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## Biochemical and Antimicrobial Activity of *Prosopis africana*

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### Abstract

Biochemical properties and antimicrobial activities of *Prosopis africana* were examined in the study. Acetone, ethanol and hexane were the solvents used to extract the phytochemicals. Standard methods were utilised in the extraction of these plant chemicals throughout. Alkaloids, tannins, terpenoids, flavanoids, glycosides, steroids and phenols were detected in varying concentrations depending on the solvent used. Zones of inhibition were determined for the three organisms tested for *S. aureus*, *S. typhi* and *E. coli* with the ethanolic extract showing the highest zone of inhibition.

**Keywords:** *Prosopis africana*, Biochemical, Inhibition, Antimicrobial, Metabolites.

### 1. Introduction

Many herbs are moving from alternatives to main stream use with a good number of the people seeking remedies and health approaches free from the side effects caused by synthetic chemicals (Adebayo, Oladele, 2012). Recently considerable attention has been paid to utilizing eco-friendly and bio friendly plant based products for the prevention and cure of different human diseases (Borokini, 2014). With the present surge of interest in phyto-therapeutics the availability of genuine plant material is becoming imperative, therefore, accurate morphological and anatomical standard of drug plant is very much essential (Adebayo, Oladele, 2012). The microscopic and macroscopic description of a medicinal plant is the first step toward establishing its identity and purity and should be carried out before any test is undertaken. Prosopis are pod bearing trees or shrubs consisting of 44 reported species which are found in arid and semi-arid regions of the world (Barku et al., 2013). *Prosopis africana* is the only tropical African prosopis species, occurring from Senegal to Ethiopia in the zone between the Sahel and savannah forests. The fact that *Prosopis africana* has found tremendous use on the local and international scene, it is a good candidate plant for research into its uses, composition and standardization as an attempt to contribute to the health and well-being of humanity (Hamad et al., 2015). There is a long and venerable history of the use of traditional medicine, Agboola stated that *Prosopis africana* is used as plant to improve dental health and promote oral hygiene (Omwirhiren et al., 2016). It is used as chewing stick by Yoruba in south western Nigeria and in vast parts of the world where tooth brushing is done with *Prosopis africana* (Rajeshwar et al., 2016). *Prosopis africana* is a perennial leguminous tree, the practice of tooth cleaning by chewing subfamily mimosidae and is mostly found growing, it's in sticks has been known since antiquity (Qadir et al., 2015). It is also widely used in the savannah regions of western Africa in many areas,

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its chewing stick persists today among many African and fermented seed are used as food condiments (Gumgumjee, Hajar, 2012). In Southern Asian communities, America and southern United States as well as isolated areas young leaves and shoots are fodder that is highly sought (Iqbal et al., 2015). In many cultures, there is this popular belief that every plant grown on the surface of the earth has medicinal properties or use. However it is impossible to provide a complete list of these uses to which must of these plants may be put (Qadir et al., 2015). Despite all the progress in synthesising chemistry and biochemistry plants are still indispensable sources of medicine in both preventive and curative ways. Hundreds of species of plants are recognized as having medicinal value, and many of them are commonly used to treat or prevent ailments and diseases. The medicinal properties of the plants are often associated barks, leaves flowers, fruits, roots or seeds (Iqbal et al., 2015). The plant belongs to the family mimosaceae, *Prosopisafricana* is a savannah tree of about 12-18m highland up to 1m in width, which may be readily distinguished by its dark rough bark, pale drooping foliage with small pointed leaf lets and sausage-shaped fruits. The flowers which usually appear around December-may are yellowish, fragrant and densely crowded in fat species about 4-6 cm long excluding the shoot stalk. *Prosopisafricana* local names kirya(hausa); Kohi(kulfulde); ayan(yaruba); sanchi(nufe); ubwa(igbo); kpaye(tiv).

