

Research Article

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Varsha M Chaudhari

of Microbiology. Department PSGVPM'sS.I.Patil Arts, G.B.Patel Science and S.T.S.K.V.S Commerce College, Shahada, District Nan durbar, Maharashtra, India

Correspondence: Varsha M Chaudhari

of Microbiology, Department PSGVPM'sS.I.Patil Arts, G.B.Patel Science and S.T.S.K.V.S Commerce College, Shahada, District Nan durbar, Maharashtra, India

Studies on antimicrobial activity of antiseptic soaps and herbal soaps against selected human pathogens

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Varsha M Chaudhari*

Abstract

In the present study antimicrobial activity of various antiseptic and herbal market soaps were determined against bacterial isolates present on the skin surface like Staphylococcus aureus, Bacillus subtilis, Escherichia coli and Pseudomonas aeruginosa using agar disc diffusion method. Various microbes are deposited on the surface of skin from the dust present in external environment which causes infection. Antimicrobial activities of various soaps on such micro flora pathogens were studied. The results obtained revealed that amonghe antiseptic soaps highest efficacy was performed by Dettol and Lifebuoy Plus soaps against Gram positive isolates but in Gram negative isolates Dettol and Savlon had maximum bactericidal activity. In Herbal soaps Neem followed by Aloe Vera was found to be most effective with all isolates as compared to Haldi Chandan soap. Pseudomonas aeruginosa was found as most resistant isolate to all soaps in the present study.

Keywords: Antiseptic and herbal soaps, Antibacterial activity, Skin flora pathogens, Diffusion.

INTRODUCTION

Antimicrobial activity of any substance is defined as its ability to either kill bacteria or inhibit the growth of bacteria. Antimicrobial activity is significant with respect to the human body in preventing diseases and skin infections. Detergents and soaps are the disinfectants required in daily practices for hygienic point. Soaps are cleaning agents, which may be liquid, solid, semisolid or powders. Soaps are used to remove dirt, including dust, microorganisms, stains and bad smells in order to maintain health, beauty and remove bad odor from the body or inanimate objects, including clothes ^[1].

Chemically soaps are the combination of fats, oils (of animal or vegetable origin) and Salt^[2]. Soaps are generally salts of free fatty acid made via saponification, where alkaline substances react with fatty acids in fats or oils. Other substances are then added to this salt of free fatty acid or soap base, to produce the different types of soaps we have. They are mainly used as surfactants for washing, bathing and cleaning^[3]. Soaps are either non antimicrobial soaps or an antimicrobial soap, also known as an antiseptic or medicated soap. An antibacterial soap can remove 65% to 85% of bacteria from human skin^[4]. Antiseptic soaps are incorporated with specified amount of germicidal substances in addition to the ordinary soap base in order to increase their antibacterial activity. These antiseptic substances impart ability for the soap to kill germs even after it has been used as residual antiseptic substances remain on the skin. It is proved experimentally that antibacterial soaps kill the bacteria at a specific concentration; they also have bacteriostatic activity and can inhibit the growth of bacteria. Herbal soaps are prepared by adding dried herbs, flowers and stems into soap base. Herbs are the natural products could be found in the treatment of almost all diseases and skin problems owing to their high medicinal value, costeffectiveness, availability and compatibility ^[5, 6]. Hence it can be used in soap base. 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^[7].

The soap should have good ingredients which have the ability to kill bacteria but not to damage body tissues. Number of bacteria including Gram Positive and Gram negative are deposited from the environment on the surface of skin and causes skin infection. Examples of these bacteria include Staphylococcus aureus^[8], Bacillus subtilis and Pseudomonas aeruginosa^[9]. Spread of infection by such bacteria can be prevented by use of antiseptic soaps, as it contains antimicrobial chemicals, but over use of soaps might results in antimicrobial resistance and even rendering a person more sensitive to allergies,

skin rashes [10].

The aim of the present research work is to compare the efficacy of locally available market herbal soaps and antiseptic soaps against skin infecting human pathogenic bacteria such as *Staphylococcus aureus*, *Pseudomonas aeruginosa, Bacillus subtilis* and *E. coli*.

It was seen clearly that Gram positive bacteria were killed at low concentration of soaps than Gram negative bacteria. The most resistant bacterium of all the soaps was appeared as *Pseudomonas* followed by *Bacillus*. This study suggests that selection of soaps and cleaning products depends on their effectiveness in killing commonly encountered pathogenic microbes in day to day activities. Selection of soaps should depend on the present environment of the person and soaps having natural ingredients are better for constant long use.

MATERIAL SAND METHODS

This study was carried out in Department of Microbiology PSGVP Mandal's ASC College, Shahada.

