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## PHYTOCHEMICAL SCREENING AND ANTIMICROBIAL EFFICACY OF EXTRACTS FROM *FICUS EXASPERATA* AGAINST HUMAN PATHOGENIC BACTERIA

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### ABSTRACT

Ethanol and water extracts of *Ficus exasperata* were screened for their phytochemical and antimicrobial activity against *Escherichia coli*, *Staphylococcus aureus*, *Streptococcus spp.*, *Salmonella typhi*, *Alpha hemolytic Streptococcus* and *Klebsiella pneumonia*. The result indicated that saponins, tannins, flavonoids, glycosides and steroids were present in all the plants part except for flavonoids which is absent in leave of water extract. The root, stem bark and leaves of the water extract were active on most of the microorganisms. But *Salmonella typhi* developed resistance against the leave for the water extract and *Escherichia coli* and *Protus (Mirabilis)* developed resistance against the root for the water extracts. The stem bark of the ethanol extracts was active on most of the microorganisms except for *Staphylococcus aureus* which developed resistance. The roots of the ethanol extract which developed resistance against most of the microorganism except for *Alpha hemolytic streptococcus* which developed zone of inhibition. The leaves of the ethanol extracts developed zone of inhibitions on *Protus (mirabilis)* and *Alpha hemolytic streptococcus* but the other microorganisms are resistance. This attest to the fact that *Ficus exasperata* contains bioactive compounds of potentially therapeutic and prophylactic significance and thus could by a promissory candidate for drug development and validates folkloric claim, as a cure for ulcer, cancer, intestinal pains and other stomach related problems, typhoid and asthma.

**Key word:** *Ficus exasperate*, *klebsiella pneumonia*, extracts, steroids, microorganisms

### INTRODUCTION

Plants are known as living things that manufactured their own food by themselves through a process called photosynthesis, in the presence of sunlight and chlorophyll. Plants are source of all, food for human and animal consumption, to provide other material for human activities, and a habitat for wildlife and birds and it also contribute to soil building and provides crude sources for most drugs. Most of the drugs that are used in modern medicine were originally from plants, although many others came from fungi (Mc. Graw Hill, 1997). Medicinal plants drug discovery continues to provide new and important drugs against various pharmacological targets including pain and inflammation (Balunas and Kinghorn, 2005). In Ghanaian traditional medicine, various parts of several plants are used either alone, or in combination thereby in the treatment of various painful inflammatory conditions (Braddock, 2007). Phytochemical components of these plants usually employed in the treatment of disease especially now that there are proposed on the integration of traditional medicine in health care programmes in Nigeria. For example, an anti-hypertensive drug obtained from *Anacardium occidentale* bark by extraction and separation of the tannin

fraction containing the antihypertensive principles using caffeine. In addition, investigation into the antimicrobial activities of these medicinal plants will reveal that those plants are potential sources for synthesis of drugs (Ebena et al., 1991). Medicinal plants represent a rich source from which antimicrobial agents may be obtained. Plants are used medicinally in different countries and are a source of many potential powerful drugs (Srivastava et al., 1996). Therefore, research into the effects of these local medicinal plants is expected to enhance the use of these plants against diseases caused by the test pathogens. However, most of these plants used on folk medicine have not been screened for their antimicrobial activity. The active principles of many drugs found in plants are secondary metabolites (Ghani, 1990; Dobelis, 1993). Therefore basic phytochemical investigation is vital. The identification and isolation of such active compounds makes it more effective therapeutic application, it prevent consumers from taking certain plants that have no medicinal value or poisonous to them. It will lead to better understanding of disease.

*Ficus exasperata* vahl, also known as sand paper tree belong to the family Moraceae with 800 species occurring in the warmer part of the world chiefly in Indonesia and Malaysia (Odunbaku et al., 2008). It is found in the savanna, rain forest beside rivers and streams. In Nigeria we have over 45 different species of *Ficus* (keay and onochie, 1964), such as *Ficus glomose*. It is a terrestrial agro-tropical shrub or a tree that grows up to about 20m tall and prefer evergreen and secondary forest habitat (Berg, 1989; Berg and Wiebes, 1992). It occurs in shrubs or tree with alternate leaves. The leaves shade soon as the fruits are ripe. The fruits are yellow with a sweet taste. The bark is gray and leaves are evergreen. The seed is 1cm long and 0.8cm broad hard with an oval structure. The leaves are 8-12cm long and 3.5-4.2cm broad. The plant is known to tolerate drought, high temperature, sandy and stony soils. (Odunbaku et al., 2008). In African tradition medicine, the leaf extract of *ficus exasperata* has been used to treat high blood pressure, rheumatism, arthritis, intestinal pains, epilepsy, bleeding and wounds (Akah et al., 1998; Ayinde et al., 2007). The roots are also used to manage asthma (chabra et al., 1990). There are some reports on scientific validation of some of the traditional uses of the plants found in literature. The hypotensive effects of the extract was significantly reduced with a prior administration of either atropine or chlorpheniramine, suggesting the probable stimulation of muscarinic receptors in the heart or release of histamine into the circulatory system thereby causing the initial fall in blood pressure (Ayinde et al., 2007). Also the extract had a significant anti-ulcerogenic property in aspirin-induced ulcerogenesis, delayed intestinal transit, increased the pH and decreased both the volume and acidity of gastric secretions (Akah et al., 1998)

