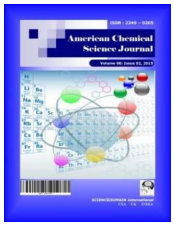

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Phytochemical Screening and Antimicrobial Activity of *Ficus sycomorus* Extracts of the Stem Bark and Leaves on Some Pathogenic Microorganisms

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Authors' contributions

This work was carried out in collaboration between all authors. Author ATM designed the study. Author ASA performed the statistical analysis. Author KG wrote the protocol and the first draft of the manuscript. Author AD managed the analyses of the study. Author HI managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

The preliminary of Phytochemical screening of methanol and ethanol extracts for both the stem-bark and leaves of *Ficus sycomorous* revealed the presence of flavonoids, glycosides, reducing sugar, resins, tannins and saponins. The result of anti-microbial activity indicates that ethanol and methanol extracts of both stem-bark and leaves show a remarkable activity on gram positive bacteria of *Staphylococcus aureus* more than the gram negative bacteria of *Salmonella typhi*. Also the extraction shows that the stem bark and leaves extract of ethanol yield 29·86% and 40.07% respectively in ethanol compared to methanol extract which yield 13.93% and 14.72% for the stem bark and leaves extract respectively.

Keywords: Flavonoids; Saponins and alkaloid; bacteria.

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1. INTRODUCTION

Ficus sycomorous (Linn) belong to the family moraceae. The plant is known for many Vernacular names such as *Baure* in Hausa, *Opoto* in Yaruba, *Ba'are* in Fulbe, subula in Arabic, *Gular* in hindi and in English was known as wild fig, strangler fig, Sycamore, sycamore fig, bush fig, common cluster fig. And also in French, it was known as *figuier sycomore*, *Sykomore.* Likewise in Spanish it was known as *Sicomoro* and in *Swahili* was known *Mukuyuchivuzi* [1]. The plant is widely distributed in Africa, South of the Sahel, North of the tropic of Capricorn excluding the central west and southern Arabian. The plant is the commonest tree that grows in savannah and high water table areas, often found along water courses such as streams and rivers, swamps and water holes [2]. *Ficus sycomorous* grows to 20 m tall and 6 m wide. They possess a heart shaped leaves with a round apex of about 14 cm long and 10 cm wide. They are always born on the leaf axis as shown in Fig. 1. The stem-bark color is yellow to orange. And on strips it reveal the yellow inner bark like all other grapes, it contains a latex as shown in Fig. 2. [3].

Fig. 1. Leaves of *Ficus sycomorus*

Fig. 2. Stem bark of *Ficus sycomorus*

In coastal areas of Nigeria, *Ficus sycomorus* are used for the treatment of some diseases like diarrhea, vomiting and mental illness. Also the stem bark of this plant is found to be a pain reliever [4]. The stem bark of this plant had been reported to be used against diarrhea, dysentery and wound infections. It was therefore imperative to screen the said part of the plant against some pathogenic organisms responsible for such diseases [5]. In Tanzania, especially in the rural areas, the leaves of the plant are used in the treatment of snakes bite, jaundice and also they are used as latex to effect for chest diseases, cold and dysentery. The stem barks of plant are used for the remedies treatment of cough, throat injection and chest pains [6].

2. MATERIALS AND METHODS

2.1 Collection and Preparation of Plant Material

The fresh leaves and stem-barks of the medicinal plant of *F. sycomorus* L. were obtained from Gwale area of Kano state located along latitude 11℃58'N and longitude of 8℃30'E in the northern region of Nigeria. The plants were identified by taxonomical study at Botanical Science Department of Bayero University, Nigeria. The stem-barks were cut into smaller pieces, the leaves were shade dried for a week and then grinded by special electric mill. The Samples were used to carry out the extraction and which were later used for further analysis [7].

2.1.1 Extraction of plant material

The methanolic and ethanolic plant extracts of 500 g were dried and crushed. Furthermore, the plant material was subjected to extraction in a soxhlet apparatus and the solvent of the extracted mixture were subjected to evaporation. The dried extracts were taken for further analysis [8].

2.1.2 Preparation of sensitivity disc

Paper discs were made from whatman No.1 filter paper using a paper puncher. 50 discs each was placed in three screw-capped bottles and sterilized by autoclaved at 121℃ for about 15 minuted as demonstrated [8]. The bottles were then removed and allowed to cool at room temperature [8].

