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## Comparison of the Effectiveness of Antibacterial Activities of Locally made Black Soap and Some Selected Medicated Soaps on Isolated Human Skin Bacteria

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### ABSTRACT

The comparative study of the effect of locally made black soap and some medicated soaps like Dettol, Delta, Tetmosol, and Septol were investigated on isolated human skin bacteria using disk diffusion method. The bacteria isolated from the skin were *Bacillus species*, *Klebsiella pneumoniae*, *Staphylococcus aureus*, and *Staphylococcus species*. The results obtained revealed that Tetmosol soap exhibited the highest level of antibacterial activity, which result in the greatest zones of inhibition than other soaps; Black soap, Dettol, Delta, Tetmosol, and Septol on the isolated human skin microflora. Tetmosol soap through this study can thus, be recommended for use since it has the potentials of treating skin diseases caused by these isolated microflora.

### INTRODUCTION

The term normal microbial flora or microbiota denotes the population of microorganism that inhabits the skin and mucous membranes of healthy normal persons. Research has shown that this normal flora now referred to as normal microbiota provide a first line of defense against microbial pathogens, assist in digestion, play a role in toxin degradation, and contribute to maturation of the immune system. Shifts in the normal microbiota or stimulations of inflammation by these commensals may cause disease as inflammatory bowel disease (Tachibana, 1976).

The density and composition of the normal flora of skin vary with anatomical location. The makeup of the normal flora depends, upon various factors including genetic, sex, age, stress, nutrition and diet of the individuals. Human skin is subject to degenerative changes due to daily exposure to environment and the impact of microorganisms. Several studies have been carried out to determine the normal flora of the skin. (Marhall, 1987) presented a comprehensive review of the normal flora of the foot. He stated that the flora of the foot is similar to that found in the other skin site. (Marples, 1992) believe that some differences exist between foot flora and other skin site.

Developmental changes in humans such as weaning, the eruption of the teeth and the onset and cessation of ovarian function invariably affect the normal flora in the intestinal tract, the oral cavity, and the vagina respectively. However, within the limits to these fluctuations, the bacteria flora of human is sufficiently constant to a given general description of the situation. The efficacy of antimicrobial soaps in reducing the number of organisms can be tested in the axilla. If different soaps are used on each axilla in the same volunteer, the transfer of antimicrobial agent from one site to another may cause difficulties in interpretation. (Jawetz *et al.*, 2010).

Soap is a cleansing agent. It cleanses by lowering the surface tension of water, by emulsifying grease, and by absorbing dirt into the foam. Ancient peoples are believed to have employed wood ashes and water for washing and to have relieved the resulting irritation with grease or oil.

Scrubbing body or hands, particularly with soaps, is the first line of defense against bacteria and other pathogens that can cause colds, the flu, skin infections and even deadly communicable diseases (Larson, 1988; Kimel, 1996). Conceptually, many people consider that an antimicrobial portion of soaps is effective at preventing communicable diseases. But now researchers highlight that too much of it can have the opposite effect spreading diseases/infections instead of preventing them (Levy, 2001; Poole, 2002). Overutilization of medicated soaps might result in antimicrobial resistance and even rendering an individual more vulnerable to microbial attacks such as opportunistic skin infections (Russell, 1998; White, 2001).

On the other hand, regardless of a wide spread availability of the so called medicated soaps; a number of communicable infectious and food borne diseases as well as poor hygienic conditions related health problems are rampant. This can partially be explained by the fact that, occasionally some of these antimicrobial consumer products could have

insufficient quantities of antimicrobials. It seems to be more of a marketing phenomenon. Unfortunately, in the long run may adversely affect the consumers, because overuse of these agents can ascribe to the emergence of drug resistant microorganisms (Russell, 1998; Beinlich *et al.* 2001; White, 2001). This instigated us to embark on evaluation of the antimicrobial effects of the so called medicated soaps.

*Sabulun salo*; a local traditional medicated soap widely used by different tribes in Nigeria, such as Hausa, Yoruba (*oṣe dudu*) and Nupe (*eko zhiko*). It is a black soap, which has been used for centuries in many African homes especially in Ghana and Nigeria. The soap is produced from a mixture of vegetable oils (palm kernel oil and shea butter) that make the soap to have antimicrobial properties recognised in the traditional African households (Getradeghana, 2000).