## 2. Materials and Methods

### Sample Collection and Preparation

The sample was collected in the area of yunbungavillage Darazo Local Government Area of Bauchi State, Nigeria. The sample was taken to the Biology laboratory of the Department of Science Laboratory Technology, School of Science and Technology, AbubakarTatari Ali Polytechnic, Bauchi, Nigeria for authentication by the botanist. The sample was washed with the distilled water and shade dried for about a week the sample was grinded using motor and pistil to obtain it coarse particles then a sieve of diameter 0.02mm was used to sieve the coarse particles so that the fine particle are obtained.

### Extraction

50 g of the prepared sample was taking and placed into the clean and dried beaker, 200 ml hexane was added and covered with aluminium foil with polythene bark tied with masking tape, this is to prevent the escape of hexane from the sample. Then the mixture was shaken up and allowed to extract the components for 24h then the hexane extract was separated using filter paper, the filtrate was placed in a clean and dried container covered and kept in a suitable place. The residue was allowed to shade dry and reweighed to get the percentage recovery. The reweighed residue was placed in a fresh and cleaned beaker then 150 ml of acetone was added and allowed for 24h, then the acetone extract was filtered and kept. Then the residue was reweighed and shade dried to obtain the second percentage recovery. The reweighed residue was also placed in a cleaned beaker then 150 ml ethanol was added and allowed for 24 h then the ethanol extract was filtered and the filtrate was shade dried to obtain the third percentage recovery. The three (3) extracts were kept in a suitable place in the laboratory for phytochemical screening and antimicrobial activity.

### Phytochemical Screening:

Test for Flavonoids, tannins, terpenoids, cardiac glycosides, steroids and phenols were done using standard methods.

### Bioassay

The anti-bacterial susceptibility was determined by using Agar well diffusion method. The entire nutrient agar surface was scaled with the inoculums suspension and allowed to dry for 5 min. The well of 6 mm was created and 7ml of each extract was poured into it, the plates were kept in the refrigerator for about 15 min to allow for proper diffusion. The extract was then incubated at 37 °C for 24 h, at the end zone of inhibition was measured in mm. This exercise was done in triplicates to ensure reliability.

### Extract Preparation for Bio-Assay

2 ml portion of the each three (3) extracts was kept in a clean and sterile container and was been covered to prevent contamination with microbes and impurities until the time for used for Bioassay.

### Preparation of the Media

28 g of nutrient agar powder was suspended in 1 L of distilled water, the mixture was heated in order the suspensions to dissolve completely. The dissolved solution was then covered with

Aluminium foil and placed in the autoclave for sterilization. The solution was then sterilized at 121 °C for 15 min, pH7. Agar was poured in 3 sterile petri-dishes and it was allowed to cool in a refrigerator and ready for inoculation.

### Bacterial Culture

The antibacterial activity was tested against three (3) clinical isolates; it includes three (3) gram negative bacteria. *Staphylococcus aureus* (*S. aureus*), *Plasmodium species* (*P. species*) and *Salmonella typhi* (*S. typhi*), the isolates were taken from AbubakarTafawaBalewa University Teaching Hospital Laboratories and were identified microscopically and on biochemical basis. Identified isolates were stored in 20 % glycerol at 20 °C and sub-cultured on the nutrient agar at 37 °C for 24h before use.

### Inoculation and Application of Extracts.

Bacterial strains were inoculated into 15 ml of sterile nutrient broth and incubated at 37 °C for 24h. After 24h the plate was removed from the oven and 3 different extracts were introduced to the plate and left for 24h.

### Minimum Inhibitory Concentration (mic).

This lowest drug concentration prevents visible microorganism growth after overnight incubation. A plate of solid growth media, after a pure culture is isolated was examined and minimum inhibitory concentration was determined.

