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Research Article

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Phytochemistry and Antimicrobial Studies of African Black Soap and its Modified Samples

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ABSTRACT

African black soap was prepared from palm kernel oil and the filtrate of cocoa pod ash. This was divided into five portions with different natural beauty enhancing organic compounds added to four portions. These samples were analyzed chemically by pH determinations, Infrared spectra analyses and phytochemical screenings. They were also screened for in vitro antibacterial activities against two Gram-positive bacteria (Staphylococcus aureus and Bacillus subtilis) and two Gram-negative bacteria (Pseudomonas aeruginosa and Escherichia coli). Their pH values were between 8.90 and 9.78. Infrared spectra analyses of the black soap revealed strong bands due to υ (C=O) frequency of the keto group at 1668 and 1560 cm^{-1} and a strong band at 1379 cm^{-1} due to υ (C-O) frequency of the ester oxygen. The spectra of the modified samples showed no complexation via these oxygen donor atoms. Phytochemical screenings revealed the presence of saponins, flavonoids and terpenoids in the black soap. Modifying black soap by adding aleo vera, camwood, lime, honey or shea butter eliminated one or two of its phytochemical components, but even facilitated its activities against some of the tested Gram-negative and Gram-positive bacteria.

Keywords: African black soap; Phytochemical; Antibacterial

INTRODUCTION

Black soap, or African black soap, also known as Alata Samina or Alata originated from West Africa. It has been used for centuries in Ghana. Its methods of preparation and secrets have been passed down from generation to generation to keep the soap close to Mother Nature and avoid exploitation and imitation [1]. African black soap can be prepared from plantain skin and it is a natural source of vitamin A and E as well as iron [2]. The plantain peels are dried and heated using a constant temperature in order to achieve a particular color, texture and fragrance. Normally the roasted plantain skin ash is mixed with hot palm oil or palm kernel oil to form the soap. The roasting of the plantain skin determines the color of the soap. The longer the plantain skins are roasted, the darker the soap. In some recipes, Cocoa Pod is used instead of Plantain skin. Cocoa pod is the shell of the cocoa fruit and it also has natural healing properties [3]. This soap making process is very delicate, and if it is not done properly with the right ingredients, there will be no soap [3]. Some artisans who make black soap have deviated from the original African recipe by replacing plantain ashes with more accessible cocoa ashes, as well as adding essential oils, herbs or dried flowers [4]. The absence of lye makes African black soap much softer, and almost putty- like when wet, with a crumbly and uneven surface. Real African black soap varies from brown to gray [5]. African black soap is known for its ability to deeply cleanse the skin's pores, remove blemishes and makeup, and help minimize razor bumps that result from shaving [1]. This report serves as the first on the physicochemical analyses, phytochemical analyses and antimicrobial studies of Cocoa pod prepared African Black soap, as well as that of its modified samples with some skin-nourishing natural products which are aleo vera, camwood, lime, honey and shea butter.

EXPERIMENTAL SECTION

Materials and Methodology

Chemical

African Black soap was prepared with little or no modifications according to literature [6]. Prepared palm kernel oil got from palm kernel seeds and cocoa pods were locally sourced for at a town called Ifeodan, Osun State, Nigeria. All other chemicals and solvents were purchased as analytical grades from Sigma-Aldrich and SAARChem.

Instrumentation

The Infrared spectra were recorded as KBR plates on a Nicolet Avatar FTIR 330 spectrophotometer. A pH meter was used for determining the pH.

Preparation of the black soap

This was prepared with little or no modifications as previously reported [6]. The prepared black soap was left for about three days in the mold in order to solidify. The finished product was soft and really malleable. When the soap was set, it was divided into five portions. Different naturally occurring beauty enhancing organic compounds were added as additives into four of the soap portions in separate labeled containers, followed by pounding manually with a pestle and mortar. Sample 1 contained 12.50 g of the black soap without any additive. Sample 2 contained half table spoon of squeezed aloe vera pounded with 12.50 g of the black soap; Sample 3 contained 0.7 g of cam wood pounded with 12.50 g of the black soap; Sample 4 contained 7 drops of lime pounded with 12.50 g of the black soap and Sample 5 had half table spoon of honey mixed with 0.7 g of Shea butter pounded with 12.50 g of the black soap.