*Ficus exasperata* (sand paper) leaves are used for polishing woods (Causins and Micheal, 2002), stabilization of vegetable oils and suppression of foaming. The plant is medicinally used for treatment of cough, hypertension and ulcer (Odunbaku et al., 2008). Despite its widespread multifunctional medicinal application there is dearth in medicinal research documentation of the plant. The aim of this work was to evaluate the antimicrobial potentials of this plant against some pathogenic bacteria and to determine the phytochemical constituent of the plant extracts.

## **MATERIALS AND METHODS**

### **Sampling and sample preparation**

The fresh sample of roots, stem bark and leaves of *Ficus exasperata* were collected in Nambare – Njoboli, Yola North Local Government, Adamawa State, and identified by Mal. Aliyu Bawuro Mustapha, Department of Crop Production, Modibbo Adama University of Technology, Yola. The fresh leaves, stem bark and roots of *Ficus exasperata* were air-dried at room temperature followed by pulverization to powdered form using a mortar and pestle. The powdered plant's parts were subjected to aqueous and organic solvent extraction.

### **Extraction**

20g of the powdered plant samples (stem bark, leaves and roots) were weight separately in different beakers and percolated with 150ml of ethanol each of these beakers were properly sealed with aluminum foil and left for 72 hours. The solutions were then filtered using a funnel fitted in a filter paper and the extracts obtained. The extracts obtained were concentrated using rotary evaporator at 40<sup>o</sup>C. The extracts were stored in a universal bottle and refrigerated at 4<sup>o</sup>C prior to use (Mann et al., 2008). The above procedure was repeated on 20g each of the powdered roots, stem-bark and leaves of the plants with the use of 150ml of distilled water.

### **Phytochemical Screening**

The preliminary phytochemical analysis of the extracts were carried out to determined the presence of tannins, flavonoids, saponins, alkaloids, steroids, phenols and glycosides as described by (Odebiyi and Sofowara, 1990) and (Fadeyi et al., 1989).

### **Determination of Antimicrobial Activity**

Clinical isolates of *Salmonella typhi*, *Escherichia coli*, *Protus (Mirabilis)*, *Staphylococcus aureus*, ( $\alpha$ -HS) *Alpha hemolytic streptococcus* and *Klebsiella pneumonia* were isolated in the microbiology laboratory of Federal Medical Centre, Yola.

### **Media Preparation**

7g of the nutrients agar in 250ml of distilled water was dissolved into the conical flask, stopped the mouth of the conical flask with cotton wool, and wrapped it with aluminum foil and then autoclave for 15 minutes 121<sup>o</sup>C.

### **Pouring of Media**

Petri dishes were displayed on the laboratory bench the molten media prepared were dispensed aseptically and then flamed and allowed to solidify.

### **Antibacterial activity**

The media prepared were inoculated with the different test organisms and it is spread using sterile bent glass rod, and allowed to dry for 15 minutes. Thereafter, various holes were made on it, and then 0.5ml of the various extracts was pipette into various holes made on the media, then allowed the diffusion of the extracts for 15 minutes. The inoculated plates

where incubated at 37°C for 24 hours. After 24 hours of incubation, diameters of zones of inhibition of various extract against the different (isolates) test organisms were measured in millimeters.

### Scoring and Reading

The results were taken by considering the zone of growth and inhibition of the organisms by the test fraction (Mackie and McCartney, 1989).

### RESULTS AND DISCUSSION

The phytochemical analysis of the ethanol and water extract from the roots, stem bark and leaves of the indigenous plant (*Ficus exasperata*) is shown in Table 1 and 2 respectively. Table 3 and 4 shows the antimicrobial efficacy of the water and ethanol fractions respectively from the extracts against micro-organisms.