## 3. Results

**Table 1.** Microscopic examination of *Prosopis africana* stems bark

Test parameters	Observation
Colour	Dark gray with ash coloured patches, reddish brown
Internal colour	Reddish brown
Texture	Hard
Shape	Curved
Ordour	Weak, musty and characteristic
Taste	Characteristic
Fracture	Outer bark is short and rough in the inner bark
Thickness	4 – 7 mm
Internal surface	Longitudinally striated
Powder colour	Reddish brown

**Table 2.** Phytochemical screening of *Prosopis africana* stem bark

Extracted	Secondary Metabolite							
	Alka- loids	Flavo- noids	Tannins	Sapo- nins	Terpe- noids	Glyc- side	Steroid	Phenolic compound
Hex E	+	-	++	-	+++	++	++	+
A C E	-	-	++	-	++	+	+	+
E E	++	-	+++	++	++	++	++	++

Hex E = Hexane Extracted  
 A C E = Acetone Extracted  
 E E = Ethanol Extracted

**Table 3.** The zone of inhibition (mm) of the antibacterial activity of hexane extracted bark of *Prosopis africana*

TEST	Conc, mg/ml	Hexane
<i>Staphylococcus aureus</i>	10 <sup>-4</sup>	8.00
	10 <sup>-4</sup>	6.00
<i>Escherichia coli</i>	10 <sup>-4</sup>	3.00
	10 <sup>-4</sup>	3.00
<i>Salmonellatyphi</i>	10 <sup>-4</sup>	12.00
	10 <sup>-4</sup>	10.00

**Table 4.** The zone of inhibition (mm) of the antibacterial activity of acetone extracted bark of *Prosopis africana*

Test	Conc, mg/ml	Acetone
<i>Staphylococcus aureus</i>	10 <sup>-4</sup>	10.00
	10 <sup>-4</sup>	9.00
<i>Escherichia coli</i>	10 <sup>-4</sup>	11.00
	10 <sup>-4</sup>	11.00
<i>Selmonellatyphi</i>	10 <sup>-4</sup>	14.00
	10 <sup>-4</sup>	12.00

**Table 5.** The zone of inhibition (mm) of the antibacterial activity of ethanol extracted bark of *Prosopis africana*

Test	Conc, mg/ml	Ethanol
<i>Staphylococcus aureus</i>	10 <sup>-4</sup>	15.00
	10 <sup>-4</sup>	13.00
<i>Escherichia coli</i>	10 <sup>-4</sup>	16.00
	10 <sup>-4</sup>	15.00
<i>Salmonellatyphi</i>	10 <sup>-4</sup>	18.00
	10 <sup>-4</sup>	17.00

#### 4. Conclusion

Accuracy morphological and anatomical standardization of medicinal plants is very essential. In this study, physico-chemical evaluation of the stem bark of *Prosopis africana* were under taken. The results of this study can be used as diagnostic tool for the standardization of *Prosopis africana* and will be help full in the characterization of the fruit drug. According to the world health organization. The macroscopic and microscopic description of a medicinal plant is the first step toward establishing its identity and purity and should be carried out before any test and undertaking.

The results obtained in (Table 2) shows that the ethanol extract also played more significance role in the bacteriocidal and/or bacteriostatic activity than the acetone and hexane extract and this is because ethanol usually extracts about 90 % of the components. Tables 3, 4 and 5 depict the zones of inhibition of the three solvents showing varying levels of inhibition. The bacteriocidal and/or bacteriostatic activity for the stem bark of *Prosopis africana* is established in the study and this data could be used as a tool for the standardization of this medicinal plant and will be helpful in the characterization of the crude drug. This parameter could be useful in properties of its monograph Africa pharmacopeia. Any crude drug which is similar to the *Prosopis africana* but whose characters significantly differ from the accepted standard will then be rejected as contaminated adulterated or done right fake.

The result of the research provides parameters for the proper identification of the stem bark of *Prosopis Africana*. The crude extract shows significant antibacterial activity. The results of the Phytochemical analysis indicates the presence of active components on the stem bark of *Prosopis africana*.

## 5. Recommendations

It is recommended that

- Further toxicity studies using different microorganism;
- Sub-acute test is planned in other to determine the long-term effect of the extract;
- Isolation of other pharmacological active compound is carried out from the plant;
- Further studies to ascertain the actual bactericidal and/or bacteriostatic mechanism of the action of the plant extraction and fraction;
- Further studies on the plant crude extract antiviral antifungal and anti-inflammatory activity.

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