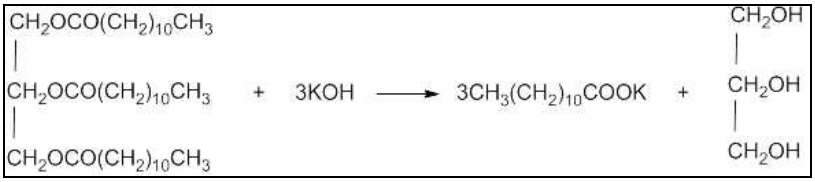
Biological

Antimicrobial screening:

Sample preparation and media used: 15.2 g of agar Mueller Hinton agar was measured into a 500 ml conical flask, 400 ml of distilled water was added (shake to dissolve) and the conical flask was covered with cotton wool and wrapped with aluminum foil paper and labeled. It was autoclaved at a temperature of 121^{o}C for 15 minutes. The agar was allowed to cool to a temperature of 45°C and aseptically poured into sterile Petri dishes. The dishes were allowed to solidify with the test bacteria. 0.5 g of the prepared samples was weighed into Mac Cartney bottles and 2.5 ml of distilled water was measured into the bottle and allowed to dissolve. Sterile filter paper disc (7 mm in diameter) was then placed inside the dissolved soap mixture and allowed to soak for 10 minutes. The plates were then inoculated with the test bacteria using the spread plate method with a sterile swab stick. The test bacteria were *Staphylococus aureus, Escherichia coli, Bacilus sp, and Pseudomonas aeruginosa.* The inoculated plates were kept on the work bench for 1hour before further work was carried out. The soaked discs were picked using forceps sterilized by flaming and placed aseptically on the inoculated Petri dishes. The discs were allowed to stick on to the surface of the agar medium before incubation. The plates were incubated at 37°C for 24 hours and antibacterial activity of the soap was measured as diameter of zones of inhibition surrounding the impregnated filter paper discs. Zones of inhibition were measured in mm [7,8].

RESULTS AND DISCUSSION

According to Dunn, 2010, the reaction for the preparation of African black soap is presented below as Equation 1 (Figure 1).



Lauric Acid Black Soap (BL) Glycerol

Equation 1: Preparation of African Black Soap (BL)

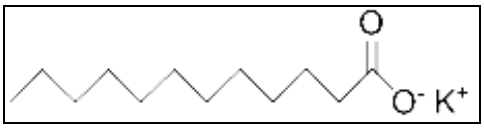


Figure 1: Structure of Black Soap (BL) [6]

pH Determinations

Table 1 shows the pH values of all the samples. Sample 1 containing black soap without any additive was the least basic with the pH value of 8.90. Sample 2 made of black soap with aloe vera had a pH value of 9.14; Sample 3 made of black soap with camwood had a value of 9.10; Sample 4 made of black soap with lime had a value of 9.73 and Sample 5 made of black soap, honey and shea butter had a pH value of 9.78.

Phytochemical Screening Analysis

Table 2 presents the results of the phytochemical screenings of the soap samples. Saponins, flavonoids and terpenoids were found present in Sample 1. Only terpenoids were found in Samples 2 and 3, while flavonoids and terpenoids were also found in Samples 4 and 5.

Table 1: pH values of the black soap samples

Samples	Additives	pH values
1	Black Soap without additives	8.9
2	Aloe Vera	9.14
3	Cam wood	9.1
4	Lime	9.73
5	Honey and shea butter	9.78

Note: pH of normal water used = 7.4

Table 2: Results of phytochemical screenings of the black soap samples

Samples	Tanins	Saponins	Flavoniods	Steriods	Terpeniods
1	-	+	+	-	+
2	-	-	-	-	+
3	-	-	-	-	+
4	-	-	+	-	+
5	-	-	+	-	+

Note: + = Present, - = Absent

Table 3: FTIR spectra analyses of the metal complexes and black soap

Compounds	υ (OH) (cm^{-1})	υ (C-H) (cm^{-1})	υ (C=O) (cm^{-1})	υ (C-O) (cm^{-1})	Others
$C_{11}H_{23}COO^-K^+$ (BL; Sample 1)	3345 bs	2918 s	1668 s	1379 s	1416 s
		2851 s	1560 s		
		2957 sh			
Sample 2	3404 bm	2918 s	1653 s	1379 s	1418 s
		2851 s	1562s		
		2957 sh			
Sample 3	3312 bs	2918 s	1668 s	1379 s	1418 s
		2851 s	1562 s		
Sample 4	3341 bs	2918 s	1665 s	1381 m	1416 s
		2851 s	1562 s		
Sample 5	3387 bs	2918 s	1653 s	1379 sh	1416 s
		2851 s	1562 s		

