



Original Research Article

***In vitro* Antibacterial Activities of Aqueous and Ethanolic Stem Bark Extracts of *Bridelia ferruginea* Benth.**

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Abstract	Keywords
<p>The folkloric claim of <i>Bridelia ferruginea</i> Benth. for treatment of bacterial infections such as dysentery and skin infection was evaluated in this study. The plant, <i>Bridelia ferruginea</i> was collected and the bark of the plant was air-dried to constant weight under shade, pulverized into a fine powder with mortar and pestle and then both aqueous and ethanolic extracts of the stem bark of the plant were used for antibacterial study at concentrations of 100 mg/ml and 200 mg/ml respectively. Four clinical isolates including <i>Escherichia coli</i>, <i>Salmonella typhi</i>, <i>Pseudomonas aeruginosa</i> and <i>Staphylococcus aureus</i> were obtained from the Microbiology Laboratory. The isolates were further identified in the Laboratory using standard biochemical tests and then sub-cultured on Nutrient Agar plates for 18 h before use. Agar- well diffusion method was used for determining the antibacterial activity of the extract. The results showed that at the concentration of 100 mg/ml, only the growth of <i>Staphylococcus aureus</i> was inhibited by the extract and at 200 mg/ml, the growth of <i>Salmonella</i> and <i>Staphylococcus aureus</i> were inhibited by the ethanolic extract. Though the test does not suggest that the extract has superior antibacterial activities compared to the standard drug, ciprofloxacin, its fair antibacterial actions could be an encouragement for its use. It is quiet difficult to directly extrapolate <i>in vitro</i> study results being that most <i>in vitro</i> proven drug activity prove to be ineffective <i>in vivo</i>.</p>	<p>Antibacterial activity <i>Bridelia ferruginea</i> Clinical isolates Folkloric claim Stem bark extract</p>

Introduction

In the last two or three decades, public dissatisfaction with the cost of prescription medications and possibly increasing toxicity of synthetic drugs have necessitated a return to herbal medications. And it is a common sight to see herbal

vendors in the streets and market places advertising their products and much degree of patronage accorded them. It is therefore in recognition of the increased value of herbal medicine in primary healthcare, along with the challenges it faced, WHO

advocated for the proper identification, sustainable exploitation, scientific development and appropriate utilization of herbal medicines which provide safe and effective remedies in medicare (Wambebe, 1998).

Bridellia ferruginea Benth. is widely employed in the traditional management of a number of disease conditions including diarrhoea, bacterial infection such as dysentery (Gill, 1992) and as an antimicrobial (Adeoye et al., 1988). *Bridelia ferruginea* is a savannah plant and the tree is about 6-15 m in height and up to 1.5 m wide. The bark is dark grey, rough and often markedly scaly (Rashid et al., 2000) and it is the commonest *Bridelia* species of the savanna woodland occurring from Guinea and Mali to Nigeria and throughout the wooded Savannah region of Africa. Since there is a clear evidence of therapeutic benefits of herbal medicine (Barrett et al., 1999), in the present study, the antibacterial activities of aqueous and ethanolic stem bark extracts of *Bridelia ferruginea* has been investigated.

Materials and methods

Preparation of plant extract

The plant, *Bridelia ferruginea* Benth. (Family: Euphorbiaceae) was collected and the bark of the plant was peeled off and air-dried to constant weight under shade; then it was pulverized into a fine powder with mortar and pestle. Both aqueous and ethanolic extracts of the stem bark of the plant were prepared and used for antibacterial study at concentrations of 100 mg/ml and 200 mg/ml each.

Drugs /chemicals

Ethanol: Manufacturer: BDH Chemicals Limited, Poole, England.

Normal saline.

Ciprofloxacin (Siprosan): Manufacturer: Divine Essential Formulations. Km. 10, Lasu-Ojo Road, Igando, Lagos, Nigeria; Batch number: 018SN; Expiry date: 06-15.

Antibacterial study

Source of test organisms: Four clinical isolates including *Escherichia coli*, *Salmonella typhi*, *Pseudomonas aeruginosa* and *Staphylococcus*

aureus were obtained from the Microbiology Laboratory of the Usmanu Danfodiyo University Teaching Hospital, Sokoto. The isolates were further identified in the Laboratory using standard biochemical tests and then sub-cultured on Nutrient Agar plates for 18 h before use.

