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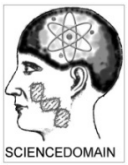

# Active Phytochemicals and Antimicrobial Properties of the Extracts of *Xylopia aethiopica* Root and Stem Bark

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***Authors' contributions***

*This work was carried out in collaboration between all authors. Author GAF designed the study, supervised the work and acted as corresponding author. Author OAO managed the literature searches, analysis of the study and compiled the manuscript for publication. Author UKO performed the antimicrobial analysis reported in this work. Author AE carried out the experimental process and also helped in literature searches. All authors read and approved the final manuscript.*

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## ABSTRACT

The use of plants to prevent and cure diseases traditionally has been a common practice all over the world even with no scientific evidence of why they have been used. *Xylopia aethiopica* is one of such plant known to be a medicinal plant of great repute in West Africa in the treatment of various diseases. In this study, phytochemical constituent and antimicrobial activities of the stem bark and root extracts of *Xylopia aethiopica* were investigated. Results obtained showed the presence of some phytochemical compounds known to be biologically active and therefore aid the antimicrobial potency of the plant material. The antimicrobial inhibition of the crude extracts of these plant parts compare quite well with the commercial antibiotic drugs used as standard reference. The results suggest that the pure form of the bioactive compound (s) responsible for this activity may prove to be a better antimicrobial drug.

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## 1. INTRODUCTION

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 [1]. Furthermore, an increasing reliance on the use of medicinal plants in the industrialised societies 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 [2]. Moreover, in this part of the world, herbal remedies have become more popular in the treatment of minor ailments, on account of the increasing cost of personal health maintenance. The study of plants used in traditional medicine requires the effective integration of information of their chemical composition, pharmacological activities of isolated compounds, as well as indigenous knowledge of traditional healers [3].

*Xylopia aethiopica* commonly known as Ethiopian/Guinea pepper, Spice tree, or Negro pepper is a common plant in Africa especially West and Central Africa with wide applications in traditional medicine in the management of several diseases. Widely distributed in Ghana, Democratic Republic of Congo, Ethiopia, Kenya, Mozambique, Nigeria, Senegal, Tanzania and Uganda where it is commonly used as condiment [4]. The plant has attracted several investigations thus revealing several documented activities. Investigations revealed that the fruit of *Xylopia aethiopica* is used locally for treatment against cough, stomach ache, dizziness, amenorrhoea, bronchitis (when smoked and inhaled), dysentery, enema, bulimia (eating disorder), lumbago and neuralgia [5]. There is the need therefore for scientific research into its bioactivity since its traditional usage may not necessarily carry with it any scientific evidence.

This study is aimed at examining the phytochemical components of *Xylopia aethiopica* with the objective of evaluating the antimicrobial effects of the bioactive compounds.

## 2. MATERIALS AND METHODS

### 2.1 Collection and Identification of Plant Samples

Fresh samples of *Xylopia aethiopica* (root and stem bark) were collected from Iyara Ijumu Local Government Area of Kogi State, Nigeria and authenticated at the herbarium of the Biological Science Department, University of Abuja, Nigeria. The samples were washed and air dried then pulverized into fine powders which were subsequently used for chemical analysis.

### 2.2 Preparation of Extracts

Cold maceration method was adopted for the extraction of plant materials. The pulverized plant parts (stem and root) were separately placed in cylindrical jars and macerated with methanol for 48 hours. The resulting extract after filtration was then left to evaporate to dryness on a hot water bath and concentrated to obtain semi-solid masses for both root and stem bark, which were subsequently stored in sterile bijoux bottles under refrigeration conditions at 4℃ [6].

Partitioning of the aqueous solution of the crude methanol extract using separating funnel was carried out with equal volume of n-hexane, chloroform and n-butanol solvents. Each extract was evaporated over a water bath, stored in separate sterile bottles and refrigerated for further analysis.

### 2.3 Phytochemical Analysis

The three partitioned (n-hexane, chloroform and n-butanol) extracts of *Xylopia aethiopica* (root and stem bark) were analyzed for the presence of tannins, phenols, volatile oils, alkaloids, saponins, flavonoids, terpenoids, resin, phlobatannins, and reducing sugar as described by various researchers [7,8,9] with little modifications.