Note: s - strong, m - medium, b - broad, bs - broad strong, bm - broad medium, sh - shoulder

Infrared Spectra Analysis

The FTIR spectra analyses of the prepared black soap (BL) and its modified samples have been presented as Table 3. The characteristic vibrational frequencies have been identified by comparing the spectra of the modified samples with that of black soap [6]. There are two potential donor sites in black soap. These are the keto-oxygen and the ester oxygen. The infrared spectrum of the prepared black soap showed a broad and strong band at 3345 cm^{-1} attributed to the stretching vibration of υ (OH) due to hydrogen bonding. This band appeared as a broad either medium or strong band having undergone a shift to higher frequencies of 3404 cm^{-1} and 3387 cm^{-1} in Samples 2 and 5, while it has also undergone a shift to lower frequencies of 3312 and 3341 cm^{-1} in Samples 3 and 4 due to the

presence of water molecules. The strong bands appearing at 1668 and 1560 cm^{-1} in the spectrum of black soap were attributed to the υ (C=O) frequency of the keto group. These bands have undergone a shift to a lower and higher frequencies of 1653 and 1562 cm^{-1} in Samples 2 and 5. In Samples 3 and 4, they have appeared at either 1668 or 1665 cm^{-1} and 1562 cm^{-1}. These do not signify any form of chelation involving the oxygen of the keto group, as expected for all the samples. In addition it also signifies that the oxygen of the keto group might only be involved in the formation of new bonds in Samples 2 and 5. The strong band at 1379 cm^{-1} has been attributed to υ (C-O) frequency of the ester oxygen [9]. This band has remained unchanged in all the samples except in Sample 4, where it has appeared as a medium band at 1381 cm^{-1}. This also signifies no involvement of the oxygen of the ester group in chelation [10] or the formation of new bonds. This is also consistent with expectations (Figures 2-6).

