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Full Length Research Paper

Antibacterial activity of soaps against daily encountered bacteria

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This study aims to check the antibacterial activity of various branded soaps against bacteria that are normally present in the environment. The proposed study includes selection of most common bacterial strains from the environment. Identification of bacterial strains was done by standard microbiological techniques, which include gram staining, biochemical testing and advanced identification by analytical profile index. Determination of minimal inhibitory concentration and minimum bactericidal activity of strains was performed by tube and microtitration method. Antibacterial soaps showed better MIC in comparison with beauty soaps. The most resistant bacterium to all the soaps is *Klebsiella pneumoniae* and *Pseudomonas aeruginosa*. It is obvious that antibacterial soaps have the antibacterial agents that can either kill or inhibit the bacterial cells. It might be possible that some bacterial strains become resistant which leads to their survival even at high concentrations of soaps.

Key word: Soaps, antibacterial activity, daily bacteria.

INTRODUCTION

Soaps play an important role in removing and killing bacteria. Although fats and oils are general ingredient of soaps but some detergents are added to enhance the antibacterial activities of soaps (Friedman and Wolf, 1996). According to Osbore and Grobe antibacterial soaps can remove 65 to 85% bacteria from human skin (Osborne and Grube, 1982). Bacteria are very diverse and present every where such as in soil, water, sewage, standing water and even in human body. Bacteria's that attacks on human body is of great importance with reference to health (Johnson et al., 2002). Transient bacteria are deposited on the skin surface from environmental sources and causes skin infections. Examples of such bacteria are *Pseudomonas aeruginosa* (Fluit et al., 2001) and *Staphylococcus aureus* (Higaki et al., 2000). The importance of hand washing is more crucial when it is associated to health care workers because of possible cross contamination of bacteria that may be pathogenic or opportunistic (Richards et al., 1995). Studies have shown that soaps containing antimicrobial active ingredients remove more bacteria as compared to plain soap (Lucet et al., 2002). Handhygiene and prevention of infection has been well recognized (HWG, 1999). The importance of hand hygiene is also there for food handlers. Food handler includes those who deals with delivers and serve food (Horton and Parker, 2002). Dr Elaine Larson published an extensive review and concluded that hand hygiene and reduced tran-smission of infections is a convincing fact (Larson et al., 1986). To investigate the antibacterial efficiency of different brands of soaps, we isolated bacteria from different environments and human skin. Identification of bacteria was done by biochemical tests (Cheesbrough, 2001) and by using analytical profile index. Minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) against these bacteria were determined. Identification of bacterial species that are most resistant to the antibacterial soaps of daily use was made. The present studies were aimed to determine the bactericidal activity/efficacy of both the antibacterial as well as beauty soaps and to determine, whether the soaps only removes the bacteria from skin or it also kills the bacteria.

MATERIALS AND METHODS

Soaps used

Safeguard soap, Lifebuoy Red soap, Lifebuoy White soap, Dettol

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Table 1. Sampling sites of Bacteria.

S/N	Bacteria	Collection area
1	*Staphylococcus aureus*	Soil
2	*Pseudomonas aeroginosa*	Waste Water
3	*Escherichia coli*	Sewage Water
4	*Pseudomonas*	Waste Water
5	*Klebsiella pneumoniae*	Polluted Soil
6	*Klebsiella pneumoniae*	Polluted Soil
7	*Enterobacter* spp.	Sewage Water
8	*Pseudomonas*	Waste Water
9	*Bacillus subtilis*	Under Nails
10	*Staphylococcus epidermidis*	Skin

bar soap, Dettol Liquid hand wash (antibacterial soaps), Lux soap, Palmolive soap, and Capri soap (Beauty soaps).

Isolation of bacteria

Different bacterial strains were isolated (Table 1). All samples were properly diluted and spread on the nutrient agar. The pH was adjusted to 7.0, incubated for 24 h at 37°C. Then inoculation was made on nutrient agar plates and incubated at 37°C for 24 h. The most abundant strain of the samples was selected, gram stained, and then purified on new plate of nutrient agar.

