

Synergistic Potentials, Comparative Antibacterial Activities of *Ficus exasperata* (Vahl) Leave Extract and Indigenous Black Soap

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Introduction

In Nigeria, West Africa and most part of the world at large, plants are vital ingredients used in inherent traditional medicine. Plant leaves and stem have been described by several authors as the repertoire of bioactive secondary metabolites [1]. To a layman, these are known as phytochemicals, while a professional will call them Phenolic. Antimicrobial activities of different fractions of plant endowment have been investigated and found useful and thus justify their continuous usage in folklore medicine [2]. The indigenous black soap is native to West Africa where it is highly prized for its ability to clear pimples, blackheads, aftershave bumps thereby ensuring a smooth, supple skin [3]. The black soap compositions have been varied by fortification to make it more sweet and succinct specifically when it serves as excipients in traditional medicine [4,5]. Plant materials as well as other compounds rich in organic matter majorly serve as fortification materials. For instance, fine egg shell powder has been used as home remedy for pimples and associated skin problem [6]. Synergistically, these compounds could enhance inherent potency of the soap resulting in more effective and useful results [7]. Microbial drug resistance is a menace that is dynamic and constantly evolving. Infections caused by microorganisms especially in the tropics are characterized by severe to mild symptoms with diverse manifestations.

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ABSTRACT

Objective: To investigate its comparative antibacterial action with the indigenous black soap.

Methods: In the present study, comparative antibacterial activities of different indigenous black soaps and ethanol leave extracts of *Ficus exasperata* (vahl) on *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Klebsiella pneumoniae*, *Staphylococcus aureus* ATCC-25923, *Pseudomonas aeruginosa* ATCC-27853, *Escherichia coli* ATCC-25922, and *Klebsiella pneumoniae* ATCC-11930, were assayed by agar diffusion method.

Results: Black soaps demonstrated higher inhibitory effects on all the bacterial pathogens. Black soap with brown eggshell powder had the highest inhibition zone of 15.83 mm on *Klebsiella pneumoniae* ATCC-11930. Similar effects were also observed with *Staphylococcus aureus* ATCC-25923 and *Pseudomonas aeruginosa* ATCC-27853. *P. aeruginosa*, *E. coli* and *K. pneumoniae* were found to be sensitive to concentrations of the crude extract at 50 mg/ml and upwards. *P. aeruginosa* showed highest inhibition of 12.0 mm and 18.68 mm at concentrations to the tune of 100 mg/ml and 125 mg/ml respectively.

Conclusions: Antibacterial activities of the indigenous black soap and leaf extracts of *Ficus exasperata* (vahl) on skin pathogens have shown that *Ficus exasperata* (vahl) could be used as additive in the indigenous black soap at the considered concentrations in order to synergistically enhance its potency.

KEY WORDS:

Ficus exasperata
Black soap
Antibacterial activity
Eggshell powder

Skin infection in the tropical developing countries had always taken its toll on the children as a result of the poor social economic status of their parents. Inadvertently, this condition has been further compounded by malnutrition and drug resistance in pathogens [5]. *Ficus exasperata* (vahl) also known as the sandpaper tree, a deciduous, and dioecious species of shrubby plant native to tropical Afri-

ca, and other Arabian countries is believed to possess superficial anti-inflammatory activities [8]. It is therefore necessary to investigate its comparative antibacterial action with the indigenous black soap in order to make relevant conclusions regarding the potentials of this plant in enhancing the antimicrobial action of the black soap when used as an additive.

Materials and Methods

Collection of materials

Clinical and type isolates of *Escherichia coli*, *Klebsiella pneumoniae*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, were obtained from The University College Hospital UCH Ibadan. Necessary biochemical test were conducted to confirm their identities and were maintained at 4 °C for subsequent usage. Plain Indigenous black soaps were bought from market (A). Indigenous black soaps were also prepared by saponification and fortified separately with brown (B) and white (C) eggshell powder, effectively at 20 g/250 mL of the molten soap. Leaf of *Ficus exasperata* was collected from Teaching and Research Farm, Federal University of Technology in Akure. The identification and authentication of the plant were accomplished in the Herbarium of the Department of Plant Science, Ekiti State University Ado- Ekiti, Ekiti State. The leaves were collected dried in a container in the laboratory for a period of five weeks. The dried leaf material was pulverized with mortar and pestle. About 200g of dried leave powder was weighed into different conical flasks and labeled. Exactly 750 mL of ethanol was added into conical flask and the extraction allowed for a period of 5 days after which the mixture was filtered with muslin cloth. The liquid extract was subjected to rotary evaporation under reduced pressure. The paste extract obtained was maintained at 4°C until further analysis.

Preparation of standard inoculum

Previous culture of test bacteria in nutrient broth incubated at 37 °C for 18 h were suspended in saline solution (0.85% NaCl) and adjusted with the aid of a spectrophotometer (Unico1100RS) to match a turbidity of 0.5 McFarland standards at wavelength of 540 nm according to the method described by [9].