Method of antibacterial test: Agar-well diffusion method (Irobi et al., 1994) was used for determining the antibacterial activity of the extract. The different isolates were spread on nutrient agar in separate plates. Wells were then bored into the agar using a sterile 5mm diameter cork borer. Equal volumes of the extracts were dispensed into the wells and the plates were allowed to stand for one hour to allow pre-diffusion of the extract into the medium (Esimone et al., 1998). The plates were incubated at 37°C for 24 h and at the end of the incubation, zones of inhibition that developed were measured and compared with that produced by ciprofloxacin which was the standard antibiotic used for the test.

Statistical analysis

Analyses were done by comparing the measurement of zones of inhibition by extract (mm) with that of orthodox drug ciprofloxacin.

Results

At the concentration of 100 mg/ml, only the growth of *Staphylococcus aureus* was inhibited by the extract. The ethanolic extract produced a zone of inhibition of 12.5 mm while the aqueous extract produced a zone of inhibition of 9.5 mm.

At 200 mg/ml, the growth of *Salmonella* and *Staphylococcus aureus* were inhibited by the ethanolic extract while the aqueous extract inhibited the growth of *Escherichia coli* and *Staphylococcus aureus*. Both the ethanolic and aqueous extracts were not effective against *Pseudomonas* at both concentrations tested.

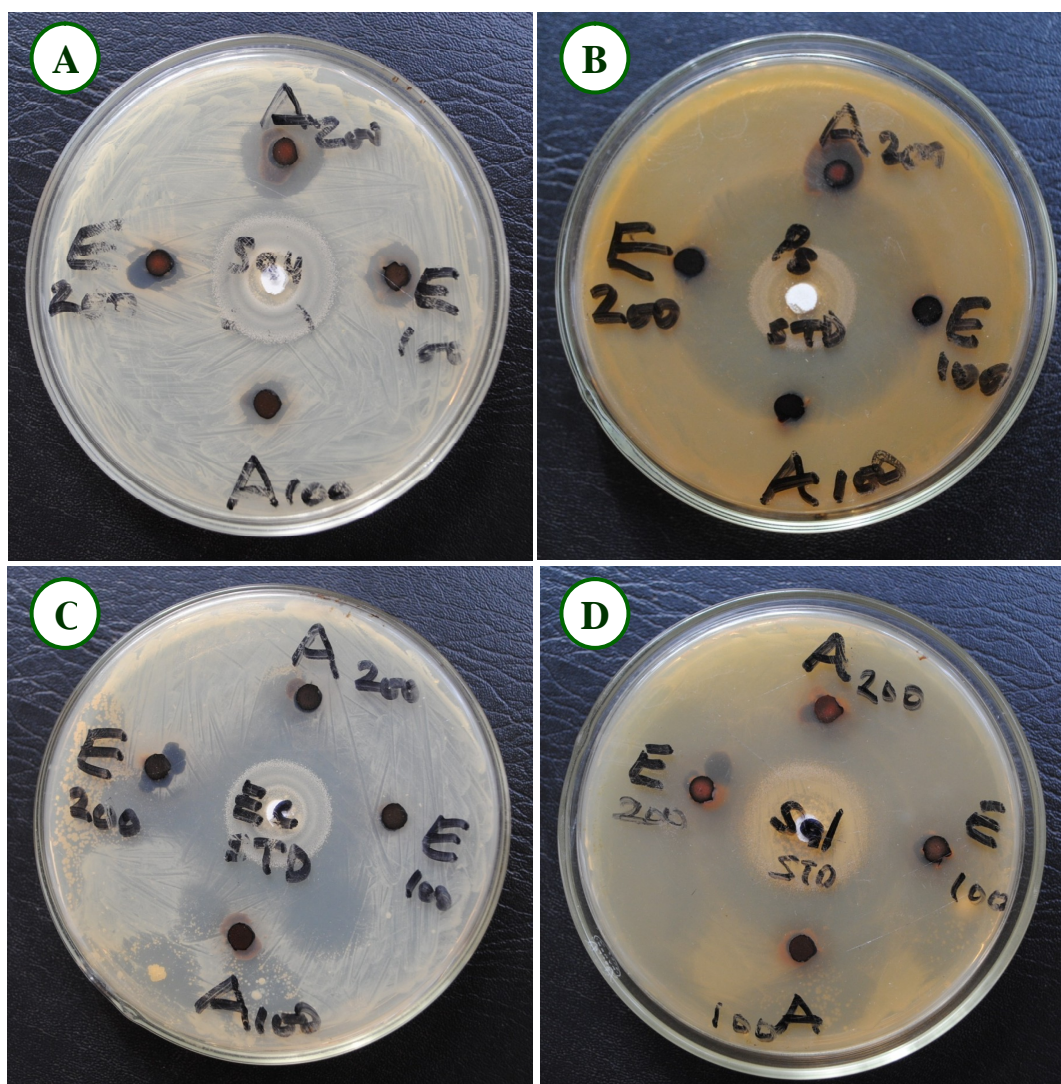
The growth of *E. coli* was inhibited by only the aqueous extract at 200 mg/ml while *Salmonella* was inhibited by only the ethanolic extract at 200 mg/ml. *Staphylococcus aureus* was inhibited by both the ethanolic and aqueous extracts at both doses tested. The standard drug, Ciprofloxacin, at a dose of 100 mg/ml inhibited the growth of all the organisms (Table 1; Fig. 1).

