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Comparative analysis on the antibacterial activity of some conventional medicated soap

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Abstract

The comparative study of the antibacterial activity of eight selected medicated soaps was evaluated for their antibacterial activity against bacteria of significance in wound infections and normal skin flora. This study was carried out to compare the antibacterial effect of the soaps and also validate the reported usefulness of the medicated soaps. Antibacterial activity test was carried out using agar plate diffusion method. Clinical isolates used include *Proteus mirabilis*, *Providencia stuartii* and *Pseudomonas aeruginosa* while isolates from normal skin flora used include *Staphylococcus aureus, Proteus mirabilis, Enterobacter kobei, Enterobacter xiangfangensis, Proteus hauseri*. All the soaps were found to possess varying antimicrobial activity in a concentration and organism dependent manner. Two and three out of the eight samples showed no excellent antimicrobial; activity against the skin and wound isolates respectively. The study showed that the tested soaps possessed antibacterial properties and they can be useful in the treatment and management of skin (wound) infections caused by bacteria if well prepared with the appropriate plant materials to target specific causative organisms. However prolonged usage of antibacterial soaps should be discouraged as this could result in the emergence of drug resistant bacteria.

Keywords: Antibacterial Activity; Medicated Soaps; Bacteria; Skin Infections

1. Introduction

Soaps are cleansing products which are a combination of fat, oil (animal or vegetable origin) and salt. Generally soap is free fatty acids produced when an alkaline substance (caustic soda) reacts with fatty acids in fats and oil to saponify them. The salt of the free fatty acid or soap base are then added to other substances that have characteristic soap like properties of detergents, surface tension lowering, wetting and emulsifying power and gel-forming properties to produce a wide variety of soaps. Soaps could be in liquid, solid, semi-solid or powdered detergent forms. Soaps are formulated differently depending on their intended purpose. They can be categorized as plain (toilet) soaps and antibacterial (medicated) soaps [1]. Medicated or antiseptic soaps usually contain additional ingredients to treat skin conditions and contain added antiseptic substances in specific amounts indicated on the soap box or leaflet. This shows how the soap can be used for different purposes [2]. As a result of the added ingredients, these soaps have been reported to have more bactericidal properties compared with non-medicated soaps [3].

Soaps are meant to clean, kill and remove germs from the body of both animate and inanimate surfaces [4]. Previously, it has been reported that soaps can remove sixty five to eighty five percent of bacterial organisms on a person's skin [5]. Scrubbing your body and hands, especially with soap has been reported as a first line of defense against bacteria and other pathogens that can cause colds, flu, skin infections and some deadly infections (6). Bacteria are said to be ubiquitous that is found everywhere such as pool of water, soil, food, sewage etc [7]. The number of friendly bacteria on the skin surface varies from person to person [6]. The human's normal flora protects the skin against the entry and multiplication of other types of harmful bacteria in the body. Transient bacteria are deposited on the surface of the

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human skin from environmental sources which can cause skin infections. Examples of such bacteria are *Pseudomonas aeruginosa* [8] and *Staphylococcus aureus* [9]. Scrubbing with soap and water does not completely remove skin bacteria as they are firmly colonized in sweat and sebaceous glands and skin folds [10]. The requirement for efficient healthcare and technological advances has made it possible to incorporate certain chemicals into soaps to make them have antiseptic/antibacterial properties [11].

Despite the widespread availability of medicated soaps health problems related to many infectious diseases, food borne diseases and poor hygiene are prevalent. This may possibly be due to the fact that some of these antimicrobial consumer products may contain insufficient levels of antimicrobial agents. Prolonged usage of such products with insufficient levels of antimicrobial agents could result in the emergence of drug resistant bacteria. This study was carried out to compare the antibacterial effect of some conventional medicated soaps and also validate the reported usefulness of the medicated soaps.

2. Materials and Methods

2.1. Collection of soap samples

Eight brands of soap samples were purchased from retail sellers and stores in Okada market in Edo state Nigeria. These samples were purchased in their original packages and taken to the laboratory. The samples were coded A- H. The constituents of the different soap samples are given in Table 1.

2.2. Preparation and Dilution of soap samples

A stock soap suspension at concentrations of 50mg/ml was prepared for all soap samples using sterile distilled water. Other concentrations (25mg/ml and 12.5mg/ml) used in this study were obtained from serial dilutions of the stock concentration.