Determination of antibacterial activities of indigenous black soaps

Susceptibilities of the test organisms to black soaps and selected antiseptic soaps were assayed using agar-well diffusion method. A 3 g of soap was scraped with sterile blade and dissolved in 10 mL of sterile distilled water to give a stock solution this was also done for selected topical creams. 0.5 mL of the standardized suspension was seeded into Mueller Hinton agar plates and PDA plates (90 mm in diameter) using pour plate method. Three wells were made on each plate using sterile core borer (8 mm) and 0.2 mL of the black soap solutions were transferred into each of the three wells (appropriately labeled) and sterile distilled water into the fourth well as the negative control

Determination of antibacterial activities of *Ficus exasperata* (vahl)

Antibacterial activities of extracts were determined by the agar well diffusion method as described by [10]. Different concentrations of the extracts were prepared using 30% Dimethyl sulphoxide (DMSO) as the reconstituting solvent and filtered using 0.4 µm sterilized membrane pore filter paper. The control was prepared by using 0.1 mL of the reconstituting solvent and incubated alongside with the extract. The experiment was accomplished in triplicates

Results

Antibacterial activities of indigenous black soap and *Ficus exasperata* (vahl)

It was discovered that black soaps had more inhibitory effects on all the bacterial pathogens. Black soap with brown eggshell powder had the highest inhibition zone of 15.83 mm on *Klebsiella pneumoniae* ATCC-11930. Similar effects were also observed with *Staphylococcus aureus* ATCC-25923 and *Pseudomonas aeruginosa* ATCC-27853. Black soap with white eggshell powder relatively possesses the lowest antibacterial activities. Generally, type isolates were more susceptible than their clinical counterparts. For the plant, *P. aeruginosa*, *E. coli* and *K. pneumoniae* were found to be sensitive to concentrations of the crude extract at 50 mg/mL and upwards. *P. aeruginosa* showed highest inhibition of 12.0 mm and 18.68 mm at concentrations to the tune of 100 mg/mL and 125

Table 1. Comparative antibacterial activities of indigenous black soaps and selected commercial antiseptic soaps at 60mg/mL

Mean inhibition Zone (mm)								
Organisms	Source	Black soap A	Black soap B	Black soap C	Soap 1	Soap 2	Soap 3	Control
* <i>S. aureus</i>		10.83±0.28 ^c	13.63±0.55 ^d	10.90±0.17 ^c				0.00±0.00
<i>S. aureus</i>	ATCC-25923	11.83±0.29 ^e	15.00±0.00 ^f	10.90±0.17 ^d	5.01±0.02 ^b	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00
* <i>E. coli</i>		11.00±0.03 ^c	12.00±0.00 ^c	11.90±0.17 ^c				0.00±0.00
<i>E. coli</i>	ATCC-25922	13.83±0.29 ^f	12.83±0.28 ^e	11.90±0.17 ^d	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00
* <i>P. aeruginosa</i>		11.98±0.29 ^c	13.67±0.28 ^d	9.83±0.29 ^b				0.00±0.00
<i>P. aeruginosa</i>	ATCC-27853	12.83±0.29 ^f	15.17±0.28 ^f	9.83±0.28 ^c	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00
* <i>K. pneumoniae</i>		8.83±0.29 ^e	9.83±0.29 ^f	8.67±0.29 ^e				0.00±0.00
<i>K. pneumoniae</i>	ATCC-11930	13.83±0.29 ^f	15.83±0.29 ^f	8.67±0.28 ^d	10.00±0.00 ^f	2.98±0.32 ^b	10.00±0.00 ^f	0.00±0.00

Values represent means ± standard deviation of triplicate readings. Superscripts of the same letter in a row are not significantly different at P≤0.05. Key: Black soap A-Plain black soap, Black soap B-Black soap with brown eggshell powder, Black soap C-Black soap with white eggshell powder, Soap 1-3-Commercial antiseptic soaps, * Clinical isolate.

Table 2. Antibacterial activities of *Fiscus exasperata*.

Concentra- tions (mg/mL)	* <i>P.aeruginos</i> <i>a</i>	<i>P.aeruginosa</i> ATCC 27853	* <i>E. coli</i>	<i>E. coli</i> ATCC 25922	* <i>K.pneumoniae</i>	<i>K.pneumoniae</i> ATCC 11930	* <i>S. aureus</i>	<i>S. aureus</i> ATCC 25923
6.25	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a
12.5	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a
25.0	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a	0.00±0.00 ^a
50.0	1.67±1.15 ^b		1.33±0.58 ^b	5.00±0.00 ^c	0.00±0.00 ^a	2.33±0.58 ^b	0.00±0.00 ^a	5.00±0.00 ^c
		2.50±0.12 ^b						
75.0	6.83±0.29 ^c	6.83	2.90±1.73 ^c	4.50±0.12 ^c	1.00±0.00 ^b	5.67±0.58 ^c	1.00±0.00 ^b	8.50±0.58 ^e
		±0.29 ^c						
100.0	12.00±0.00 ^d	11.00±0.00	6.80±0.35 ^d	8.55±0.58 ^e	2.00±0.00 ^c	6.00±0.00 ^d	3.00±0.00 ^c	10.20±0.05 ^d
		^d						
125.0	18.68±0.58 ^e	18.50	8.00±0.00 ^e	10.00±0.00 ^d	3.93±0.12 ^d	8.33±0.58 ^e	3.33±0.58 ^d	10.20
		±0.58 ^e						±0.05 ^d

Values with the same alphabet along the column are not significantly different P<0.05 * Clinical isolate.