Identification of isolated bacteria

Identification of bacteria was done by using different biochemical tests. These tests were based on the gram stain reaction of bacterial strains. Tests includes, oxidase test, catalase test, urease test, motility test, acid production from glucose, mannitol, sucrose, lactose, maltose, coagulase test, Dnase test, indole test, eosine methylene blue test, triple sugar iron reactions, methyl red test, voges proskauer test, and nitrate reduction test following chesseborugh (Cheesbrough, 2001). For confirmation of gram negative bacteria, analytical profile index (biomereux) was performed according to manual instructions (Table 2).

Strain maintenance

All strains were grown on nutrient agar plates at 37°C for 48 h. Strains were stored at -70°C in 50% sterile glycerol and TSB (Aulet, 2001). Minimum inhibitory and bactericidal concentrations of different soaps To determine minimum inhibitory concentrations of different soaps, two different methods were used. One was tube method (Cappuccino and Sherman, 1992) and other was micro titration plate method (Johnson et al., 2002). Bactericidal concentrations of soaps were determined following Cappuccino and Sherman (1992).

RESULTS

The present study suggested that the choice of soap should be that which does not affect the facial tissues as well as effective against disease causing bacteria in a small amount. For the determination of MBC and MIC, soaps of daily use were employed (Figures 1 and 2). Safeguard soap MBC is 250 mg/ml and its MIC was Safeguard is an antibacterial soap that has bactericidal observed at 125 mg/ml for *Staphylococcus aureus*. Safeguard is an antibacterial soap that has bacteric idalagents in it. For *Pseudomonas aeroginosa* (1) its MBC was at 500 mg/ml and MIC was at 250 mg/ml. If it is compared with the *S. aureus* it clearly showed that it was killed at high concentration of soaps. The MBC ofsafeguard soap against strain of *P. aeroginosa* strain 2 was observed at 250 mg/ml and its MIC was observed at 25 mg/ml. For *E. coli* MBC was 125 mg/ml and its MIC was observed at 62.5 mg/ml. Safeguard soap was used against *Klebsiella pneumoniae* its MBC at 500 mg/ml and its MIC was observed at 250 mg/ml. The MBC of safeguard soap against this strain of *Pseudomonas* wasobserved at 250 mg/ml and its MIC was observed at 125mg/ml (Figure 3). If it is compared with the first strain of *P. aeroginosa* two prominent differences were observed which showed that strain was more sensitive than the first one. Dettol soap showed MBC at 250 mg/ml and MIC was observed at 125 mg/ml against *S. aureus.* These values were compared to the values obtained from safeguard soap. These were almost equal to the values of safeguard. It might be estimated that the antibacterial activity of safeguard and dettol soap were almost the same against this organism. Lifebuoy red (antibacterial): This soap showed its MBC at 250 mg/ml and MIC was observed at 125 mg/ml against *S. aureus*. The comparison of safeguard, lifebuoy and dettol soaps revealed the equivalence of MBC and MIC values. It was also estimated that the organism might be sensitive to the antibacterial-soaps.-Lifebuoy-(Red)-is-also--an tibacterial soap, it showed its MBC against is *Pseudomonas* spp. At 350 mg/ml and MIC was seen at 175 mg/ml which much lower than safeguard and dettol soap. The antibacterial agents used in this soap showed more antibacterial activity in comparison with the above mentioned soaps. This soap showed its MBC at 250 mg/ml and MIC at 150 mg/ml or close to it. For *Escherichia coli* and *Klebsiella*, this soap showed its MBC at 500 mg/ml and MIC was seen at 250 mg/ml which is very high concentration of the