2.1.3 Preparation of sub culture

To the volumetric flask, 2 g of nutrient agar was dissolved in 60 cm^3 of distilled water and then autoclaved at 121℃ for about 15 minutes. It was removed and then allowed to cool at room

temperature. The media was poured into plates (petriedishes) which was allowed to cool and solidify. The plates were inoculated singly with the organisms which are *Staphylococcus aureus*, *Salmonella typhi*. Incubation was carried out at 37℃ for 24 hours as demonstrated by [8].

2.1.4 Preparation of solution/serial dilution

The stock solution were prepared by dissolving 0.002 g of each methanolic and ethanolic extracts of stem-bark and leaves of *Ficus sycomorus* in 2 cm^3 of DMSO to obtain concentrations of 1000 µg/cm^3. Two different concentrations were prepared from the stock solution of 500 µg/cm^3 and 250 µg/cm^3. These were obtained by mixing stock solution (0.5 cm^3) with 0.5 cm^3 DMSO that is 0.5cm^3 DMSO was subsequently added to the stock after removal of 0.5 cm^3. The solutions were introduced singly into each bottle containing 50 discs and allowed to stay for some time at room temperature to ensure maximum absorption of solution by the discs [8].

2.1.5 Preparation of inoculums

The standardized inoculums of the bacterial isolates were swabbed onto the surface of nutrient agar in separate petridishes. This was followed by placing the prepared discs of the methanolic and ethanolic extracts of stem-bark and leaves of *Ficus sycomorus* and standard tetracycline discs onto the surface of inoculated media. The plates were incubated at 37℃ for 18 to 24 hours after which zones of growth inhibition of each sample was observed [8].

3. RESULTS AND DISCUSSION

The extracts fraction of the leave and stem-bark of *Ficus sycomorus* as presented in Table 1, which shows that ethanol solvent extract from stem bark and leaves produces a high yield of 29.86% and 40.07% compared to methanol extract from the stem bark and leaves which yield 13.93% and 14.72% respectively. Which relates to the polarity of the solvents used and this can be seen as demonstrated by Lamba et al. [9]. It can also be inferred from Table 1, that Distilled water is the most polarized solvent with a lower yield of 4.46% and 4.01% from the stem bark and leaves extract respectively. This trend is associated with the polarity of the solvents extract, the higher the polarity of solvent the lower the yield and vice versa. The Phytochemical analysis as shown in Table 2, which reveals the presence of; flavonoids, saponins, alkaloids, reducing sugars and glycosides which are the secondary metabolites from the stem-bark extracts of *Ficus Sycomorus*. Flavonoids and glycosides have been found to stimulate β-cells, increase insulin secretion or possess an insulin like effect as demonstrated by Oumar et al. [10]. The methanol and ethanol extract from the stem-barks and leaves were further subjected to antimicrobial activity test. The plant extracts were tested against two bacteria isolate; *Salmonella typhi* and *Staphylococcus aureus*. The antimicrobial screening results of various fractions in different solvents from the leaves and stem bark of *F. sycomorus* were shown in Table 3 and Table 4 respectively. The ethanol stem-bark extracts were found to have a high remarkable activity on gram positive bacteria isolate *Staphylococcus aureus* with a maximum bactericidal inhibition zones of 15.0±0.71 mm in diameter at 250 µg/ml and a minimum inhibition zones of 9.5±0.5 mm in diameter at 500 µg/ml respectively as shown in Table 3. Also the gram negative bacteria isolate of *Salmonella typhi* yield a high bactericidal activity with a maximum inhibition zones of 8.5±0.31 mm in diameter at 250 µg/ml and minimum inhibition zones of 7.0±0.81 mm in diameter at 1000 µg/ml respectively as shown in Table 3. Likewise the ethanolic leaves extract show a similar trend of inhibition on *Staphylococcus aureus* with a maximum inhibition zones of 11±0.51 mm in diameter at 250 µg/ml and minimum inhibition zones of 7.0±0.41mm in diameter at 1000 µg/ml respectively. Whereas the *Salmonella typhi* show a minimum inhibition zones of 7.5±0.71 mm in diameter at 1000 µg/ml and a maximum inhibition zone of 10±0.20 mm in diameter at 250 µg/ml respectively on the ethanolic leaves extract as shown in Table 3. Similar fluctuation trend of inhibition zone was reported by Kunjal Bhatt et al. [11] and Uma et al. [12]. This may be due to the fact that at higher concentrations, the rate of diffusion may perhaps be varied and hence, it might not be available to react with the microorganisms. The methanol stem-bark extracts show a high remarkable sensitivity on gram positive bacteria isolate of *Staphylococcus aureus* with a maximum bactericidal inhibition zones of 11.0±0.50 mm in diameter at 250 µg/ml and a minimum inhibition zones of 8.0±0.95 mm in diameter at 500 µg/ml respectively as shown in Table 4. Also the gram negative bacteria isolate of *Salmonella typhi* yield a high bactericidal activity with a maximum inhibition