2.3. Isolation and Identification of Skin isolates

Skin swab samples were obtained by gently rubbing the outer skin surface of healthy Igbinedion University Okada students. Samples were randomly taken among the students. Swab sticks were moistened with; peptone water before taking samples. Swab sticks were used to inoculate previously prepared Mannitol salt agar and Macconkey agar media. Samples were processed using standard microbiological techniques, as previously described [12] Culture plates were incubated afterwards at 37^{o}C for 24 hours. Isolates were sub-cultured on nutrient agar plates to obtain pure colonies. Matrix-assisted laser desorption ionization time of flight (MALDI-TOF) mass spectrometry (Bruker Daltonik GmbH, Bremen, Germany)analysis was used for species identification.

2.4. Test organisms

Clinical wound isolates used include *Proteus mirabilis, Providencia stuartii* and *Pseudomonas aeruginosa* were obtained from the Department of medical microbiology Laboratory, University of Benin Teaching Hospital, Benin city, Edo state, Nigeria while isolates from normal skin flora used include *Staphylococcus aureus, Proteus mirabilis, Enterobacter kobei, Enterobacter xiangfangensis, Proteus hauseri* obtained from the skin surface of healthy igbinedion University Okada students. Isolates obtained were employed in the investigation. The wound isolates were pre-identified; hence, research ethics approval was not required. No contact was produced with patients and the original samples from the hospital. Informed consent was not required by the institution. The isolate data were obtained from clinical records and anonymously handled. Approval from the Igbinedion university ethical committee was duly obtained for samples obtained from the healthy students. The document IUO/ethics/056/24 was initiated and obtained for the study. All the participants were duly informed about the purpose and procedures of the study.

2.5. Antimicrobial assay

The standard agar diffusion method recommended by CLSI [13] was employed. A 24-hour culture of all isolates was prepared in sterile nutrient broth after which serial dilutions of the sub-cultured organisms were performed to obtain a concentration of 10^{-2}. A volume of 0.1ml of 10^{-2} concentration of the isolates was inoculated on already prepared and solidified Mueller-Hinton agar and gently swabbed over the different plates. The experiment was carried out in duplicates. A sterile cork borer (6mm) was used to make holes in the agar for the introduction of the different concentrations of the soap samples and controls. Gentamicin 10υg/ml was used as positive control and sterile distilled water was used as negative control. The plates were allowed to stand for one hour to ensure adequate diffusion of the soap before incubating the plates. The diameter of zones of inhibition was measured in mm after incubating the plates for 24 hours at 57^{o}C and the mean of duplicate experiments were recorded.

3. Results

Eight skin isolates were identified by MALDI-TOF MS as *Staphylococcus aureus* (2 isolates), *Proteus mirabilis* (3 isolates), *Enterobacter kobei*(1 isolate), *Enterobacter xiangfangensis*(1 isolate), *Proteus hauseri*(1 isolate). Four clinical wound isolates retrieved from the hospital were confirmed by MALDI-TOF MS as *Proteus mirabilis, Providencia stuartii* and *Pseudomonas aeruginosa.* All the soaps were found to possess varying antimicrobial activity in a concentration and organism dependent manner (Table 2 and 3). Soaps A, F and G were observed not to have excellent antimicrobial activities against the isolates compared to the other samples and the Gram positive cocci were observed to be more susceptible to the soaps compared with the Gram negative organisms. Only three and two skin and clinical wound isolates respectively were susceptible to the positive control. The soaps showed better antimicrobial activity against the isolates compared with the positive control (10υg Gentamicin)

