ISSN 2348-1218 (print)
International Journal of Interdisciplinary Research and Innovations ISSN 2348-1226 (online)
Vol. 11, Issue 1, pp: (64-68), Month: January 2023 - March 2023, Available at: www.researchpublish.com

STUDIES ON CHARACTERIZATION AND ANTIMICROBIAL SUSCEPTIBILITY OF FACIAL MICROFLORA AND EFFICACY OF DIFFERENT FACE WASHES AGAINST ISOLATES

Saranya Thulasi[1], Bharathi Balasubramanian[2]*, Deepa C. Philip[3]

[1] Department of Medical Lab Technology, MMM College of Health Sciences, Chennai

[2*]Associate Professor, Department of Medical Lab Technology, MMM College of Health Sciences, Chennai

[3] Principal, MMM College of Health Sciences, Chennai

[*]Corresponding Author: Bharathi Balasubramanian

DOI: https://doi.org/10.5281/zenodo.7759220

Published Date: 22-March-2023

Abstract: **A diverse microbial flora is associated with the infections of skin and mucous membrane of every human being. The pathogenic bacteria can be easily removed by the use of antibacterial soaps, face washes, etc. It is common now to find out a wide array of antibacterial face wash products that claim to do miracles, though their actual results are questionable. Antibacterial face washes are popular means of prevention. But it is important to know how effective they are. In this study, 50 samples were collected from the faces belong to the age groups 20-45 years by using sterile cotton swabs moistened with sterile normal saline. Organisms were cultured on different culture plates and identified by biochemical tests. Most commonly isolated organisms from the face include *Staphylococci, Streptococci, Micrococci, Pseudomonas & E.coli.* Antimicrobial activity of antibiotics was done on isolates using MHA by disc diffusion method. Simultaneously, the antimicrobial susceptibility of the isolates was analysed with commonly known face washes namely Clean & Clear, Himalaya, Fair & lovely and Lakme. Out of four face washes used, clean&clear was the most potent antibacterial face wash that inhibit all the isolates. Fair&lovely shown least antibacterial activity against all the isolates.**

Keywords: **Microbial flora, *Streptococci, Micrococci, Pseudomonas, E.coli,* antibacterial face wash.**

I. INTRODUCTION

Hygiene is the practice of keeping oneself and one's surroundings clean in order to prevent illness or disease. Consequently, skin hygiene includes both skin cleaning and also taking care of its health. The human body which contain about 10^{13} cells, routinely harbour about 10^{14} bacteria, which constitute the normal microbial flora. The normal microbial flora is relatively stable with specific genera populating various body regions during particular periods in an individual's life. Normal flora may aid the host or may harm the host, or may exist as commensals. Though existing clinical studies have provided invaluable information about the abundance and types of microbes on the skin (1) they fail to address their function. The skin is a milieu for controlled bacterial growth and it supports the growth of commensals bacteria, which protect the host from pathogenic bacteria. Since the skin is the primary external coating of the human body and is exposed to the environment, it is inhibited by a number of bacteria (2).

Some of the studies stated that long term and transient bacterial resident flora isolated from the skin include those from the general *Staphylococcus*, *Streptococcus*, *Corynebacterium*, *Propionobacterium and Pseudomonas* (3). By colonizing the skin, the commensals microbes can restrict the colonization of other hostile microorganisms by competitive exclusion. The environment of the skin also predisposes the skin to selective colonization. Glands of the skin secrete compounds called fatty acids. Many organisms will not tolerate these fatty acids. Also sweat contains a natural antibiotic known as Dermicidin. The normal flora appears to be more tolerant to the Dermicidin than the skin is encouraged by the normal physiological condition of the body (4). In contrast to the protection, skin microbes can cause infection if they gain entry to other body parts through cut or during surgical procedure or due to malfunctioning immune system. Bacteria and other microbes that are normal resident of the skin case some six to ten percent of common hospital acquired infection (5).

