRACT INTO THREE DIFFERENT COMPONENTS

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ABSTRACT

The stem bark of *Maerua angolensis* is traditionally used to reduce pain and also in management of epilepsy, peptic ulcer, diarrhea, arthritis, diabetes etc. In this study preliminary test was carried out to determine the most active of the stem bark extracts of *Maerua angolensis*, which was extracted with hexane, acetone and methanol in order of their increasing polarity. The extracts were screened against antimicrobial activity against gram +ve bacteria (*Staphylococcus aureus*), gram –ve bacteria (*Escherichia coli*) and fungi (*Candida albicans*) using the cup plate agar diffusion method. The result showed that the methanol extract is most active. The methanol extract which is the most active of all the extract was then successfully fractionated into acidic, basic and neutral plant components.

Keywords: *Maerua angolensis*, antimicrobial, fractionation, stem bark, extract

INTRODUCTION

Medicinal plant is any plant which has one or more of its organs, contain substances that can be used for therapeutic purpose or which are precursors for the synthesis of useful drugs (WHO, 1992).

Maerua angolensis is a tropical plant that is found in most parts of Africa and is mostly found in the rocky areas and ant hills. It is planted on graves in Nupe areas of Nigeria (Iliya *et al.*, 2014). It is an important medicinal plant which is used in managing various types of diseases such as diabetes, epilepsy, peptic ulcer, diarrhea, skin infection, arthritis etc. (Iliya *et al.*, 2014). Traditional medicine healers used either the stem bark, leaves or a combination of the two or all to treat patients (Iliya *et al.*, 2014).

In Nigeria, *Maerua angolensis* is found mostly in the North eastern and North western parts of the Country. The plant is called *CICIWA* in Hausa

language, *LEGCEL BAALI* by Fula-Fulfulde of Nigeria and BEAD BEAN TREE in English. *Maerua angolensis* is traditionally used to treat Jaundice, Psychosis, vomiting, boils pimples, bad spirits and prevent abortion (Hassan, 2012).

The aim of this work is to fractionate the active extracts into acidic, basic and neutral components.

Different solvent extracts of the leaf had been reported to exhibit antimicrobial activities (Benneh *et al.*, 2018; Yusuf *et al.*, 2017; Ayo *et al.*, 2013). Yusuf *et al.* (2017) reported an activity of 200 µg/mL against clinical isolates of *Staphylococcus aureus* and *Escherichia coli* for the leaf ethanol-extract. A methanol extract of the leaf was found to be active against *S. aureus* (ATCC 13704), *Streptococcus pyogenes* (Local strain), *Corynebacterium ulcerans* (Local strain), *Bacillus subtilis* (NCTC 8230), *E.coli* (NCTC 10418), *Salmonella Typhi* (ATCC 9184), *Klebsiella pneumonia* (ATCC 10031), *Pseudomonas*

aeruginosa (NCTC 6750), *Neisseria gonorrhoeae* (NCTC 10341) and *Candida albicans* (ATCC 10231) at 50 mg/m (Ayo et al., 2013). The broad antimicrobial activity is believed to be responsible for the wound healing properties of the plant and its use in infectious diseases in ethno medicine (Ayo et al., 2013). The plant has been demonstrated to exhibit strong anti-oxidant activity by Meda et al. (2013). Further studies suggested that the bark is non-toxic in anti-inflammatory doses, supporting ethno medical use of the plant in managing inflammation (Meda et al., 2013; Ayo et al., 2013). It is found in bush and rocky areas of the Savannah in tropical Africa. Various parts of the plant notably the leaves, roots and stem barks are claimed to relieve pain and also used to manage psychosis, epilepsy, diabetes, peptic ulcer,

diarrhea and arthritis in the traditional medicine (Magaji et al., 2009).

Phytochemical study indicated that the methanolic stem bark extract has steroids, saponins, tannins, terpenoids, flavonoids, alkaloids, glycosides, carbohydrates and proteins (Adamu et al., 2007). Many of these bioactive substances are involved in the modulation of pain sensation. *Maerua angolensis* known to contain bioactive substances involved in the modulation of pain sensation and used traditionally in the treatment of pain might also have some protective effects on the withdrawal syndrome of morphine dependence in its analgesic activity. This study, therefore, assessed the analgesic effect of the petroleum ether/ethyl acetate extract and fractions prepared from the stem bark of *Maerua angolensis* in the mouse tail-flick test using Hargreaves thermal hyperalgesia model. The effect of the extract and fractions on morphine dependence was also assessed in mice (Adamu et al., 2007).