Table 1 Ingredients of the Soaps tested

Soap	Ingredients
A	Soap base, aqua, sodium chloride, triclosan, trichlorocarbanilide (0.5% w/w), colour and perfume
B	PK/TL oil, sodium hydroxide, aqua, natural extracts
C	Information as regards the ingredients of the soap could not be retrieved
D	Information as regards the ingredients of the soap could not be retrieved
E	soap base, water, glycerin, fragrance, antibacterial agent, chloroxylenol 0.3% w/w, sodium chloride, BHT, tetra sodium EDTA, pine oil, titanium dioxide, CI 19140, total fatty matter NLT 65% w/w when packed
F	Sodium Palmate, Sodium Palm Kernelate, Aqua, Silica, Talc, Parfum, Glycerin, Sodium Chloride, 4-Chloro-3,5-Xylenol, Calcium Oxide, CI 77891, Limonene, Sodium C14-16 Olefin Sulfonate, Tetrasodium EDTA, Etidronic Acid, PEG-7 Amodimethicone, Sodium Laureth Sulfate, Trideceth-10, Sodium Sulfate, Hydroxypropyl Cyclodextrin, Tetradecene, Tetrabutyl Ammonium Bromide, Acetic Acid, o-Phenylphenate, Sodium Hydroxide, Methylchloroisothiazolinone, Methylisothiazolinone, CI 11680.
G	Soap base, Water, Glycerin, Talc, Fragrance, Menthol, Disodium, Distyrylbiphenyl Disulfonate, (Lauryl Alcohol, Phenoxyethanol, 2-Benzylheptanol and Decylene Glycol) 0.1%, Cl 77891, Cl 74160.
H	Soap Base, Perfume, Pine Oil, TCC (Trichlorocarban), TiO2(Titanium dioxide), Water, Glycerine, EDTA (Ethylenediaminetetraacetic acid), BHT (Butylated hydroxytoluene), EHDP (Etidronic acid), Colourant.

Table 2 Antimicrobial activity of the soaps on the skin isolates

Soaps	Conc used mg/ml	M1	M9	M15	F5	F7	M19	M10	F19
A	50	9	9	10	8	8	7	9	16
	25	9	13	11	8	7	9	14	15
	12.5	7	14	10	16	7	9	16	13
B	50	17	13	16	11	11	13	18	18
	25	12	9	10	16	8	9	10	7
	12.5	10	13	10	17	8	7	8	11
C	50	11	10	10	17	11	14	18	11
	25	13	9	7	15	10	10	8	9
	12.5	11	14	7	17	9	7	6	7
D	50	12	15	13	12	13	13	14	12
	25	14	12	13	10	8	8	10	8
	12.5	16	12	15	10	12	7	13	15
E	50	13	10	17	8	9	11	12	14
	25	12	11	11	7	9	12	7	14

	12.5	9	13	7	7	14	11	15	17
F	50 25 12.5	9 11 9	14 12 13	8 12 10	17 19 15	6 11 6	9 10 12	15 22 10	8 13 13
G	50 25 12.5	10 13 7	12 13 14	10 12 9	9 10 11	10 12 8	10 8 6	13 12 7	12 14 16
H	50 25 12.5 +	13 14 14 R	8 12 15 R	14 15 6 R	11 19 12 R	10 10 11 20S	11 15 16 R	17 13 7 20S	15 11 10 20S

M1 *Enterobacter Kobei*, M9 *Enterobacter xiangfangensis* M15 *Proteus hauseri* F5 *Staphylococcus aureus* M19 *Staphylococcus aureus* F7 *Proteus mirabilis* M10 *Proteus mirabilis* F19 *Proteus mirabilis* + Positive control 10ug Gentamicin R-Resistant S- Sensitive

Table 3 Antimicrobial activity of the soaps on the clinical wound isolates

	Conc used mg/ml	**2**	**7**	**16**	**30**
A	50 25 12.5	11 12 13	11 13 12	12 15 17	11 13 16
B	50 25 12.5	16 17 11	16 11 10	15 13 15	18 18 12
C	50 25 12.5	18 13 12	18 13 10	19 15 10	11 15 14
D	50 25 12.5	17 16 10	22 21 18	18 14 15	17 13 11
E	50 25 12.5	16 16 13	15 16 12	20 20 15	15 18 15
F	50 25 12.5	12 13 11	15 16 14	14 14 14	11 15 9
G	50 25 12.5	15 17 12	15 18 16	14 14 15	15 16 15
H	50 25 12.5 +	14 16 11 R	20 17 17 19S	15 16 11 20S	16 16 19 ND

2 *Proteus mirabilis* 7 *Providencia stuartii* 16 *Proteus mirabilis* 30 *Pseudomonas aeruginosa* + Positive control 10ug Gentamicin R-Resistant S-Sensitive ND- Not determined

4. Discussion

Soaps are employed mainly for washing or bathing with the aim of removing dirt and microorganisms present on the skin surface. The act of washing or scrubbing the body with the soap is meant to lead to a reduction in the number of microorganisms on the skin and this can contribute to a reduction in the incidence of skin infections. Whatever choice of soap an individual chooses to use, it should be such that will not affect the sensitive skin and should also be effective against potential disease-causing microbes present on the skin.

