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# Antimicrobial activities of crude methanolic extract of *Bauhinia rufescens* (Caesalpiniaceae) against some pathogenic microorganisms

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**Abstract**
The aerial part of the plant – *Bauhinia rufescens* (Caesalpiniaceae) which has been implicated for various traditional medicinal applications was studied. The plant extract was screened for its antimicrobial activities and for secondary metabolites. Phytochemical analysis of the crude methanol extract revealed the presence of saponins, tannins, and anthraquinones. Alkaloids, flavonoids and cardiac glycosides were not detected. The extract was active against pathogenic fungi-*Trichophyton tonsurans*, *Aspergillus flavus* and *cadida albicans*. A concentration dependent antibacterial activity was observed against *Staphylococcus aureus*, *Bacillus cereus*, *Proteus mirabilis*, *Pseudomonas aeruginosa* and *Klebsiella pneumoneae*. However, the extract was inactive against *Salmonella parathypie A*. The minimum inhibitory concentration (MIC) of the plant extract against *Staphylococcus aureus*, *Bacillus cereus*, *Pseudomonas aeruginosa* and *Proteus mirabilis* were obtained between 12.5 mg /ml – 25.0 mg/ml. The results obtained in this study have provided evidence and justification for the ethno medical use of the plants in the treatment of microbial diseases.

**Keywords:** *Bauhinia rufescens*, antifungal and antibacterial activity, leprosy

## Introduction
Over the last decade, interest in drugs of plant origin and their uses in various diseases management have increased in many developed countries since plants used in traditional medicine are more likely to yield pharmacologically active compounds than developing new drugs synthetically (WHO, 1956). Interestingly, a significant percentage of the world population still rely on folkloric remedies for various disease conditions.
*Bauhinia rufescens* is a shrub (Synonymous) *Pilostigma rufescens* (Lam); *Adenolabus rufescens* (Lam); *Bauhinia adansoniana* (Guill & Perrott), belonging to the family Caesalpiniaceae and tribe Carcideae. *Bauhinia rufescens* is commonly referred to as matsattsagi by the Hausas in Kano, Zaria and Sokoto; It is kwon as Nammmarahi in Fulani; Kharroub in Sudan and Kulkul in Shuwa Arab (Dalziel, 1956) [3]. It is a much branched small tree reaching 5-8m in height, with greenish-yellow to white flowers. It is evergreen and under ordinary Sahel condition it behaves like deciduous species. The plant is well distributed from Benin to Cameroon, Chad, Ghana, Ivory Coast, Mauritania, Niger, Nigeria and Senegal. The infusion or the decoction of the root bark is drunk for the cure of fever and dysentery (Gill, 1992) [5]. It is also used to tan native leather and as a medicine for diarrheal. People of Barkin Ladi in plateau state use the infusion of the bark and the aerial part of the plant including the seed in the treatment of leprosy (Babalola, 2006) [1]. There is no available scientific literature on *Bauhinia rufescens* to support the ethno medical use of the plant. However, many members of the Caesalpiniaceous family have been investigated and reported to possess remarkable pharmacological activity. *Piliostigma thonningii* (Schum) is a tropical plant used in the treatment of wounds, chronic ulcers, diarrheal, toothache, cough and bronchitis. The aqueous ethanolic extract was reported to possess wide spectrum antimicrobial activity. The extract also significantly inhibit prostaglandin synthetase enzyme test for anti - inflammatory activity (Mbewuike *et al.*, 1997) [7]. The antibacterial and anti- inflammatory activities of the plant was attributed to the flavonoid content (C – Mehtyl flavanols: 6,8 – di – C – Methyl quercetin 3 – methylether, quercetin,6-C-methylquercetin 3-methylether, 6-8 – di – C-methyl Kaemferol 3, 7 dimethyl ether and 6,8 – di C-methyl kaemferol 3,7 dimethyl ether. These flavonoids have also been reported to posesse anti-allergic and anti-cancer activity (Code *et al*., 1988) [2]. This study reports on the phytochemical, antifungal and antibacterial activity of methanolic extract of aerial part of *Bauhinia rufescens*.

## Materials and methods
Plant Material. Aerial part of the plant was collected in a bush in Barkin Ladi, Plateau state in November, 2005. The material was air-dried and ground into coarse form. The plant was duly authenticated by Dr. I. Abdulkarim, Federal College of Forestry, Jos, where voucher specimen has been deposited.

### Extraction
The crushed plant material (60g) was extracted with methanol using the Soxhl et extractor. The extract was filtered, concentrated and kept in the refrigerator for use.

### Phytochemical screening
The crude methanol extract was screened for the presence of alkaloids, tannins, glycosides, steroid/ triterpenes, flavonoids,

saponins, and anthraquinones using standard methods (Harborne, 1973; Trease and Evans, 1959) [6, 9].

**Determination of antimicrobial activity**

**The following test organisms were used in this study**

**Bacterial:** *Staphylococcus aureus*; *Proteus mirabilis*; *Klebsiella pneumonia; Salmonella paratyphie A*; *Bacillus cereus*; *Pseudomonas aeruginosa*. The fungi were *Trycophyton tonsurans*; *Aspergillus flavus* and *Candida alibicans*. The bacteria and fungi were clinical isolates obtained from School of Medical Laboratory Technology, NVRI, Vom and Department of Dermatophilosis, NVRI, Vom. The strains were maintained and tested on nutrient agar (bacteria) and sabroud dextrose agar (fungi). The antimicrobial activity test was carried out using Agar well diffusion and dilution techniques and MIC's by tube dilution method (Elizabeth, 2001; Russel & Furr, 1997) [4, 8]. In the agar well diffusion method, plates of nutrient agar seeded with the various bacterial cultures and plates of sabroud dextrose agar seeded with the fungus were used, the agar thickness was about 10mm in a 90mm plate.