MATERIALS AND METHOD

Plant Sampling

The plant (*Maerua angolensis*) was collected from the bush area of Toro Local Government area of Bauchi State and was identified by a botanist of Biological Sciences Department, Abubakar Tafawa Balewa University Bauchi State, Nigeria

Preparation of Sample

The stem bark of *Maerua angolensis* was collected and dried under shade for the period of three weeks and powdered with a laboratory pestle and mortar and after which it was sieved to obtain a fine powder. The powdered material was packed in clean polythene bags in order to avoid contamination (Zhang, 2004).

Extraction Method

The extraction was carried out using soxhlet extractor with hexane, acetone and methanol in order of their increasing polarity. 100g of the powdered stem bark of *Maerua angolensis* was weighed accurately using LA 164 weighing machine and introduced into a porous thimble made of Whatmann filter paper and placed in the inner tube (Chamber) of the Soxhlet fitted to 500 cm³ round bottomed flask containing about 250 cm³ of the solvent and the set up was mounted on a burner and held in place with a retort stand and clamps. The power button was switched on and the temperature was adjusted closely to the boiling

point of the solvent in use. The extraction process was run for about 8hrs until colorless liquid was seen for some time in the sample chamber or the extractor. The extract solution was filtered while hot and concentrated on a rotary evaporator at about 40°C and then kept for some days in the oven at about 40°C in order to eliminate or remove the remaining solvent. The process was repeated with each of the solvents (Magaji et al., 2009).

Biological Assay

This is the study of antimicrobial activity of the crude extracts against some microorganisms. It was used as a guide to determine the active components of the stem bark extracts.

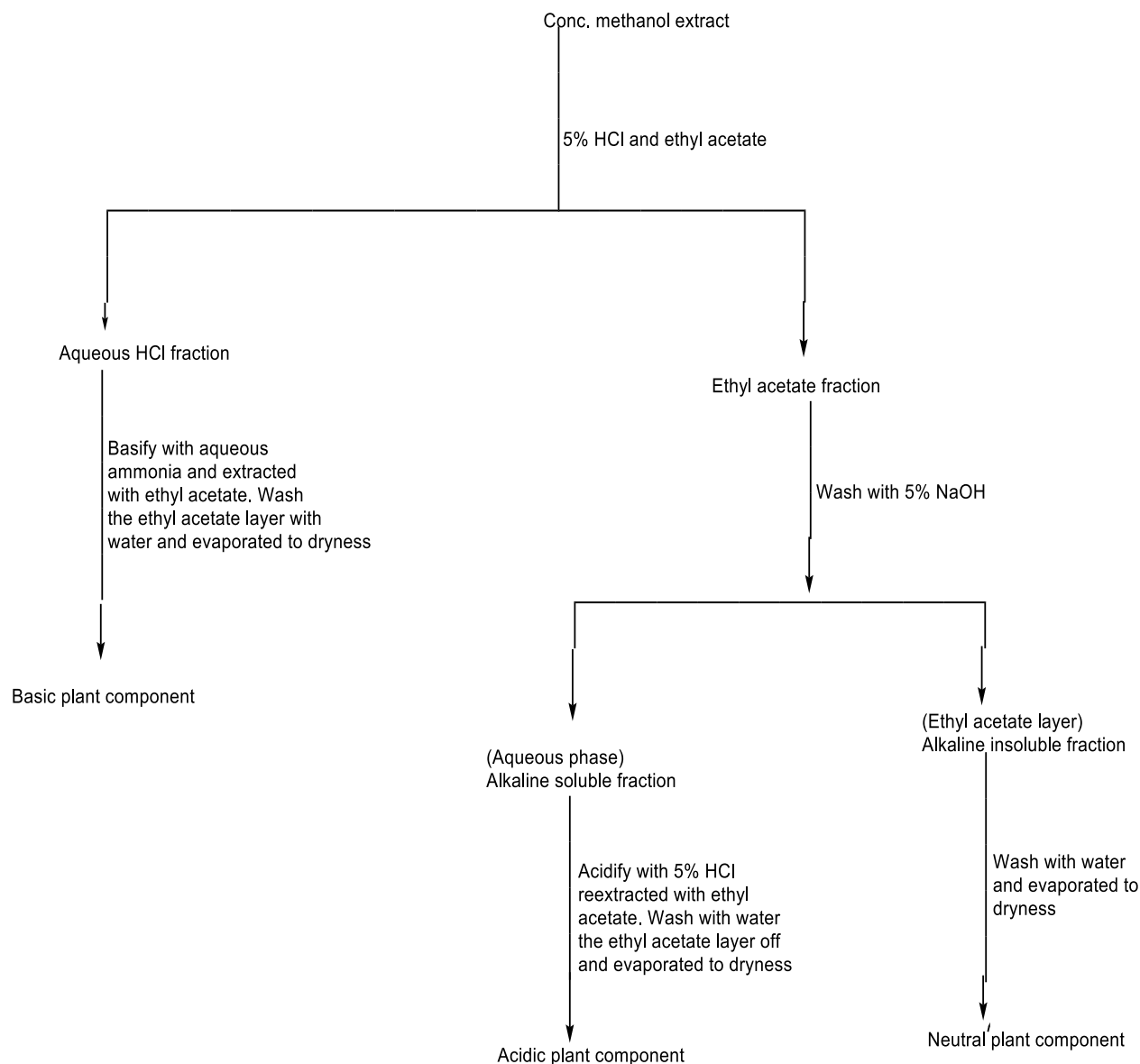
Test Organisms: Three different test organisms were selected based on their varying characteristics. They include gram positive bacteria (*Staphylococcus aureus*), gram negative bacteria (*Escherichia coli*) and fungi (*Candida albicans*). The test organisms were obtained from the Biological sciences department of ATBU. The choice of these pathogens was based on their implication or disease caused in human such as diarrhea, skin diseases, diabetes, dysentery, peptic ulcer, epilepsy, arthritis etc.