### 2.4 Antimicrobial Analysis of the Extracts

The bioactivity of *Xylopia aethiopica* plant extracts were tested using well-agar diffusion method as described by Cheesbrough [10]. Four standard bacteria strains (*Staphylococcus aureus, Escherichia coli, Klebsiella pneumonia, and Pseudomonas aeruginosa*) and a fungus (*Candida albican)* were obtained from the Microbiology laboratory of University of Abuja Teaching Hospital, Gwagwalada, Abuja. The bacteria and fungus strains were activated and cloned three successive times in CLED agar and nutrient agar respectively, and they were stored in nutrient slants at 4℃. Subsequent activation and test were done on nutrient agar medium.

Six different dilutions of each of the extracts were made using their respective solvent (n-hexane, chloroform and n-butanol). Different weight of the extract ranging from 100 mg to 600 mg were weighed into six different test tubes and 1 ml of the corresponding solvents of each extracts (n-hexane, chloroform and n-butanol) were added into the test tubes and allowed to mix thoroughly. These gave six different concentrations of the extracts from 100 mg/ml to 600 mg/ml. The bacteria and yeast colonies on the nutrient agar were streaked to cover the entire surface of a new nutrient containing six wells and 0.1 ml each of the resulting mixture was transferred into each of the 6 well and incubated again for 24 hours at 37℃. Zone diameter of inhibition of growths were measured and recorded to the nearest millimeter. Standard antibiotic drugs (septrin, gentamycin and tetracycline) were used as positive control to provide and evaluate the degree of inhibition of the extracts.

## 3. RESULTS AND DISCUSSION

The results obtained for the phytochemical screening carried out on *Xylopia aethiopica* are as presented in Table 1.

The n-Hexane and n-Butanol extracts of the stem and root Extracts of *Xylopia aethiopica* showed no antimicrobial activity against the test microorganism. Only the chloroform extracts showed inhibition against some of the isolates.

Result of the phytochemical analysis carried out on the three extracts of the stem bark and root of *Xylopia aethiopica* is as shown in Table 1. Results from the stem bark analysis showed the presence of tannins, phenol, volatile oils, alkaloids, saponins, flavonoids, resins and phlobatannins in at least one of the three solvents used for the extraction but all extracts showed negative test to reducing sugar. Also, the root extracts of the *Xylopia aethiopica* showed positive test to phenol, alkaloids, saponins, flavonoids and terpenoids in at least one of the three solvents used for the extraction but showed negative test to resin, reducing sugar and phlobatannins in all the solvents used. The absence of reducing sugars from both plant parts can be due to ecological and geographical factors, age of plants, and method of extraction or extracting solvents or probably not a secondary metabolite in the plant parts.

These compounds are known to be biologically active and their presence in the plant parts can be said to be responsible for the antimicrobial activities of the plant material. These bioactive components are known to be bactericidal, pesticidal and fungicidal in nature [11]. For example, the anti-inflammatory, anti-allergic and antimicrobial actions of flavonoids are well documented and account for the use of decoctions of several plants materials in the treatment of various diseases in humans. Cytotoxic and carcinogenic effects have also been reported [12]. Also, Saponins have been reported as possible anti-carcinogens whose reaction mechanisms include direct cytotoxicity, immune-modulatory effects, bile-acid binding and normalization of carcinogen-induced cell proliferation [13]. Thus these results establish that *Xylopia aethiopica* has potential as a source of important bioactive molecules for the treatment of microbial infections.

The result for the antimicrobial activities of the chloroform extract against 4 clinical isolates: *Escherichia coli, Staphylocollus aureus, Pseudomonas aeruginosa and Klebsiella pneumonia* and yeast isolate, *Candida albicans*, for the stem bark and root extracts are as shown in Table 2 and Table 3 respectively while that of the positive control drugs are as shown in Table 4. The n-hexane and n-butanol extracts of

**Table 1. Phytochemical screening of the stem bark and root extracts of *Xylopia ethiopica***

| Phytochemicals | Stem | | | Root | | |
|---|---|---|---|---|---|---|
| | n-Hexane | Chloroform | n-Butanol | n-Hexane | Chloroform | n-Butanol |
| Tannins | - | + | + | - | - | - |
| Phenols | - | - | + | + | + | - |
| Volatile oils | + | + | + | - | - | - |
| Alkaloids | + | + | + | - | + | - |
| Saponins | + | - | + | + | + | + |
| Flavonoids | + | + | - | - | - | + |
| Terpenoids | - | - | - | + | - | - |
| Resins | - | - | + | - | - | - |
| Phlobatannins | - | + | + | - | - | - |
| Reducing sugar | - | - | - | - | - | - |