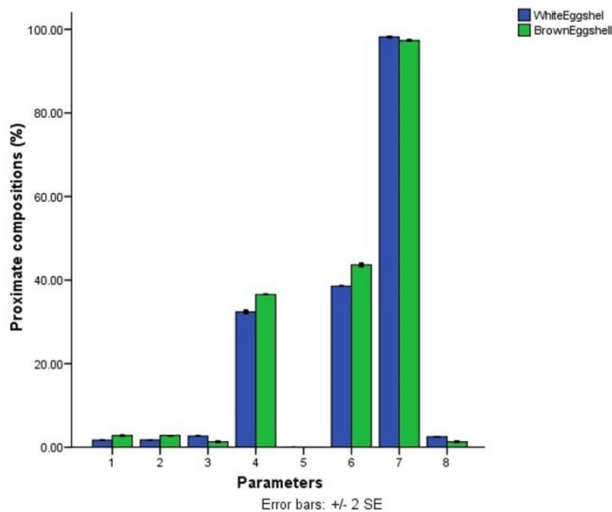
mg/mL respectively. *S. aureus* however, demonstrated mild sensitivity at 75mg/mL concentration giving rise to zones of inhibitions of 1.0 mm; 0.33 mm and 1.0 mm respectively but showed better susceptibility to the crude extract at higher concentrations. Type cultures were also relatively more susceptible to the plant extract.

Proximate compositions of the eggshell powder

Organic matter is the most prominent proximate content of the eggshell samples with an average range of 97.20 to 98.29% and closely followed by carbohydrate with a range of 43.41 to 38.53%. The eggshells were low in crude fat content, calculated fatty acid and crude fibre

content. The total ash of the brown eggshell was higher than the white eggshell (Figure 1).

Figure 2. Proximate compositions (%) of eggshell powder.



1=Moisture Content; 2 = Total Ash; 3 = Crude Fat; 4 = Crude Protein; 5 = Crude Fibre; 6 = Available Carbohydrate; 7 = Organic Matter; 8 = Fatty Acid

Discussion

Antibacterial activities of the indigenous black soaps and *Ficus exasperata* leave extracts have shown that they contain biologically active molecule or molecules that can alter biochemical process and are hence of therapeutic importance. The similar effects of the soaps and plant on *Pseudomonas aeruginosa* and *Staphylococcus aureus* have further lent credence to their therapeutic succinctness as surface active agent. *P. aeruginosa* is a Gram negative bacterial that is capable of survival in conditions of little or no specific substrate; this can only mean that the organism easily metabolize any available substrate as enabled by its inherent genetic endowment allowing it to grow and spawn in extreme conditions which ordinarily would not have supported it [11]. This has however led to increased virulence and enhanced drug resistance. Antibacterial activities of the black soap against these pathogens when compared with the commercial antiseptic soaps could be as a result of their novel constituents. The observed higher resistance in the so called commercial antiseptic soaps could be predicated on their substandard active constituents, bad manufacturing practices and excessive profit quest by manufacturers as opined by [12]. Response of the test bacterial to *Ficus exasperata* (vahl) leave extract was noticeable from 50mg/mL and above. This specifically

means that antibacterial action will be dose based implying the active ingredients increase as the dosage increases. This is beneficial in the sense that it will not only enable us determine the effective plant concentrations on using it to fortify the black soap; it also as well indicates applicable safety red flags during its clinical appraisal. Antimicrobial activities of plants have been generally linked to their compositions of several different secondary metabolites that act in synergy [13]. Since the previous submission of [2], [14] had stated that the indigenous black soap contains chemicals of plant origin; there is therefore a likelihood of synergy when *Ficus exasperata* is used as black soap additive. [7] had posited that antimicrobial potency of the black soap on bacterial is due to its complex novel constituents which can be further enhanced by plant additives. The past submission of [6], that eggshell may contain biologically active compounds agree with the results from proximate analysis of the eggshell powders used in this research. Eggshells are good sources of organic matter which can make them to be suitable for other purposes such as in feed formulations for animals. This is also in line with the submission of [15]. Other differences observed in the proximate composition could be because the local fowl is a good scavenger that can survive on little or no readymade food when compared with that of artificial breed with constant access to controlled feed. Antibacterial activities of the indigenous black soap and leave extract of *Ficus exasperata* (vahl) on skin pathogens such as *Pseudomonas aeruginosa*, *Staphylococcus aureus* and other transiently borne skin pathogens have shown that *Ficus exasperata* (vahl) could be used as additive in the indigenous black soap at the considered concentrations in order to synergistically enhance its potency.

Conflict of Interest

We declare that we have no conflict of interest.

Acknowledgements

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