Sample Collection

Soaps used

Different types of herbal and antiseptic soap samples, which are commonly used by humans, were purchased from standard pharmaceutical shop from Shahada. The content and expiry dates of all soaps were noted. Antiseptic soaps used in the present experiment were Dettol, Savlon, Lifebuoy Plus and herbal soaps used for the present experiment were purchased from Patanjali stores, Shahada, which includes Haldi Chandan, Aloe Vera and Neem soap.

Isolation of microorganisms

Bacterial cultures used for the present study includes *Staphylococcus* aureus, Bacillus subtilis, E. coli and Pseudomonas aeruginosa were isolated in laboratory.

Sterile moistened cotton swab sticks were used to collect skin swabs samples from the hands, necks and faces of the students from the Department who were exposes to heavy dust during their bus up-down.

The samples collected with swab sticks were then used to inoculate already prepared nutrient and LB agar plates. The cultured plates were then incubated at 37°C for 48 hours. Biochemical characterization and identification of the test pathogen was also carried out using the Bergey's Manual of Systematic Bacteriology.

Preparation and dilution of soap samples extract

With the help of sterile sharp knife soaps were scraped atone side. 250 mg and 500mg of each soap sample was weighed and dissolved in 1mlof sterile distilled water separately and used for the preparation of discs.

Preparation of Sterile Disc

Filter paper discs were prepared from Whatman's No.3 filter paper. Discs of 6mm size were prepared in Petri plate and sterilize in an autoclave at 121^{0} C for 15 minutes. Each sterile disc was incorporated individually with 250 mg and 500 mg of soap solution. Paper discs soaked in the soap solution were allowed to stand for a period of one hour to ensure full saturation of the soap preparations. The discs were then aseptically removed from soap solution and allowed to dry in an oven at 25°C.

Assay of Antimicrobial Activity

Overnight cultures were kept ready for anti-microbial activity. Assay of the antimicrobial activity of soaps were done by disc diffusion method.

Discs diffusion assay

Agar disc diffusion method ^[11] was used to detect antimicrobial assay. The standardized 0.1 ml saline suspension of test organisms were inoculated on the surface of sterile Mueller-Hinton agar plates. Sterile filter paper discs prepared from different concentrations of the various soap samples were aseptically transferred directly into the surface of plates with the help of a sterile forceps. All plates were incubated at 37^{0} C for 24-48 hours and then were examined for zone of inhibition around the disk. The zone of inhibition was determined by measuring the diameter in millimeters of zone to which the soap inhibited the growth of the organism.

Statistical Analysis

The data obtained from the above study was analyzed statistically using Analysis of Variance (ANOVA).

RESULTS AND DISCUSSION

In general soaps are used for cleaning purposes and in order to remove dust and microbes present on the surface of skin. The choice of soap varies from person to person but it should not affect the sensitive skin and it should be effective against disease causing microbes present on skin.

The present research investigation was carried out to determine the antimicrobial efficacy of antiseptic soaps like Dettol, Savlon, Lifebuoy Plus and herbal soaps like Haldi Chandan, Aloe Vera and Neem against skin micro flora isolates *Staphylococcus* aureus, *Bacillus subtilis*, *E. coli* and *Pseudomonas aeruginosa*.

Results obtained from the experimental data revealed that most of the studied antiseptic soaps have antimicrobial activity, though to varying degrees as indicated by the inhibition of the growth pattern of the isolates. Varied levels of effectiveness by soaps were observed against the isolated skin flora pathogens.

Table 1: Diameter of Zone of Inhibition (mm) on Staphylococcus aureus by various soaps

	Antiseptic soaps			Herbal soaps			
	Mean±S.D.						
Concentrations	Dettol	Savlon	Lifebuoy Plus	Haldi Chandan	Aloe Vera	Neem	
250mg/ml	31.0 ± 1.0	12.0 ±1.2	9.0 ±1.0	9.0 ±1.2	12.0 ±1.2	13.0±1.5	
500mg/ml	42.0 ±1.5	13.6 ± 1.2	11.2 ±1.2	10.2 ±1.0	14.0 ± 1.5	14.4±1.2	

Table 2: Diameter of Zone of Inhibition (mm) on Bacillus subtilis by various soaps

	Antiseptic soaps			Herbal soaps			
	Mean ± S.D.						
Concentrations	Dettol	Savlon	Lifebuoy Plus	Haldi Chandan	Aloe Vera	Neem	
250mg/ml	22.0±1.0	10.0 ±0.5	8.4 ± 1.2	NA	NA	11.0±1.0	
500mg/ml	30.0±1.5	11.5 ±1.0	11.0 ±1.0	11.4 ±1.5	12.0 ±1.5	13.2 ±1.0	