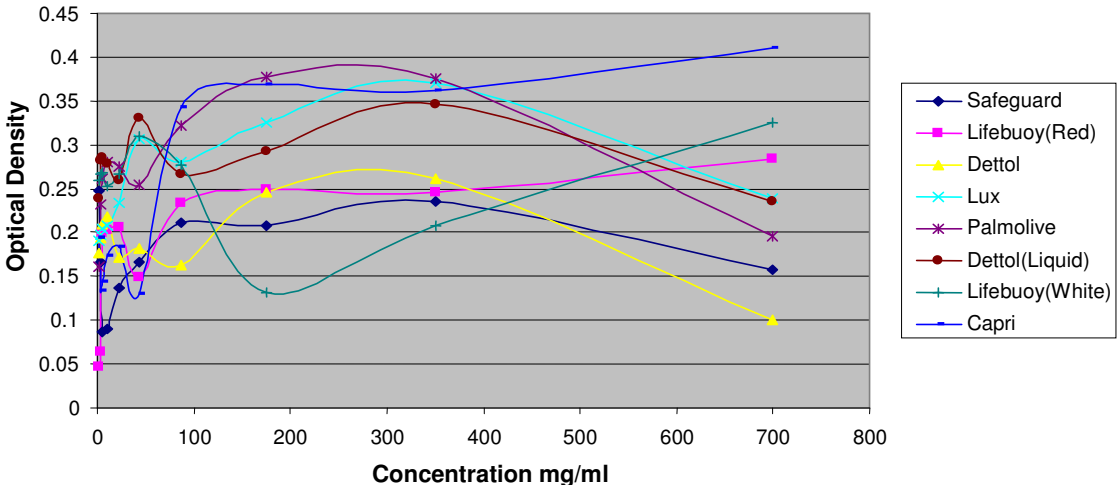

Figure 1. Antibacterial activity of different soaps against *Pseudomonas aeroginosa.*

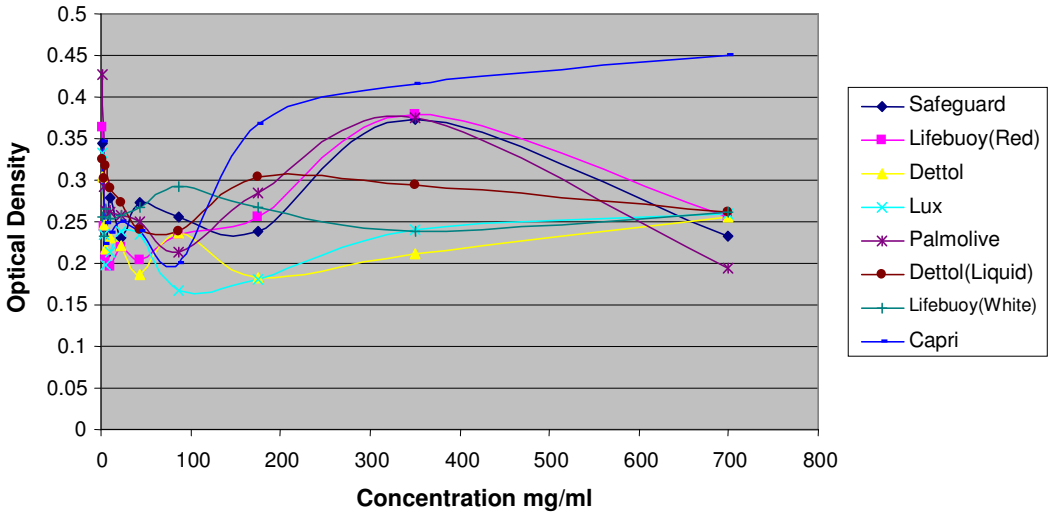

Figure 2. Antibacterial activity of different soaps against *Klebsiella pneumoniae.*