The assessment of the antimicrobial properties of the conventional medicated soaps in this study showed that the soaps possessed varying antimicrobial activity against healthy skin bacteria flora tested in a concentration dependent manner indicating that the soaps have constituents with antimicrobial properties. Aside the varying concentration dependent activity, the inhibitory antimicrobial action observed was also organism dependent. Majority of the soaps were active against the Gram positive organisms than the gram negative organisms. *Staphylococcus aureus* was the only Gram positive organism isolated from the skin of healthy students. Result from this study slightly correlates with a previous study on the antimicrobial assessment of some Nigerian herbal soaps [14]. The study reported that Gram positive organisms especially the gram positive cocci including *S. aureus, S. epidermidis*, and *S. capitis* were inhibited to a large extent by most of the herbal soaps tested. This is important and of significance as most skin infections such as acne, impetigo, furuncles and carbuncles are caused by this group of Gram positive organisms [14, 15, 16] and the use of these soaps against such infections is justified by the results of the study. Another previous study that assessed the comparison of antimicrobial activity of locally produced soaps and conventional medicated soaps on bacterial isolates from skin and wound reported the soaps tested showed varying levels of activity against *S. aureus* and *P. aeruginosa* [17]. This also correlates with results from this study as the soap samples were observed to varying inhibitory activity against all isolates tested. Santos-Junior [18] reported the evaluation of antibacterial and antifungal activity of antimicrobial soaps. The soaps tested were effective against the bacterial species *Staphylococcus aureus, Pseudomonas aeruginosa,* and *Acinetobacter baumannii*, and the fungus species *Candida albicans*. None of the soaps tested in that study showed inhibitory effect against the growth of *Escherichia coli*, *Proteus mirabilis*, and *Enterobacter cloacae*. This slightly contrasts with results from this study. The soaps tested had inhibitory effects on the isolates tested including *Staphylococcus aureus, Pseudomonas aeruginosa* and *Proteus mirabilis*. The variation in the size of the antimicrobial activity inhibition zones observed may be due to the increase in viscosity resulting from the higher concentrations of soap. When increasing the concentration of the samples, there may be difficulties in its diffusion and an increase in the degree of interactions with the solid culture medium [18]. The observed variability in antibacterial activity may also be possibly due to difference of antimicrobial active ingredient contents, type of formulations and repeated uses of the agents, which might have made some of the bacteria isolates less susceptible.

The soaps in this study were observed to have better antimicrobial activity against the isolates compared with the positive control (10υg Gentamicin). This confirms that the soaps had constituents that had antimicrobial properties. Constituents in the soaps with antimicrobial properties include triclosan, trichlorocarbanilide and chloroxylenol. Generally, antimicrobial soap could be any cleaning soaps to which antimicrobial active ingredients have been added. These ingredients kill bacteria and other microorganisms, although they are not effective on viruses

Soaps are meant for reduction of the inoculum sizes of microorganisms (pathogenic and non-pathogenic). Nonpathogenic microorganisms include the normal flora which is of two types: the resident flora that is the normal flora of the skin and other human body parts, and transient flora that are usually picked up from objects or other human beings [11, 19]. Antimicrobial soap products are usually obtained to stay healthy, with an intention to protect from potentially harmful organisms. However care has to be exercised in the prolonged use of these products as these could result in the potential increase in antibiotic-resistant pathogens in the environment making treatment of microbial infections more difficult to treat.

This study shows that in case of skin infections associated with the test organisms used in this study these soaps can be considered for treatment as most of the samples show satisfactory antibacterial activity. Irrational and long-time usage of these products should be discouraged as a result of the observed antimicrobial effects of the soaps. Topical antimicrobial products should be designed to meet the specific needs of users. This will result in the product more likely to have a long, useful, and profitable usage.

Compliance with ethical standards

Disclosure of conflict of interest

No Conflict of Interest is declared.

Statement of ethical approval

The wound isolates were pre-identified; hence, research ethics approval was not required. No contact was produced with patients and the original samples from the hospital. Informed consent was not required by the institution. The isolate data were obtained from clinical records and anonymously handled. Approval from the Igbinedion university ethical committee was duly obtained for samples obtained from the healthy students. The document IUO/ethics/056/24 was initiated and obtained for the study.

Statement of informed consent

All participants were duly informed about the purpose and procedures of the study.

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