In recent time, the soap has been improved industrially into more presentable forms (although many people still prefer the traditionally prepared one) with different trade names such as '*VillageFresh*', '*Dudu Osun*' '*Zee Black Soap*' etc. The attribute of the soap includes gentleness on the skin, rich lather, protection against skin disorders (including rashes, eczema, scabies) treatment of skin infection (such as ringworm), protection of even skin toning and smoothness of the skin (Getradeghana, 2000). Black soap can be used on the hair, face and body. Black soap has anti-aging properties and can reduce fine lines and wrinkles for youthful, smooth skin. Dark spots and blemishes are evened out and the natural ingredients effectively cleanse and deodorize. Black soap does not contain specific antimicrobial ingredient, many people prefer this soap because it does not cause resistant bacteria growth. Black soap is also a natural source of vitamins A, E and iron which helps to strengthen the skin and hair (Tachibana, 1976).

The major fatty acids in palm kernel oil are lauric acid (C12, 48%), myristic acid

(C14, 16%) and oleic acid (C18, 15%). Certain fatty acids (medium chain saturates) and their derivations have adverse effects on various microorganisms (Kabara, 1978). Monolaurin has been specifically found to have adverse effect on potentially pathogenic microorganism. Isaac and Thomas, (1991) reported the inactivation of *S. epidermidis* and group B Gram positive *Streptococcus* by lipases with high monolaurin content. The lauric acid content of the palm kernel oil has the additional beneficial function of being formed into monolaurin in human or animal body (Emg, 2000). This means that palm kernel oil may have higher antimicrobial effect *in vivo*. The palm kernel oil sample with highest lauric acid value have the highest effect on *S. aureus*, *Streptococcus* sp and *C. albicans*, this confirms that lauric acid is the antimicrobial agent in palm kernel oil (Ugbogu, 2006). *S. aureus* and *Streptococcus* sp. which cause skin and wounds infections are inhibited minimally by palm kernel oil. Although the antimicrobial activity observed is low, Kabara (1978) has shown that the use of this type of inhibitory agent does not lead to the development of resistant organisms.

Analysis of the kernel reveals the presence of phenolic compounds such as garlic acid, catechin, epicatechin, gallate as well as quercetin and transcinnamic acid (Steven *et al.*, 2003), some of which are known to have antimicrobial activities.

Medicated soaps to a large extent remove dirt and disrupt cytoplasmic membrane to kill microorganisms (Tachibana, 1976). It also works against enveloped virus like human immunodeficiency virus (HIV). Several antimicrobial substances are found in medicated soaps and they have various mode of action on various skin microflora.

This study is aimed at identifying bacteria commonly present on the human skin and to compare the effectiveness of some medicated soaps and locally prepared black soaps on the bacteria isolates obtained from the skin.

## MATERIALS AND METHODS

Sterile swab sticks were purchased from pharmaceutical store in Ilorin. The medicated soap (deltol, septol, tetmosol and delta) and black soap were obtained from ultra modern market, Ilorin.

### Collection of Samples

Skin swab were randomly collected from both male and female of Department of Biological Sciences, Alhikmah University, Ilorin, using a sterile swab stick that were dampened with sterile peptone water for each sample collected. The samples were collected from the hand, armpit, face and legs of different students and were later transferred to the laboratory for isolation.

### Collection and Preparation of the Soap

The soap (medicated and black soap) were made into small pieces using a sterile blade, one gram (1g) of each soap sample were dissolved in 9 ml of sterile distilled water to make a stock solution of 10<sup>-1</sup>. This was stored in a refrigerator in a well-sealed container to avoid further dilution.

### Preparation of Medicated and Black Soaps Dilution

Five sterile bottles, each containing 0ml, 2.5ml, 5ml and 7.5ml of sterile distilled water were properly labeled 100%, 75%, 50%, 25% and were dissolved respectively in 10ml, 7.5ml, 5ml, and 2.5ml of each soap samples. The solutions were mixed properly and stored in a refrigerator in well-sealed containers to avoid further dilution.