Fortunately, these problematic bacteria can be easily removed by washing with ordinary soaps. However washing with harsh soaps or very frequent washing can increase the acidity of the skin, which can counteract some of the protective fatty acid secretion. Also the physical act of washing will shed skin cells. If washing is excessive the protective microbial flora will be removed, leaving the newly exposed skin susceptible to colonization by another, potentially harmful microorganisms. Health care workers, who scrub their hands frequently, are prone to skin infection and damage. Many hand and face washes now available with antimicrobial properties capable of acting against such infections. There are several reasons to use a well formulated cleansing product. The skin on the face and the neck is vascular rich and exposed to the elements constantly (6). For its protection, there is a hydro lipid barrier known as the acid mantle composed of Sebum and perspirations. This mantle protects the skin from sun, the dehydrating effects of the environment and invasion by bacteria and fungi. The mantle that is referred to as acid because the pH of the skin is between 4.5 and 5.5. Soap is alkaline with a pH between 9 and 12. It takes several hours for the acid mantle to re-establish itself (7). Dry skin can take up 4-8 hours to replenish. Conditions such as acne, eczema and itchy skin are associated with alkaline skin, which is further worsened by using soap or detergents based face wash products, more easily allowing bacteria to enter the disrupted protective barrier. High quality cleansing products are designed to remove impurities, infective bacteria, make up and sloughing skin cells without irritating or stripping the skin (8).

Due to increasing case of drug resistance pathogens, it is essential to know how to protect skin against possible infections. Antibacterial face washes are popular means of prevention. But it is important to know how just effective they are (9). To prove this, in our study four commercially available branded face washes are selected and observed how successfully they removed the bacteria. The study reflects that the antibacterial activity of different face washes against microorganisms isolated from the face and also to reveals which is the most potent face wash.

II. RESEARCH METHODS

Isolation of microorganisms from face

Sterile swab sticks damped with sterile saline were used to collect samples from face which were inoculated into nutrient agar plates and incubated at 37^{O} C for 24 hours. The most prominent isolates were selected; gram staining was done as a preliminary process and inoculated into peptone water. The colony characters were recorded and further identification of the organisms was carried out by biochemical characterization (10).

Identification of isolated organisms

Identification of the bacteria was done by using different biochemical tests based on the Gram staining reaction. Different biochemical tests were used in the study such as

a) Indole Test
b) Methyl Red Test
c) Voges Proskauer Test
d) Urease Test
e) Catalase Test
f) Citrate UtilizationTest
g) Oxidase Test
h) Hanging Drop preparation for Motility Testing

Antibiotic Sensitivity Test (AST) of isolates

Antibiotic sensitivity of the isolated facial microflora against four common antibiotics viz, Penicillin, Erythromycin, Tetracycline & Ampicillin was performed using disc diffusion method. 0.1ml of selected bacterial culture was uniformly spread on Muller hinton agar (MHA) plates to prepare Lawn culture. Sterile antibiotic discs were placed on the surface of MHA plates at a distance 2 cm using a sterile forceps. The plates were incubated at 37^{O} C for 24 hours and after incubation, zone of inhibition was measured (11).

ISSN 2348-1218 (print)
International Journal of Interdisciplinary Research and Innovations ISSN 2348-1226 (online)
Vol. 11, Issue 1, pp: (64-68), Month: January 2023 - March 2023, Available at: www.researchpublish.com

Antibiotic activity of face washes

Face washes of different brands like, Himalaya, Clean & Clear, Lakme, and fair &lovely were purchased from local market and used to compare the antibacterial activities according to the standard method. The dilution was prepared by dissolving 0.1 ml of face wash in 10 ml of sterile distilled water and used for the test. 0.1 ml of selected bacterial culture was uniformly spread on MHA plates and lawn culture was made with the isolates. Wells were made and each well incorporated with 20-30 µl of diluted face washes followed by incubated for 24 hours at 37^{O} C. The zone of inhibition was determined by measuring the diameter in millimeter of zone to which the face washes inhibit the growth of organisms.

III. RESULTS

About 50 different strains were isolated from face of 50 different individuals. Out of this, 14 samples were contain *Micrococci*, 14 samples were contain *Staphylococci.* *Streptococci* were present in 11 samples while *E.coli* and *Pseudomonas* were present in 05 and 06 samples respectively (Table 1.0).