Table 3: Diameter of Zone of Inhibition (mm) on E. coli by various soaps

	Antiseptic soaps			Herbal soaps		
	Mean ± S.D.					
Concentrations	Dettol	Savlon	Lifebuoy Plus	Haldi Chandan	Aloe Vera	Neem
250mg/ml	18.0 ± 1.0	12.0±0.5	15.0 ± 1.5	9.5 ±0.5	12.5 ± 1.0	10.2 ±0.5
500mg/ml	25.0 ±1.0	16.0 ± 1.0	17.5 ±1.0	11.8 ±1.0	13.2 ± 0.5	12.4 ± 1.5

Table 4: Diameter of Zone of Inhibition (mm) on Pseudomonas aeruginosa by various soaps

		Antiseptic soaps	5		Herbal soaps	
	Mean ± S.D.					
Concentrations	Dettol	Savlon	Lifebuoy Plus	Haldi Chandan	Aloe Vera	Neem
250mg/ml	16.2±1.0	10.0 ± 0.5	14.2 ± 1.2	NA	NA	10.3 ±1.0
500mg/ml	18.6 ±1.5	13.5 ±1.5	16.5 ±1.0	NA	NA	14.2±1.2

NA: Zone not appeared

When the efficacy of the antibacterial soaps were compared using the disc agar diffusion method, analysis of variance for the Means of antimicrobial activities among the different soaps revealed the positive correlations (P<0.05). The results of the zone of inhibitions using the organisms showed that there were significant differences (P<0.05) on the various microorganisms used for the study. *Staphylococcus aureus* have more zone of inhibition (42mm) while *Bacillus* have zone of inhibition (30mm) followed by *E. coli* (25mm) and *Pseudomonas* have (18 mm). Significant differences were observed in the zone of inhibitions in all types of antiseptic and herbal soaps used for the study.

Analysis of variance also revealed that positive correlation exists among the different concentrations of antiseptic and herbal soaps used for the study. It was observed that significant differences (P<0.05) exists among the different concentrations of soaps used for the study with 500mg/ml, higher concentration having higher zone of inhibitions were observed than lower concentration.

Analysis of variance revealed that there is no significant difference (P>0.05) exists among the two types- antiseptic and herbal soaps. When the antibacterial activity of antiseptic and herbal soaps were compared Neem (14.4mm) and Aloe Vera (14.0mm) soaps have similar zone of inhibition with Savlon soap(13.6mm) in *Staphylococcus aureus*. With other pathogens also results showed similar zone of inhibition.

Results obtained showed that Dettol was found to be most effective against all the pathogenic strains tested having the highest zone of inhibition (42mm) against *Staphylococcus aureus* and 30 mm against *Bacillus subtilis* at the highest concentration 500 mg/ml when used. Followed by Dettol, Savlon also inhibits the growth of *Staphylococcus* but least zone was appeared for *Bacillus* species. Among the antiseptic soaps Lifebuoy Plus showed least zone of inhibition against *Staphylococcus* and *Bacillus* but it inhibits the growth of *E. coli* and *Pseudomonas* species.

Among the different herbal soaps studied Neem showed highest antimicrobial activity against all pathogen studied as compared to Haldi Chandan soaps. Haldi Chandan exhibited the least antibacterial activity with zone of inhibitions of 10.2 mm for *S. aureus*, 11.4 mm for *Bacillus* and 11.8 for *E. coli* species. In herbal soaps Aloe Vera was found to effective against *E. coli* but *Pseudomonas* was not found to be inhibited by Haldi Chandan and Aloe Vera soaps also.

Pseudomonas exhibited higher resistance to soaps, no zone of inhibitions were recorded for this isolate. Savlon was found to be as efficient as Dettol in inhibiting the growth of pathogens.

CONCLUSION

The soaps are cleaning agents routinely used for cleaning purposes and removing germs. Soaps and detergents disrupt the microbial cell membrane and disrupt cells proteins. Soaps tested in the present research work showed varied levels of activity against the pathogenic microbes tested. Hence Dettol followed by Savlon among other soaps showed antibacterial activity and can be used to prevent skin infections and transmission of skin pathogens when used in hand washing. However, prolonged used of these soaps could lead to development of microbial resistance and allergic reactions to skin. The main aim of this study was to identify the antimicrobial activity of antiseptic soaps and herbal soaps. Antiseptics soaps possess active germicidal chemical agent, it kills pathogen but its prolonged use should be avoided. As herbal soaps revealed similar antimicrobial activity like that of antiseptic soaps, they are more effective and considered better in our daily life. The soap is routine requirement of life, but it should have good active ingredients or plant extracted material which has the ability to kill bacteria but not to harm body tissues when used constantly.

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