### Media Preparation

Three culture media used for this study, Nutrient Agar, Blood Agar and Mueller Hinton Agar were prepared according to the manufacturer's specification.

### Isolation of Microorganisms

The samples collected with swab sticks were inoculated on already set petri-dishes using streaking method. The culture plates were labeled and incubated at 37°C for 24 hours in an inverted position to prevent condensed moisture from dripping into the media or bacteria colonies. Biochemical characterization and identification of the

organisms were carried out using the Bergey's Manual of Determination Bacteriology, 9<sup>th</sup> edition (1994).

#### **Preservation of pure cultures of microorganisms**

Sub culturing was carried out for different colonies that grow on the mixed culture so as to obtain pure cultures of all isolates. This was done by streaking on a fresh, sterile solidified nutrient agar. The plates were then labeled and incubated at 37°C for 24hrs in an inverted position.

The pure culture growths were transferred into McCartney bottles containing nutrient agar and were incubated at 37°C until a visible growth was noticed, this was stored in the refrigerator to serve as stocked culture.

#### **Characterization and identification of isolates**

The bacteria isolated were characterized using the colonial characteristics, cellular morphology and biochemical properties. The colonial characteristics of the bacteria isolates were determined using the following parameters; Size, Elevation, Surface appearance, Edge, Opacity and Shape, while the cellular and biochemical properties were determined using tentative identification of isolates by Gram staining ; the colony character and the cell morphology were supplemented with biochemical tests ; Catalase test, Coagulase test, Citrase test, DNase test.

#### **Susceptibility test**

Disks of 7.0mm diameter was bored in the laboratory, it was then wrapped with foil paper and sterilized in a hot air oven at 100°C for 1 hour. The disks were later soaked in the different soap concentration for 1 hour to ensure that the disks were fully saturated. The disks were then aseptically transferred directly into the sensitivity plates. If not to be used immediately, the disks were aseptically removed from soap solution and were allowed to dry in a drying oven at a 25°C. They were then packed into sterile bottles, corked and stored in the refrigerator for use from time to time for susceptibility test. The impregnated disks were applied

immediately to the surface using sterile fill contact with the inoculated medium and to moisten the disk. The plates were then incubated at 37°C for 18-24 hours in an inverted position (Ndukwe *et al.*, 2005; Aliyu, *et al.*, 2009).

#### **Determination of minimum inhibitory concentration (MIC)**

This was determined according to the National Committee for clinical standard (1999). Five sterile test tubes containing 8ml of peptone water were properly labeled with different concentration of soap samples, i.e (100%, 75%, 50%, 25%). 1ml of each concentration of soap sample dilution was then introduced into corresponding test tube. Each set of 5 test tubes containing different concentration was challenged by 1ml of standardized test organism (A set of tube per organism) and were incubated at 37°C for 24 hours. This process was repeated for all other samples using a new set of five sterile test tubes containing 8ml of peptone water. The lowest concentration of each soap sample that completely prevented the growth of organisms microscopically was taken as the minimum inhibitory concentration (MIC).

#### **Determination of the minimum bactericidal concentration (MBC)**

According to the method of National Committee for clinical standard (1999), a small volume of about 0.5ml was removed from the test tubes in MIC in which there was no visible growth after incubation and spread over the surface of a sterile oven dried nutrient agar plate (without any agent) to determine the minimum concentration of soap required to kill the organisms. This was indicated by lack of growth on transfer to new plate. Growth on transfer to new plate indicates a bacteriostatic effect, the lowest concentration of soap sample indicating a bactericidal effect (i.e plate without growth) was taken as the minimum bactericidal concentration.

## **RESULTS AND DISCUSSION**

The normal human skin harbors microorganisms that can be grouped into transient and resident flora (Tachibana,

1976). Microorganisms differ in their nutritional requirements and level of susceptibility to antimicrobial agents. The effect of soaps on the skin microflora has not been widely studied. There is no soap that contains the required ingredient that suits all individual skin. Here skin is the main site of exposure to soap come into focus. The human skin is the main site of exposure to soaps, therefore reactions exhibited by individual skin differs from soap to soap. The different concentration of soaps tested on the different organisms isolated from human skin shows that the soaps contained antimicrobial activities which inhibited the

growth of the organisms, *Bacillus species*, *Klebsiella pneumoniae*, *Staphylococcus aureus*, and *Staphylococcus* species to different degrees.