Table 1: Number of samples containing isolates

Sl No	Isolates	No. of Samples Containing Isolates
01	*Micrococci*	14
02	*Staphylococci*	14
03	*Streptococci*	11
04	*E.coli*	05
05	*Pseudomonas*	06

Antibiotic Sensitivity Test (AST) of Isolates

Antibiotic sensitivity test showed that almost all the isolates were sensitive to erythromycin and tetracycline. Statistical analysis of AST shown in Table 2, *Micrococci* was produced highest zone of inhibition to Erythromycin followed by *Staphylococci*, *E.coli*. The least zone of inhibition was produced by *Pseudomonas*. Tetracyclinee showed highest antibiotic activity against *Pseudomonas*. Tetracyclinee exhibited least antibiotic activity against *Staphylococci*.

Staphylococci, *Streptococci* and *Pseudomonas* were resistant to Penicillin and Ampicillin. Most of the strains of *E.coli* and *Micrococci* were susceptible to all the four antibiotics.

Table 2: Statistical Analysis of AST

Organism	Zone of inhibition in mm			
	Erythromycin	Tetracyclinee	Ampicillin	Penicillin
Micrococci	12.68±3.15	11.93±2.92	5.66±3.24	6.20±2.67
Staphylococci	12.64±1.86	11.61±2.54	R	R
Streptococci	12.16±1.76	13.64±2.41	R	R
E.coli	12.50±1.66	15.50±1.00	4.70±4.30	6.46±3.81
Pseudomonas	12.22±1.24	16.08±2.55	R	R

*Values are in mean ± standard deviation R- Resistant

Antibacterial Activity of Face washes against Isolates

The facial microflora isolated were identified as *Staphylococci, Streptococci, Micrococci, Pseudomonas and E.coli* subjected to antibacterial activity of face washes and the results are tabulated in table 3.0

Table 3: Antibacterial activity of face washes against bacterial isolates

Isolates	Zone of inhibition in mm			
	Clean&clear	Himalaya	Lakme	Fair&lovely
Staphylococci	20	16	13	10
Streptococci	16	14	12	8
Micrococci	17	14	11	8
Pseudomonas	12	11	10	6
E.coli	10	15	12	6

Antibacterial activity of four face washes viz, Clean & Clear, Himalaya, Fair & lovely and Lakme showed that all the isolates inhibited by all the four brands of face washes.

- The Clean & Clear was the ***most potent*** among the four face washes as it shown highest Antibacterial activity against all the isolates. Clean & clear Exhibited highest zone of inhibition against *staphylococci* (20.00 mm) followed by *Micrococci* (17.00 mm), *Streptococci* (16.00 mm) and *Pseudomonas*(12.00 mm). The least zone of inhibition showed by *E.coli* (10.00 mm).

- Himalaya was the **second** most potent face wash among the four. Himalaya had highest antibacterial activity against *staphylococci* (16.00 mm). Also *E.coli* was largely inhibited by Himalaya (15.00 mm). least zone of inhibition showed by *Pseudomonas* (11.00 mm).

- In terms of Antibacterial activity among the above four face washes, Lakme at third position. *Staphylococci* showed highest zone of inhibition towards Lakme (13.00 mm) and the least zone of inhibition produced by *Pseudomonas* (10.00 mm).

- *Fair & lovely* shown **least** antibacterial activity against all the isolates among the four.

IV. DISCUSSION

According to this study, face washing is an effective method for removing majority of facial pathogenic microorganisms. In this study about 50 different strains were isolated from volunteers. The most frequent organisms isolated were *Micrococci, Staphylococci, Streptococci, E.coli, and Pseudomonas*. These organisms were subjected to antibiotic sensitivity test against four common antibiotics viz Erythromycin, Tetracyclinee, Ampicillin, Penicillin. Antibiotic sensitivity test showed that almost all the isolates were sensitive to Erythromycin and Tetracycline. Most of the strains resistant to Penicillin and Ampicillin. The isolated organism then subjected to antibacterial activity of four commercially available face washes such as Clean&clear, Himalaya, Lakme and Fair&Lovely.