zones of 10.0±0.50 mm in diameter at 500 µg/ml and minimum inhibition zones of 9.25±0.45 mm in diameter at 250 µg/ml respectively as shown in Table 4. Also the methanolic leaves extract show a similar trends of inhibition on *Staphylococcus aureus* with a maximum inhibition zones of 9±0.20 mm in diameter at 500 µg/ml and minimum inhibition zones of 7.8±0.20 mm in diameter at 1000 µg/ml respectively. Whereas the *Salmonella typi* show a minimum inhibition zones of 7.5±0.71 mm in diameter at 1000 µg/ml and a maximum inhibition zones of 10±0.20 mm in diameter at 250 µg/ml respectively on the methanolic leaves extract as shown in Table 4. While the gram negative bacteria isolates of *Salmonella typhi* only exhibit maximum inhibition zones of 7.0±0.4 mm in diameter at 500 µg/ml whereas there is insensitivity effect at 250 µg/ml and 1000 µg/ml concentrations respectively.

Table 1. Percentage yield of the extracts fraction of *Ficus sycomorus* in different the extracts

Solvents	Weight(g) SBE	Weight(g) LE	Weight(%) SBE	Weight(%) LE
Ethanol	7.50	6.80	29.86	40.07
n-hexane	2.31	2.10	9.20	12.38
Petroleum ether	2.00	1.88	7.96	11.08
Chloroform	1.89	2.00	7.52	6.54
Ethylacetate	6.80	1.90	27.07	11.20
Methanol	3.50	2.50	13.93	14.72
Distilled water	1.12	0.68	4.46	4.01

Where; SBE; Stem-bark extract and LE; Leaves extract

Table 2. Phytochemical screening result for stem bark of *ficus sycomorus extracts*

Solvent extract	Reducing sugar	Alkaloid Dragendorff's	Alkaloid Meyer's	Saponins	Tannins	Steroid	Flavanoid
Ethanol	+	+	+	+	+	_	+
Methanol	+	+	+	+	+	_	+
Aqueous	_	+	_	+	_	_	+
n-hexane	+	+	_	+	+	_	+

Where + = present and − = absent

Table 3. Inhibitory activity of ethanol extract of the stem bark and leaves *F. sycomorus* against two bacterial Isolates

Bacteria	Extract 250 µg/ml	Extract 500 µg/ml	Extract 1000 µg/ml	Tetracycline 250 µg/ml
Stem–bark				
Staphylococcus aureus	15±0.71	9·5±0.50	10·0±0.20	16.00±0.80
Salmonella typhi	8·5±0.31	8·0±0.95	7·0±0.50	12·25±0.40
Leaves				
Staphylococcus aureus	11±0.50	8·0±0.95	7·5±0.40	12·25±0.40
Salmonella typhi	10±0.20	8·5±0.50	7·5±0.71	14·00±0.25

Values are mean inhibition zone (mm) ±SD of four replicates

Table 4. Inhibitory activity of methanol extract of the stem bark and leaves extract of *F. sycomorus* against two bacterial Isolates

Bacteria isolate	Extract 250 µg/ml	Extract 500 µg/ml	Extract 1000 µg/ml	Tetracycline 250 µg/ml
Stem bark				
Staphylococcus aureus	11±0.50	8.0±0.95	9.50±0.40	12.25±0.40
Salmonella typhi	9.25±0.43	10.0±0.50	9.75±0.10	14.0±0.40
Leaves				
Staphylococcus aureus	8.50±0.40	9.0±0.20	7.8 0±0.20	15.24±0.83
Salmonella typhi	_	7.0±0.40	_	12.30±0.40

Values are mean inhibition zone (mm)±SD of four replicates

4. CONCLUSION AND RECOMMENDATION

The ethanolic and methanol extracts from the stem bark and leaves shows a remarkable antimicrobial activity against the test organisms. It was also found that the ethanol extract yield a high amount of the extract compared to the methanol extract. And also the extracts revealed the presence of flavanoids, steroids, tannins, saponins, alkaloids and reducing sugar. Flavonoids and glycosides have been found to stimulate β-cells, increase insulin secretion or possess an insulin like effect Further study should be done on the plant extracts by testing on the isolates of the multi resistant microorganisms. Lastly there is need of to isolation more chemical component presence in the plant.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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