The microorganism isolated from human skin at different parts (Armpit, face, leg and hand) from different male and female shows that *Bacillus* species were most predominant occurring microorganism in all the parts of the skin samples while *Staphylococcus* species were the least occurring microorganism from the isolated parts of the skin sample as presented in Table 1.

Table 1a: Microorganisms isolated from male human body parts

| Isolates |   | Gram staining  | Catalase Test | Coagulase Test | DNase test | Citrate Test | Organisms                     |
|----------|---|----------------|---------------|----------------|------------|--------------|-------------------------------|
| Armpit   | 1 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 2 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 3 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 4 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 5 | cocci +cluster | +             | +              | +          | -            | <i>Staphylococcus aureus</i>  |
| Face     | 1 | cocci +cluster | +             | +              | +          | -            | <i>Staphylococcus aureus</i>  |
|          | 2 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 3 | cocci +cluster | +             | +              | +          | -            | <i>Staphylococcus aureus</i>  |
|          | 4 | cocci +cluster | +             | +              | +          | -            | <i>Staphylococcus aureus</i>  |
| Leg      | 5 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 1 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 2 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 3 | cocci +cluster | +             | +              | +          | -            | <i>Staphylococcus aureus</i>  |
|          | 4 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 5 | cocci +cluster | +             | +              | +          | -            | <i>Staphylococcus aureus</i>  |
|          | 6 | cocci +cluster | +             | -              | -          | -            | <i>Staphylococcus species</i> |
| Hand     | 7 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 1 | cocci +cluster | +             | +              | +          | -            | <i>Staphylococcus aureus</i>  |
|          | 2 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 3 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 4 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 5 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 6 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |
|          | 7 | + rod          | +             | -              | -          | -            | <i>Bacillus species</i>       |

Table 1b: Microorganisms isolated from female human body parts

| Isolates |                | Gram staining  | Catalase Test | Coagulase Test | DNase test | Citrate Test                 | Organisms                     |
|----------|----------------|----------------|---------------|----------------|------------|------------------------------|-------------------------------|
| Armpit   | 1              | cocci +cluster | +             | +              | +          | -                            | <i>Staphylococcus aureus</i>  |
|          | 2              | + rod          | +             | -              | -          | -                            | <i>Bacillus species</i>       |
|          | 3              | - rod          | +             | -              | -          | +                            | <i>Klebsiella pneumonia</i>   |
|          | 4              | + rod          | +             | -              | -          | -                            | <i>Bacillus species</i>       |
|          | 5              | cocci +cluster | +             | +              | +          | -                            | <i>Staphylococcus aureus</i>  |
| Face     | 6              | cocci +cluster | +             | +              | +          | -                            | <i>Staphylococcus aureus</i>  |
|          | 1              | cocci +cluster | +             | +              | +          | -                            | <i>Staphylococcus aureus</i>  |
|          | 2              | cocci +cluster | +             | +              | +          | -                            | <i>Staphylococcus aureus</i>  |
|          | 3              | + rod          | +             | -              | -          | -                            | <i>Bacillus species</i>       |
|          | 4              | + rod          | +             | -              | -          | -                            | <i>Bacillus species</i>       |
|          | 5              | cocci +cluster | +             | -              | -          | -                            | <i>Staphylococcus species</i> |
| Leg      | 6              | cocci +cluster | +             | +              | +          | -                            | <i>Staphylococcus aureus</i>  |
|          | 7              | cocci +cluster | +             | +              | +          | -                            | <i>Staphylococcus aureus</i>  |
|          | 1              | + rod          | +             | -              | -          | -                            | <i>Bacillus species</i>       |
|          | 2              | - rod          | +             | -              | -          | +                            | <i>Klebsiella pneumonia</i>   |
|          | 3              | cocci +cluster | +             | -              | -          | -                            | <i>Bacillus species</i>       |
|          | 4              | - rod          | +             | -              | -          | +                            | <i>Klebsiella pneumonia</i>   |
|          | 5              | - rod          | +             | -              | -          | +                            | <i>Klebsiella pneumonia</i>   |
|          | 6              | cocci +cluster | +             | -              | -          | -                            | <i>Staphylococcus species</i> |
| Hand     | 7              | - rod          | +             | -              | -          | +                            | <i>Klebsiella pneumonia</i>   |
|          | 8              | + rod          | +             | -              | -          | -                            | <i>Bacillus species</i>       |
|          | 1              | cocci +cluster | +             | +              | +          | -                            | <i>Staphylococcus aureus</i>  |
|          | 2              | + rod          | +             | -              | -          | -                            | <i>Bacillus species</i>       |
|          | 3              | + rod          | +             | -              | -          | -                            | <i>Bacillus species</i>       |
|          | 4              | + rod          | +             | -              | -          | -                            | <i>Bacillus species</i>       |
|          | 5              | - rod          | +             | -              | -          | -                            | <i>Klebsiella pneumonia</i>   |
|          | 6              | - rod          | +             | -              | -          | -                            | <i>Klebsiella pneumonia</i>   |
| 7        | cocci +cluster | +              | +             | +              | -          | <i>Staphylococcus aureus</i> |                               |
| 8        | - rod          | +              | -             | -              | +          | <i>Klebsiella pneumonia</i>  |                               |