Out of Four commercially available face washes I used, Clean & clear is the most potent antibacterial face washes that inhibiting all the isolates at the Dilution test. Himalaya was the second most potent face wash among the four. Fair & lovely shown least antibacterial activity against all the isolates among the four. The study conducted by Bojar and Holland (2002) about antibacterial activities of medicated soaps on selected human pathogens concluded that Crusader soap had the highest antibacterial activity (25mm, against *Staphylococcus aureus).* Significant differences were observed on the different concentrations of soap preparations used in the work. *Staphylococcus aureus* was very sensitive to most of the antibacterial soaps used, while *E.coli* showed higher resistance to the soaps (12).

The medicated soaps analyzed have bacteriostatic and bactericidal effects on the test pathogens while complete resistance was shown by some of the test isolates even at higher concentrations of the soap preparations used (13). The cloth washing soap had no antibacterial effect on the tested pathogens. The use of medicated soaps is thus recommended in homes, schools, offices and hospitals as a way of minimizing or stopping infections that are hitherto spread through the hands (14).

V. CONCLUSION

Many of the environmental impurities and cosmetic products are not water soluble and so washing the skin with simple water would not be sufficient to remove them. Substances capable of emulsifying them into finer particles are to be used for making these fat soluble impurities water soluble. Herein, cleansers ie face washes fit into the picture. They are surface—active substances (i.e. emulsifiers/ detergents/ surfactants/ soaps) that lower the surface tension on the skin and remove dirt, sebum, oil from cosmetic products, microorganisms, and exfoliated corneum cells in an emulsified form. Hence, the study recommends that, whenever the people are visiting infection prone areas like hospitals, industries, crowded areas and after long travelling, can use face washes like Clean&clear followed by Himalaya. That will help to remove the pathogenic bacteria from the face and protect the skin from the colonization of other pathogenic microorganisms.

REFERENCES

[1] Roth R, James WD. Microbial ecology of the skin : resident flora, ecology , *infection journal.* 1989; 20:367-390.

[2] Fredricks DN. Microbial ecology of human skin in health and disease. *Journal investigation dermatology*. 2001; 6:167-169.

[3] Cogen AL, Nizet V, Gallo RL. Skin microbiota; a source of disease or defence? *British Dermatology.* 2008; 158:442-455

[4] Harder J, Bartel S, Christopher E, Schroder M. A peptide antibiotic from human skin. *Nature*. 1997; 387:867.

[5] Bojar KA, Holland KT. The human cutaneous microbiota and factors controlling colonization. *World Journals Microbiology Biotechnology* 2002; 18:889-903.

[6] Feroze, K., Elsayed, A and Tarek, TA. Antimicrobial activity of commercial antibacterial handwashes and soaps. *Indian Dermatology Online Journals.* 2014; 5(3): 344-346.

[7] Grice EA, Kong HH, Conlan S. Topographical and temporal diversity of the human skin microbiome. 2009; *Science* 324:593-595.

[8] Selvamohan,, V and Sandhya, T. Studies on the bactericidal activity of different soaps against bacterial strains. *Journal of Microbiology and Biotechnology Research* 2012; (5): 646- 650

[9] Wootton C.I, Bell.S, Mayxay.M. Assesing skin disease and associated health related quality of life in a rural Lao community. *BMC Dermtology* 2008; 18, Article number: 11.

[10] Brockman ER. Bergey's Manual of Systematic Bacteriology. 1986. Volume 3. Edited by Sneath PHA, Mair NS, Sharpe ME, Holt JG. Williams & Wilkins, Baltimore.

[11] Bauer, A.W, Kirby, W.M, Sherris, J.C and Jurck, M. Antibiotic susceptibility testing by a standard single disc method. *American Journal Clinical Pathology.* 1966; 451: 493-496

[12] Barak O, Treat JR, James WD. Antimicrobial peptides: Effect of innate immunity in the skin. *Advanced Dermatology* 2005; 2:357-374.

[13] Cheesbrough M. District laboratory practice in tropical countries, part 2. Cambridge University Press, Cambridge. 2005; 159- 162.

[14] Obi CN. Antibacterial activities of some medicated soaps on selected human pathogens. *American journal of microbiological research*. 2014. Volume 2(6): 178-181.