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The Effect of *Bridelia ferruginea* Bark Extracts on Some Pathogenic Micro-Organisms

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Abstract

The effects of extracts of *Bridelia ferruginea* on *Bacillus subtilis, Eschericia coli, Staphylococcus aureus, Aspergillus niger* and *Fusarium solani* were examined in this study. The results obtained revealed that methanol extract was the most effective on *Bacillus substilis* and *Escherichial coli* while ethanol extract was most effective on *Staphylococcus aureus*. In the fungi isolates the ethanol extracts had the highest % growth inhibition though the values obtained were considerably lower than those of the control experiments in the two fungi species.
Key words: *Bridelia ferruginea*, Bark Extracts, Pathogenic Micro-Organisms

Introduction

*Bridelia ferruginea i*s an indigenous medicinal species in Nigeria. Though present in the forest vegetation but it is commonly found in the savanna. It is usually a gnarled shrub, which sometimes reach the size of a tree when grown in a suitable environment. The bark is dark grey, rough and even markedly scaly. Its common vernacular names in Nigeria include Kirni, Kizni (Hausa), Mareni (Fulani), Iralodan (Yoruba), Ola (Igbo), Urareju (Owo).

B. ferruginea bark extract is being used for milk coagulation and also in lime juice for the formulation of traditional gargle “ogun efu” (Orafidiya *et al*., 1996). It is also used as a purgative and vermifuge (Cimanga *et al*., 1997). Iwu (1984) asserted that the plant has molluscidal activities, while Adeoye *et al* (1999) reported that the bark extract of the plant has antimicrobial activities against some micro-organisms known to cause enteric and secondary upper respiratory tract infection.

The study being reported here examined the effect of *B. ferruginea* bark extract on some

pathogenic micro-organisms which include *Aspergillus niger, Bacillus subtilis, Escherichia coli, Fusarium solani* and *Staphylococcus aureus.*

Key words: Bark extract, *Bridelia ferruginea,* pathogenic microorganisms.

Material and Methods:

Collection and Preparation of Sample:

Fresh barks of *Bridelia ferruginea* were obtained from the local herb market situated at Oja-Oba, Owo, Ondo State, Nigeria. They were air dried and the dried samples were grinded with the aid of milling machine into powder and the resulting powder kept in a clean air tight container.

Preparations of the Extracts

*Aqueous Extraction***:**

The methods of Sofowora (1986) were used in this study. A 40g portion of the powder was dispersed in 200ml of distilled water, stirred very well and left for 48 hours; thereafter the extract was filtered through sterile muslin cloth. The extract obtained was concentrated in vacuum using a rotary evaporator. Necessary precautions were taken to avoid contaminations during the experiment.

Organic Solvent Extraction:

The solvent used were ethanol, methanol and acetone. A 40g of the powder was dispersed in 200ml of each of the organic solvents in 250ml conical flask. Thereafter, the flask containing the mixtures were covered and left for 75 hours in the laboratory at room temperature. The extract was equally filtered through sterile muslin cloth and the filtrates were then concentrated in vacuum using a rotary evaporator.

Extracts Dilution

The extracts were diluted using di-methyl sulphoxide (DMSO). Aqueous extract was diluted using sterile distilled water, 1g of the extract was mixed with 10ml of DMSO and also 1g of the extract was equally mixed 19ml of DMSO.

Culture Media Preparation

The media used for the tests were nutrient agar, and potato dextrose agar. They were prepared from commercial dehydrated products and were made up or reconstituted according to the manufacturer instructions. In the nutrient agar, 2.8g of the powder was dissolved in 100ml of distilled water in a conical flask but in the potato dextrose agar, 3.5g was dissolved in 100ml of distilled water. Dissolution was achieved by stirring the mixture on a magnetic stirrer hot plate in a conical flask with a sterile glass rod until it completely dissolved. The conical flasks were cotton-plugged, sealed with aluminum foil and autoclaved at 121^0c for 15 minutes.