Procedure for Fractionation: The plant material was exhaustively extracted with methanol in a soxhlet apparatus. Concentration of the extract gave residue (A). The extract was partitioned between 5% HCl and ethyl acetate to give fractions

B and C respectively. The ethyl acetate fraction C was wash several times with 5% NaOH and partitioned. The aqueous phase contained the alkaline soluble fraction D. The ethyl acetate layer contained the alkaline insoluble fraction E. Fraction E was washed with water and evaporated to dryness in order for it to give a neutral plant extract. The alkaline soluble component was obtained by acidifying D fraction with 5% HCl and re-extracted

with ethyl acetate. The ethyl acetate layer was washed with water and evaporate to dryness to yield the acidic plant component F. The initial aqueous HCl fraction B was basified with aqueous ammonia and extracted with ethyl acetate. Evaporation of this layer after washed with water gave G component which is basic in nature (Mitscher, 2009).

THE FRACTIONATION PROCEDURE IS DIAGRAMATICALLY SHOWN BELOW



RESULTS AND DISCUSSION

Table 1: Results show the nature and quantity of the crude extracts of the stem bark of *Maerua Angolensis*

Solvents	Color of extracts	Texture	Weight of powdered sample (gram)	Weight of crude extracts (gram)
Hexane	Dark green	Oily and sticky liquid	100	8.00
Acetone	Dark green	Semi-solid	93.5	9.40
Methanol	Brown	Semi-solid	76.5	10.01

Table 2: Results of antimicrobial activity of the stem bark extracts of *Maerua Angolensis* on the test organisms

Organisms	Zone of Inhibition		
	Hexane	Acetone	Methanol
<i>Staphylococcus aureus</i>	10	14	25
<i>Escherichia coli</i>	12	15	26
<i>Candida albicans</i>	08	12	12

Table 3: Color and quantity of most active extract (methanol extract)

Component	Color of extracts	Quantity of extract (g)
ACID	Dark green	0.8
BASE	Brown	0.7
NEUTRAL	Dark green	1.6

DISCUSSION OF RESULTS

From the results obtained in table 1; appreciable recovery of 8.00, 9.40 and 10.01 were obtained using hexane, acetone and methanol respectively. It shows that methanol solvent extract yield more compared to acetone solvent and acetone solvent extract yield more compare to hexane solvent, this can be as a result of their polarity, which shows that the higher the polarity the higher the rate/yield of extraction.

Table 2 which shows the results of the antimicrobial activities on the test organisms revealed that both acetone and methanol extracts showed appreciable activities against *Escherichia coli*, *Staphylococcus aureus* and *Candida albican* while hexane extract showed no activity against

Candida albican and *Staphylococcus aureus* but was active against *Escherichia coli* (Ehssan et al., 2014).

The methanol extract was successfully fractionated into acidic, basic and neutral components and each of the components obtained from fractionation process was tested using litmus paper for confirmation.

CONCLUSION

Hexane, acetone and methanol solvents were sequentially used to extract stem bark of *Maerua angolensis* and the most active extract was successfully fractionated into acidic, basic and neutral components. The most active extract was known from the results of antimicrobial test run on the selected organisms.

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