Table 1. Effect of *Bridelia ferruginea* extracts on selected bacterial organisms zone of inhibition in mm).

Organism	100 mg/ml		200 mg/ml		S
	E	A	E	A	
<i>Escherichia coli</i>	-	-	-	11	42
<i>Pseudomonas aeruginosa</i>	-	-	-	-	48
<i>Salmonella typhi</i>	-	-	14	-	40
<i>Staphylococcus aureus</i>	12.5	9.5	11	18	28

Key: E = Ethanolic extract; A = Aqueous extract; S = Standard Drug (Ciprofloxacin)
 Values = Zones of inhibition in mm;

Fig. 1: Effect of *Bridelia ferruginea* on selective bacteria (A) *Staphylococcus aureus*, (B) *Pseudomonas aeruginosa*, (C) *Escherichia coli* and (D) *Salmonella typhi*.



(E₁₀₀ - Ethanolic extract at 100 mg/ml; E₂₀₀ - Ethanolic extract at 200 mg/ml; A₁₀₀ - Aqueous extract at 100 mg/ml; A₂₀₀ - Aqueous extract at 200 mg/ml; STD - Standard drug)

Discussion

There has been reports of plant with antibacterial properties *Pimenta dioica* (Marjorie, 1999), and results of this study showed that the extract of *Bridelia ferruginea* demonstrated some antibacterial property against *E. coli*, *S. aureus*, *salmonella*. Among these organisms, some are implicated in the aetiology of dysentery which may further support the traditional use of *Bridelia ferruginea* for the treatment of skin diseases (De Bruyne et al., 1997) and dysentery (Gill, 1992).

Moreover, recent report by Jonathan et al. (2014) clearly demonstrated the antidiarrhoeal effects of aqueous stem bark extract of *Bridelia ferruginea* which is also in support of traditional use of the plant. Only *Pseudomonas*, known for its resistance to several antibiotics showed no response to the extract. It suffices here to mention that the antibacterial effect was quite weak on comparing with the reference orthodox antibiotic ciprofloxacin which showed very wide area of inhibition in this study.

Though the use of the plant extract in the treatment of dysentery is not superior to the treatment option offered by available orthodox drugs like ciprofloxacin the antidiarrhoeal effect of the aqueous extract proved effective (Jonathan et al., 2014) and on that note, coupled with its fair *in vitro* antibacterial activity, its use could be encouraged. It should be noted that it is quite difficult to directly extrapolate *in vitro* study results (Rothman, 2002) because most *in vitro* proven drug activity are known to be ineffective *in vivo*, because of issues associated with drug delivery, toxicity, and or other issues which are not represented in *in vitro* studies (De Clercq, 2005).

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