Infrared Spectra

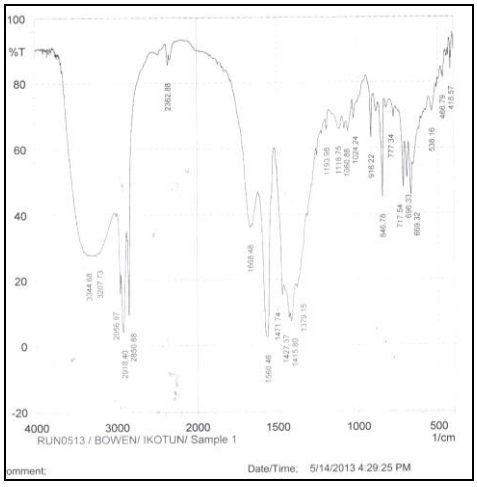


Figure 2: Infrared spectrum of black soap sample 1

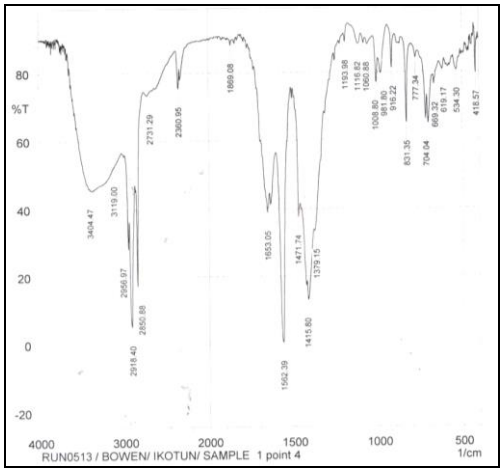


Figure 3: Infrared spectrum of sample 2

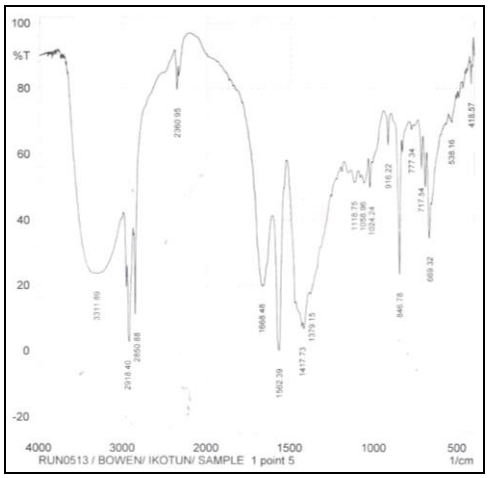


Figure 4: Infrared spectrum of sample 3

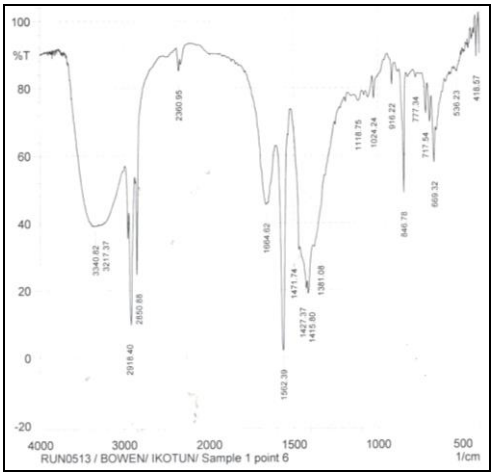


Figure 5: Infrared spectrum of sample 4

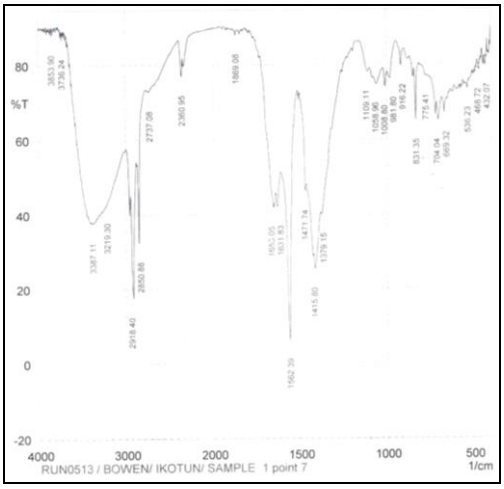


Figure 6: Infrared spectrum of sample 5

Antimicrobial Analysis

The results of the antimicrobial activities of these various soap samples have been presented as Table 4. In consistence with literature [11], black soap (Sample 1) was not active against any of the tested microorganisms.

Sample 2, which included aloe vera added to the black soap was active against the two tested gram-negative bacteria *Pseudomonas aeruginosa* and *Escherichia coli*. Sample 3, which is black soap with cam wood, was active against the Gram-positive bacterium, *Bacillus subtilis* and the Gram-negative bacterium, *Escherichia coli.* Sample 4 containing lime as an additive to black soap was active against the Gram-negative bacterium, *Escherichia coli* and Sample 5 containing honey and shea butter was also inactive against all tested microorganisms.

Table 4: Antimicrobial activities of black soap samples

Micro Organism	*Staphylococcus aureus* (mm)	*Bacillius subtilis (mm)*	*Pseudomonas aeruginosa* (mm)	*Escherichia-coli*. (mm)
Sample 1	-	-	-	-
Sample 2	-	-	12	10.5
Sample 3	-	8.5	-	9
Sample 4	-	-	-	10.5
Sample 5	-	-	-	-

Note: Diameter of the filter paper used = 7mm

Sample 1 =black soap without additives
Sample 2= black soap and Aloe vera
Sample 3= black soap and cam wood
Sample 4= black soap with lime
Sample 5= black soap with honey + shea butter

CONCLUSION

Nowadays skin changes are due to prolonged exposure to sunlight, types of cosmetics and soaps applied on skin with ozone depletion and air pollution. All these may also cause the skin to age. Thus the skin requires regular cleaning with soaps such as black soap which possess a deep cleansing ability. Results of the phytochemical screenings of the prepared black soap showed that it also possesses some secondary metabolites: saponins, flavonoids and terpenoids, which actually enhance its body nourishing abilities. This research has also shown that modifying black soap with some natural organic compounds like aleo vera, camwood, lime, honey and shea butter did not necessarily destroy its phytochemical components, but even facilitated its activities against some of the tested Gram-negative and Gram-positive bacteria. Therefore the addition of naturally occurring organic compounds to black soap will not only improve its deep cleansing ability, skin moisturizing effect, but also facilitate the ability to destroy some of the microorganisms which the skin encounters on daily basis.

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