Inoculums Preparation

The micro-organisms used were *Bacillus subtilis, Eschericia coli, Staphylococcus aureus, Aspergillus niger* and *Fusarium solani.* Pure isolates of these micro-organisms were obtained from the laboratory of Federal Medical Centre, Owo, Ondo State, Nigeria. Broth cultures were prepared for the preservation of pure isolates and the stock culture were used for the experiment.

Determination of Antimicrobial Properties of the Crude Extract by Agar Diffusion Method

1ml of the test organisms (24 hours old culture) was aseptically injected into sterile plates. 20ml sterilized nutrient agar (NA) was poured on top of the test organism aseptically after it has been allowed to cool to about 45^0c. The medium was mixed gently for even distribution of the inoculums within the media and allowed to solidify at room temperature (25^0c). Sterile cork borer of 8mm in diameter was used to make five (5) wells on the solidified agar into which 0.5ml extracts of the sample (water and solvent extracts) were aseptically introduced into the well separately with the aid of syringe and it was labeled.

A control experiment was set up with well containing standard antibiotic, streptomycin powder at 0.2mg/ml. The plates were incubated at 37^0c for 24 hrs. Zones of inhibition were observed around each well after 24 hrs and recorded appropriately against the extract. Results were quoted as the radii (mm) of the zone of the inhibition around the well.

In the fungi isolates, 5ml of the extract was impregnated with 25ml of sterile potato dextrose agar (PDA) and allow to set at 25^oc. A sterile 8mm cork borer was used to inoculate the fungi isolate at the centre of the plate. A control experiment was also set up with PDA plate containing no extract while a standard antifungal agent (Benlate) was equally, used at 0.5mg/ml, to impregnate another control plate separately prior to inoculation. The plates were incubated at 27^oC for 72 hrs. The radical growth of the fungal isolates was recorder at every 24 hrs. The % mycelia growth inhibition was recorded at the end of the experiment, using the following relation:

$$\% \text{ growth inhibition} = \frac{\text{Mycelia growth in the control} - \text{Mycelia growth in the treated sample}}{\text{Mycelia growth in the control}} \times 100$$

Results and Discussion

Tables 1 and 2 show the effects of *Bridelia ferruginea* bark extracts on some pathogenic microorganisms, that is, *Bacillus substilis, Staphylococcus aureus, Escherichia coli, Aspergillus niger* and *Fusarium solani*. Table 1 revealed that the methanol extract was most effective than the other extracts on *Bacillus substilis* and *Escherichial coli* while ethanol extract was most effective on *Staphylococcus aureus*. In the fungi isolates the ethanol extracts had the highest % growth inhibition though the values obtained were considerably lower than those of the control experiments in the two fungi species (Table 2).

The results obtained from this study tend to suggest that extract from the bark of *Bridelia ferruginea* could be used in the treatment of food borne diseases, such as diabetes, cough, black tongue, boils and wound infections. It could also be used in the control of food spoilage organisms. It could be suggested however that further research should be conducted to further confirm the promising results demonstrated in this study.

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Table 1. Zones of Inhibition (Diameter in mm) in *Bridelia ferruginea* extracts-treated microorganisms.

Microorganisms	**Extracts**				
	Methanol	**Ethanol**	**Acetone**	**Aqueous**	**Streptomycin**

Bacillus subtilis	5.50	5.40	5.30	5.00	15.00
Staphylococcus aureus	6.00	6.50	4.30	4.00	18.00
Escherichia coli	5.30	5.25	5.20	5.18	17.00

Table 2. Growth inhibition (%) *in Bridelia ferruginea* extracts-treated microorganisms.

Microorganisms	**Extracts**				
	Methanol	**Ethanol**	**Acetone**	**Aqueous**	**Streptomycin**
Aspergillus niger	5	10	4	3	100
Fusarium solani	12	15	10	5	100