*Key: +=Present, - = Not present*

**Table 2. Zone of inhibition (mm) of the antimicrobial activities of the chloroform extract of stem *Xylopia aethiopica***

| Isolates | Concentration (mg/ml) | | | | | |
|---|---|---|---|---|---|---|
| | 100 | 200 | 300 | 400 | 500 | 600 |
| *S. aureus* | 0 | 0 | 0 | 0 | 0 | 0 |
| *E. coli* | 0 | 0 | 0 | 0 | 0 | 0 |
| *K. pneumonia* | 3 | 5 | 8 | 8 | 10 | 10 |
| *P. aeruginosa* | 10 | 13 | 14 | 14 | 16 | 17 |
| *Candida albican* | 0 | 4 | 5 | 5 | 8 | 8 |

**Table 3. Zone of inhibition (mm) of the antimicrobial activities of the chloroform extract of root *Xylopia aethiopica***

| Isolates | Concentration (mg/ml) | | | | | |
|---|---|---|---|---|---|---|
| | 100 | 200 | 300 | 400 | 500 | 600 |
| *S. aureus* | 0 | 0 | 0 | 0 | 0 | 0 |
| *E. coli* | 2 | 6 | 2 | 2 | 7 | 8 |
| *K. pneumonia* | 0 | 0 | 3 | 3 | 5 | 5 |
| *P. aeruginosa* | 3 | 5 | 10 | 15 | 16 | 16 |
| *Candida albican* | 4 | 6 | 8 | 14 | 15 | 16 |

*Keys: E. coli- Escherichia coli, S. aureus - Stapylococcus aureus, K. pneumonia – Klebsiella pneumonia, P. aeruginosa - Pseudomonas aeruginosa, C. albican - Candida albican*

**Table 4. Zone of inhibition (mm) of the antimicrobial activities of the control drugs against test isolates**

| Isolates | Tetracycline (40 mg/ml) | Gentamycine (20 mg/ml) | Septrin (100 mg/ml) |
|---|---|---|---|
| *S. aureus* | 10 | 14 | 0 |
| *E. coli* | 30 | 25 | 18 |
| *K. pneumonia* | 35 | 20 | 14 |
| *P. aeruginosa* | 30 | 27 | 15 |
| *Candida albican* | 0 | 0 | 0 |

the stem and root did not show antimicrobial activity against the selected test microorganisms. High activities of the stem extract were observed against *Pseudomonia aeruginosa* followed by activites against *Klebsiella pneumonia and Candida albican.* No any activities were observed against *E. coli* and *S. aureus*. The chloroform extracts of the root of *Xylopia aethiopica* showed antibacterial activity and antifungal activity against *Pseudomonia aeruginosa, E. coli, Klebsiella pneumonia* and *Candida albican* but did not show any activity against the growth of *Staphylococcus aureus.* The observed variation in sensitivity pattern of the tested clinical isolates used may be related to the genetic diversity among the organisms. Tetracycline, Gentamycine, and septrin were used as positive controls and yielded zones of inhibition higher than all the extracts.

Based on this study, the chloroform extracts of the stem and root of *Xylopia aethiopica* were found to have significant antimicrobial and antifungal activity. This current study showed that the chloroform extracts of the stem and root of *Xylopia aethiopica* possess antimicrobial effect against the growth of pure isolates of *Pseudomonia aeruginosa, E. coli, Klebsiella pneumonia* and *Candida albican*.

## 4. CONCLUSION

The demonstration of the antimicrobial activity of *Xylopia aethiopica* in this study has therefore provided scientific basis for its use in ethnomedicine/traditional medicine as a local health remedy for the treatment of diseases such as cough, rheumatism, infections, stomach ache, calmative etc. This preliminary study has emerged a good foundation required for further on-going research on the active component in *Xylopia aethiopica* plant to culminate in appropriate collaborations for achievement of dream drugs.

## CONSENT

It is not applicable.

## ETHICAL APPROVAL

It is not applicable.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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