Microflora at 25%, the zones of inhibition of *Bacillus species* were very active with tetmosol soap and dettol soap (14.0mm each) while septol soap were less active (9.0mm). zone of inhibition of *Klebsiella pneumoniae* was very active with delta soap (10.0mm), black soap and septol soap were not active while dettol soap were least active with a value of 8.0mm. with

*Staphylococcus aureus*, tetmosol soap was very active (13.0mm), black soap and septol soap shows similar activity at 10.0mm each, deltal soap was less active while septol soap were not active. Tetmosol soap was very active (20.0mm) while black soap and septol were less active to the organism, as shown on Table 2.

Table 2: Effect of soaps against isolated human skin bacteria at 25% concentration

| Organisms                     | Zones of inhibition in mm |        |        |          |       |
|-------------------------------|---------------------------|--------|--------|----------|-------|
|                               | Black soap                | Septol | Dettol | Tetmosol | Delta |
| <i>Bacillus species</i>       | 11.0                      | 9.0    | 11.0   | 14.0     | 14.0  |
| <i>Klebsiella pneumonia</i>   | N.A                       | N.A    | 8.0    | N.A      | 10.0  |
| <i>Staphylococcus aureus</i>  | 10.0                      | N.A    | 10.0   | 13.0     | 9.0   |
| <i>Staphylococcus species</i> | 10.0                      | 10.0   | 12.0   | 20.0     | 15.0  |

N.A= Not Active

At 50% microflora, the zones of inhibition were active with tetmosol soap (15.0mm) while septol soap were less active on *Bacillus species*. Delta soap (10.0mm)

were very active on *Klebsiella pneumonia* with black soap (7.5mm) it was less active while septol soap and tetmosol soap were not active on the organism. Zone of inhibition

were very active with tetmosol soap (13.0mm) on *Staphylococcus aureus* while septol soap were less active on the organism, tetmosol soap (23.0mm) was very active on

*Staphylococcus* species while septol soap were less active on the organism as presented on Table 3.

Table 3: Effect of soaps against isolated human skin bacteria at 50% concentration

| Organisms                     | Zones of inhibition in mm |        |        |          |       |
|-------------------------------|---------------------------|--------|--------|----------|-------|
|                               | Black soap                | Septol | Dettol | Tetmosol | Delta |
| <i>Bacillus</i> species       | 14.5                      | 9.0    | 13.0   | 15.0     | 14.0  |
| <i>Klebsiella pneumonia</i>   | 7.5                       | N.A    | 8.5    | N.A      | 10.0  |
| <i>Staphylococcus aureus</i>  | 11.5                      | 9.0    | 10.5   | 13.0     | 10.0  |
| <i>Staphylococcus</i> species | 14.5                      | 11.0   | 14.5   | 23.5     | 17.0  |

N.A= Not Active

Microflora at 75%, the zones of inhibition was very active with black soap (16.0mm) on *Bacillus* species while septol soap (9.5mm) were less active on the organism. Delta soap (10.0mm) were very active on *Klebsiella pneumonia*, dettol soap (9.0mm) were less active on the organism

while septol soap and tetmosol soap were not active on the organism. The zone of inhibition on *Staphylococcus aureus* and *Staphylococcus* species with tetmosol soap (15mm; 23.5mm respectively) were very active while septol soap were less active on the two organism as presented on Table 4.