**Table 1:** Phytochemical analysis of ethanol extracts from roots, stem bark and leaves of *Ficus exasperata*.

Phytochemical components	Roots extract	Stem-bark extract	Leaves extract
Tannins	+	+	+
Flavonoids	+	+	+
Saponins	+	+	+
Glycosides	+	+	+
Alkaloids	-	-	-
Steroids	+	+	+
Phenols	+	-	+

+ = Present; - = Absent

**Table 2:** Phytochemical analysis of water extracts from roots, stem bark and leaves of *Ficus exasperata*.

Phytochemicals component	Roots extract	Stem bark extract	Leaves extract
Tannins	+	+	+
Flavonoids	+	+	-
Saponins	+	+	+
Glycosides	+	+	+
Alkaloids	-	-	-
Steroids	+	+	+
Phenols	+	-	+

+ = Present; - = Absent

**Table 3:** Antimicrobial efficacy of water extracts from roots, stem bark and leaves of *Ficus exasperata* (zones of inhibition in mm)

Test organisms	Water extracts			Ofloxacin (control)
	Roots	Stem-bark	Leaves	
<i>Salmonella typhi</i>	10	15	R	30
<i>Escherichia coli</i>	R	23	21	31
<i>Staphylococcus aureus</i>	12	21	19	21
<i>Protus (Mirabilis)</i>	R	18	25	30
<i>Alpha haemolytic streptococcus (αHS)</i>	25	28	28	R
<i>Klebsiella pneumonia</i>	21	25	18	31

R = Resistance

**Table 4:** Antimicrobial efficacy of ethanol extracts from roots, stem-bark and leaves of *Ficus exasperata* (zones of inhibition in mm)

Test organisms	Water extracts			Ofloxacin (control)
	Roots	Stem-bark	Leaves	
<i>Salmonella typhi</i>	R	25	R	30
<i>Escherichia coli</i>	R	26	R	31
<i>Staphylococcus aureus</i>	R	R	R	21
<i>Protus (Mirabilis)</i>	R	28	17	30
<i>Alpha haemolytic streptococcus (αHS)</i>	15	17	R	R
<i>Klebsiella pneumonia</i>	R	22	26	31

R = Resistance

The preliminary phytochemical tests carried out on the powdered roots, stem-bark and leaves extracts revealed that *ficus exasperata* contain tannins, flavonoids, saponins, glycosides and steroids, with no traces of alkaloids. Phenols are present in root and leaves extract but absent in stem bark extracts (Table 1). Table 2 shows presence of saponins, tannins, steroids and glycosides, with no trace of alkaloids. Flavonoid is present in root and stem bark but absent in leaves and phenols is absent in stem bark extract but present in root and leaf extracts in the tested plants part in an indication that the plant is of pharmacological importance. The presence of flavonoids indicates the natural occurring phenolic compound, with beneficial effects in the human diet as antioxidants and neutralizing free radicals. Table 4 showed the antimicrobial activities of the ethanolic extract of the root stem-bark and leaves. The stem-bark ethanolic extract was active against *Salmonella typhi*, *Escherichia coli*, *Staphylococcus aureus*, *Protus mirabilis*, *Alpha haemolytic Streptococcus* and *klebsiella pneumonia* and the ethanolic extract from the leaves was only active on *Protus mirabilis* and *klebsiella pneumonia*. While ethanolic extract from the root inhibited the growth of only *Alpha haemolytic streptococcus (α-HS)*.

Table 3 showed the antimicrobial efficacy of the water extract of roots, stem-bark and leaves. The stem-bark of the water extract was active against all the tested microorganisms. The leaf of the water extract was also active for all the tested microorganisms except for *Salmonella typhi* which showed resistance while the root of the water extract also showed resistance to *Escherichia coli* and *Protus mirabilis*. The activity of the extracts against *Salmonella typhi* and *Escherichia coli*, which are potential causative agents of abdominal ailment agreed with previous work (Sonibare et al., 2006), reported that young leaves of *Ficus exasperata* used as remedy for some abdominal ailments. The decoction of the leave has been established to exhibit significant reduction in intestinal motility in addition to its anti-ulcer activity with no sign of toxicity (Gamaniel et al., 1970). In this work, the extracts of the plant's parts (roots, stem-bark and leaves) inhibited the growth of *Escherichia coli* to a high degree. These bacteria are responsible for various stomach related illnesses. *Salmonella typhi* is causative organism of typhoid fever, a systemic infection associated with the consumption of contaminated food (Adebayo and Ishola, 2009b), while *Escherichia coli* is responsible for a number of food and water related illnesses that manifest themselves in the form of diarrhea (Adams and Moss, 1999).

Water extracts (roots, stem- bark and leaves) were all active on the organism compared with the standard (Ofloxacin). The standard used (Ofloxacin) is active on all the micro-organisms tested both in water and ethanol extracts except, *Alpha haemolytic streptococcus* (α-HS). The inhibition of micro-organisms by the extracts has justified their use for the treatment of cough, typhoid, ulcer and malaria in the traditional medicine. These analyses suggest that the ethanol extracts of the different parts of the plant of the indigenous plant contain active agents and could be promising candidates for drugs development and validate their tribal claims as a cure for some human ailments. This assertion is also confirmed as their extracts indicates a relatively moderate number of phytochemicals. Clinical and toxicity studies should be carried out on the extract to ascertain it safety when used.

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