**Media**

Sabroud Dextrose Agar (SDA, product of Oxoid Ltd, Basmagstoke Hampshire England, ($p^H$ = 5.6 ± 0.2), Nutrient Agar (Product of i.d.g., Topley House, 52 Wsahlane, B196-UK, $P^H$=7.3±0.2) were used for testing the antifungal activity. Nutrient broth (BHI, $P^H$ = 7.4) was used for the overnight culture of the bateria.

**Result & discussion**

The result of antimycotic activity of the crude methanol extract of *Bauhinia rufescens* as presented in Table 1, shows moderate activity against *Candida albicans*. Table 2 shows significant antifungal activity of the plant extract against *Trichophyton tonsurans* and *Aspergillus flavus. The activity of the plant extract on these pathogenic fungi increased with increase in concentration.* The antibacterial activity of the plant extract as shown in Table 3 indicates that the extract significantly inhibited the growth of *Staphylococcus aureus*, *Bacillus cereus*, *Pseudomonas aeruginosa* but inactive against *Salmonella paratyphie A*. The result of phytochemical screening revealed the presence of anthraquinones, saponins and tannins but alkaloids were not detected. The minimum inhibitory concentrate (MIC) of the plant extract against *Staphylococcus aureus*, *Proteus mirabilis*, *Bacillus cereus*, *Pseudomonas aeruginosa* were obtained between 12.5mg/ml-25.0mg/ml. The antibacterial activity of the plant extract were more pronounced on the gram positive bacteria and moderately active against gram negative bacteria. It is noteworthy the fact that the plant extract was active against *Pseudomonas aeruginosa* which is known for its obstinate resistance to most antimicrobial drugs including Streptomycin used in this study.

**Table 1:** Result of Antimycoti of Activity of Methanol Extract of Bauhinia *rufescens on Candida albicans*

| Extract concentration | Population of organinism growth description | | | | control |
|---|---|---|---|---|---|
| | 1CF | ½CFU | ¼ CFU | ⅛CGU | |
| 200 mg/ml | TG | MG | MG | MG | DG |
| 100 mg/ml | DG | DG | DG | DG | DG |
| 50 mg/ml | DG | DG | DG | DG | DG |
| 25 mg/ml | DG | DG | DG | DG | DG |

**Key:** CFU – Colony forming unit = 1 x $10^6$; Control solvent (methanol); TG- Moderate growth; DG- Dense growth.

**Table 2:** Result of Antifungal Activity of Crude methanol Extract of *Bauhininia rufescens*

| *Fungi* | Growth Diameter(mm) Concentration of Crude Extract Incorporated | | | | | |
|---|---|---|---|---|---|---|
| | 200mg/ml | 150mg/ml | 100mg/ml | 50mg/m | +ve Control | -Ve Control |
| *A. Flavus* | 15.0 | 16.0 | 20.0 | 31.0 | 11.5 | 85.0 |
| *T. Tonsurans* | 14.0 | 14.0 | 35.0 | 49.0 | 11.0 | 80.0 |

**Key:** Nizoral – (+ve Control) 2.5µg/ml; Negative Control – Methanol.

**Table 3:** Result of Antibacterial Activity of Crude Methanol Extract of *Bauhinia rufescens*

| *Bacteria* | *Zones of Inhibition (mm) Extract Dillution (mg/ml)* | | | | +ve Control | -Ve Control | MIC *(mg/ml)* |
|---|---|---|---|---|---|---|---|
| | 25mg/ml | 50mg/ml | 100mg/ml | 200mg/ml | | | |
| *S. aureus (G + ve)* | 12.0 | 14.0 | 15.0 | 17.0 | 22.0 | NI | 12.5 |
| *B. cereus (G + ve)* | 13.0 | 14.0 | 16.0 | 18.0 | 32.0 | NI | 25.0 |
| *P. aeruginosa(G -ve)* | 10.0 | 11.0 | 12.0 | 13.0 | NI | NI | 25.0 |
| *K. pneumonia (G-ve)* | 7.0 | 7.0 | 7.0 | 7.0 | 20.0 | NI | NI |
| *S. parathpie A (G-ve)* | NI | NI | NI | NI | 25.0 | NI | NI |
| *P. mirabilis (G-ve)* | 7.0 | 11.0 | 12.0 | 14.0 | 11.0 | NI | 12 |

**Key:** +ve Control = Streptomycin, 2.5mg/ml; -ve Control = Solvent (methonal); NI-ND inhibition; ND-Not determined; MIC=Minimum inhibition Concentration.

**Table 4:** Result of Phytochemical Analysis of Methanol Exract of *Bauhinia rufescens*

| *Alkaloid* | Saponins | Tannins | Flavonoids | Anthraquinones | Cardiac glycosides |
|---|---|---|---|---|---|
| - | +++ | +++ | - | ++ | - |

**Key:** +++ - appreciable; + + - moderate amount; + - trace amount; - - not detected.

**Conclusion**

The plant *Bauhinia rufescens* possesses interesting antimicrobial active against common fungi and bacteria. The study has provided scientific justification for the ethno medical use of the plant in the treatment of dysentery, diarrheal and syphilis. The findings in this study call for a more detailed investigation of *Bauhinia rufescens* to harness

in its potential as a bio-resource for active therapeutic agents for effective management of leprosy and other microbial infections.

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