Table 4: Effect of soaps against isolated human skin bacteria at 75% concentration

| Organisms                     | Zones of inhibition in mm |        |        |          |       |
|-------------------------------|---------------------------|--------|--------|----------|-------|
|                               | Black soap                | Septol | Dettol | Tetmosol | Delta |
| <i>Bacillus</i> species       | 16.0                      | 9.5    | 14.0   | 15.0     | 14.0  |
| <i>Klebsiella pneumonia</i>   | 9.5                       | N.A    | 9.0    | N.A      | 10.0  |
| <i>Staphylococcus aureus</i>  | 11.5                      | 10.0   | 11.0   | 15.0     | 10.0  |
| <i>Staphylococcus</i> species | 14.5                      | 11.5   | 14.5   | 23.5     | 18.0  |

N.A= Not Active.

Table 5 shows that at 100%, the zones of inhibition on *Bacillus* species were very active with black soap (20.5mm), while septol soap (9.5mm) were less active on the organism. Black soap (10.0mm), Tetmosol (10mm), and Delta (10mm) were very active on *Klebsiella pneumonia*, while dettol soap

(9.5mm), were less active with septol soap not active on the organism. Tetmosol soap (15mm; 24mm) were active on both *Staphylococcus aureus* and *Staphylococcus species* respectively while septol soap (10mm; 11.5mm) were less active on both organism.

Table 5: Effect of soaps against isolated human skin bacteria at 100% concentration

| Organisms                     | Zones of inhibition in mm |        |        |          |       |
|-------------------------------|---------------------------|--------|--------|----------|-------|
|                               | Black soap                | Septol | Dettol | Tetmosol | Delta |
| <i>Bacillus</i> species       | 20.5                      | 9.5    | 14.0   | 16.0     | 16.0  |
| <i>Klebsiella pneumonia</i>   | 10.0                      | N.A    | 9.5    | 10.0     | 10.0  |
| <i>Staphylococcus aureus</i>  | 13.0                      | 10.0   | 11.0   | 15.0     | 11.0  |
| <i>Staphylococcus</i> species | 18.0                      | 11.5   | 16.0   | 24.0     | 18.0  |

N.A= Not Active.

The MIC of all the soap samples (25%/ml) were the same on *Bacillus species* and *Staphylococcus species*, it was higher with tetmosol soap (100%/ml) on *Klebsiella pneumonia* while septol soap were not active.

MIC were higher with septol soap (75%/ml) on *Staphylococcus aureus* while black soap, dettol soap and tetmosol soap were less, as shown in Table 6.



Table 6: MIC of soaps on the isolated human skin microflora in %/ml

| Organisms                     | Black soap | Septol | Dettol | Tetmosol | Delta |
|-------------------------------|------------|--------|--------|----------|-------|
| <i>Bacillus species</i>       | 25         | 25     | 25     | 25       | 25    |
| <i>Klebsiella pneumonia</i>   | 75         | N.A    | 50     | 100      | 25    |
| <i>Staphylococcus aureus</i>  | 25         | 75     | 25     | 25       | 50    |
| <i>Staphylococcus species</i> | 25         | 25     | 25     | 25       | 25    |

MIC=Minimum Inhibitory Concentration

N.A= Not Active

The MBC were higher with delta soap (100%/ml) on *Bacillus species* with Black soap (50%/ml) were less, while septol soap (75%/ml), dettol (75%/ml), Tetmosol (75%/ml), exhibit the same MBC on the organism. MBC were absent with all the soap samples on *Klebsiella pneumonia*. Septol soap (100%/ml) has higher MBC on *Staphylococcus aureus* while black soap,

dettol soap, tetmosol soap show similar MBC on the organism and delta soap were not active. MBC on *Staphylococcus species* were higher with septol soap (75%/ml), while tetmosol soap (50%/ml); black soap (50%/ml) and dettol soap (25%/ml); delta soap (25%/ml) shows similar MBC on the organism as presented in Table 7.

Table 7: MBC of soaps on the isolated human skin microflora in %/ml

| Organisms                     | Black soap | Septol | Dettol | Tetmosol | Delta |
|-------------------------------|------------|--------|--------|----------|-------|
| <i>Bacillus species</i>       | 50         | 75     | 75     | 75       | 100   |
| <i>Klebsiella pneumonia</i>   | N.A        | N.A    | N.A    | N.A      | N.A   |
| <i>Staphylococcus aureus</i>  | 75         | 100    | 75     | 75       | N.A   |
| <i>Staphylococcus species</i> | 50         | 75     | 25     | 50       | 25    |

MBC= Minimum Bactericidal Concentration

N.A= Not Active

At all concentration, Tetmosol shows the most antimicrobial effect on *Staphylococcus species*, the inhibition of the growth pattern of the isolates indicates the varying abilities of the organisms to resist the antimicrobial effect of the soaps. However these variations could be due to the differences in the nature and structures of the bacteria cell wall since it is the ultimate target of any antimicrobial agent or disinfectant. The result shows that the black soap and Tetmosol exhibited high level of antimicrobial activity which is the ability of the soaps to inhibit the growth or destroy the normal microflora.

The active ingredient in the soap is what distinguishes one type of the soap from another. The Tetmosol soap is found to contain Tetramethylthiuram monosulphide as the active antimicrobial agents. These chemical compounds function by denaturing all disrupting cell activity and interfering with microbial metabolism. These depend on a number of factors such as the inherent

properties of the organisms, contact time, the composition of the soaps, concentration of individual formulation and skin sensitivity. Traditional black soap lacks a key ingredient used in killing microorganisms such as Tetramethylthiuram monosulphide, instead when the soap is scrubbed into the skin; it helps release oils on the surface of the skin that can kill bacteria and rinsing microorganisms away on the skin and preventing the emergence of mutating bacteria.

The antibacterial activity of Black soap on gram positive bacteria can be attributed to the presence of cell wall, which is made up of mainly peptidoglycan. Peptidoglycan is found to be distorted by long chain fatty acids that are found in palm kernel oil an active ingredient in Black soap. The activity of the soap against *S. aureus*, *S. species* and *Bacillus species* therefore, could be attributable to the palm kernel oil present in the soap as reported by Ugboju, (2006); that palm kernel oil has inhibitory effect on *S.*

*aureus* and *Streptococcus* sp. The major fatty acids in palm kernel oil used for the production of Black soap are lauric acid, myristic acid and oleic acid. Certain fatty acids (medium chain saturates) and their derivatives have adverse effects on various microorganisms (Kabara, 1978). Monolaurin has been specifically found to have adverse effect on potentially pathogenic microorganism. Isaac and Thomas (1991) reported the inactivation of *S.epidermidis* and group B Gram- positive *Streptococcus* by lipases with high monolaurin content.

However, there were no observed bactericidal effects on *Klebsiella pneumoniae* by the soaps at all concentrations used. *Klebsiella pneumoniae* being Gram negative organism has little peptidoglycan in its cell wall and this may hinder the activity of the active components of the soaps. The resistance of *Klebsiella pneumoniae* to antimicrobial agents is usually due to chromosomal mutation which lowers the permeability of the bacteria to the agents or acquisition of resistance (R) plasmids and transposons (Arora, 2004). Therefore, the resistance showed by *Klebsiella pneumoniae* to the soaps may be due to chromosomal mutation which may have resulted to lower permeability of the bacterial cell.

The result shows that Tetmosol exhibited the highest level of antibacterial activity on the test organisms (*Bacillus* species, *Klebsiella pneumoniae*, *Staphylococcus aureus*, and *Staphylococcus* species) than other soaps (Black soap, Dettol, Delta and Septol) which is the ability of the soaps to inhibit the growth or destroy the normal microflora that are associated with various skin infections. It is therefore recommended that further studies should be conducted on Tetmosol such as its antifungal and antiviral effect on skin microflora.

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