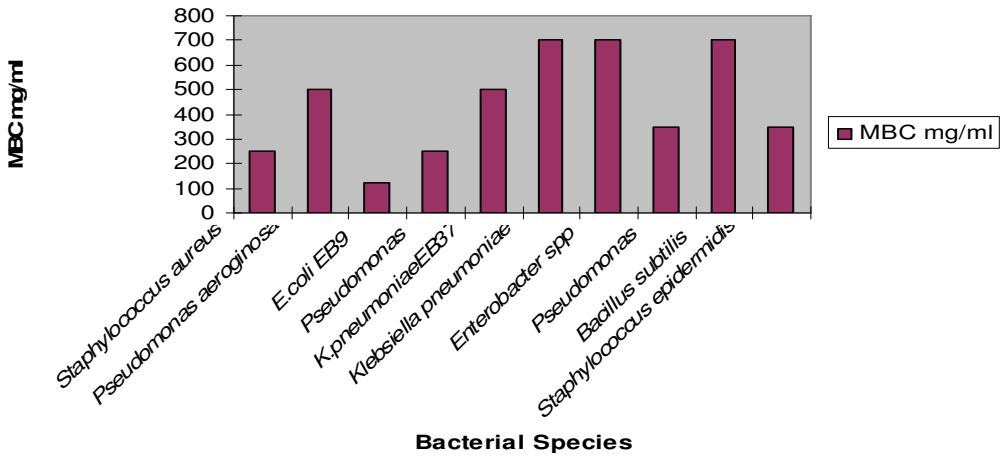

Figure 3. Safeguard soap activity against different bacteria species.

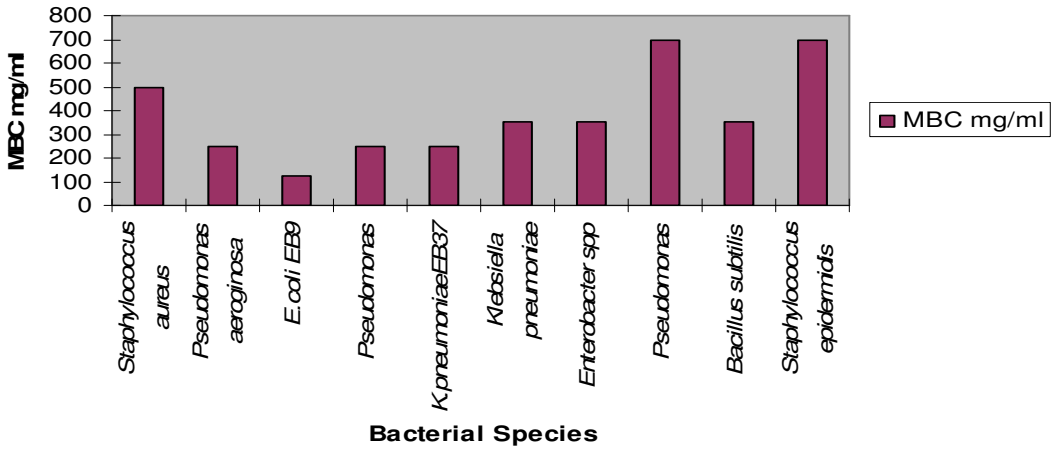

Figure 4. Lifebuoy (White) soap activity against different bacteria species.

Table 2. Characteristics of the bacterial strains.

Test	Bacteria									
	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**
Oxidase	NA	+ve	-ve	+ve	-ve	-ve	-ve	+ve	+ve	-ve
Catalase	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve
Motility	NA	+ve	+ve	+ve	-ve	-ve	+ve	+ve	+ve	-ve
Lactose	NA	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve
EMB	NA	NA	+ve	NA	-ve	-ve	-ve	NA	NA	NA
Indole	NA	-ve	+ve	-ve	-ve	-ve	-ve	-ve	NA	NA
Citrate	NA	+ve	-ve	+ve	+ve	+ve	+ve	+ve	+ve	NA
V P	NA	-ve	-ve	-ve	+ve	+ve	+ve	-ve	NA	NA
M R	NA	+ve	+ve	+ve	-ve	-ve	-ve	+ve	NA	NA
TSI	NA	R/R/-/-	Y/Y/+/-	R/R/-/-	Y/Y/+/-	Y/Y/+/-	Y/Y/+/-	R/R/-/-	NA	NA
Urease	NA	-ve	-ve	-ve	+ve	+ve	-ve	-ve	NA	+ve
Mannitol	+ve	+ve	NA	+ve	NA	NA	NA	+ve	NA	-ve
Maltose	NA	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	+ve
Pigment	Golden	Green	-ve	Blue	-ve	-ve	-ve	Blue	-ve	-ve
Coagulase	+ve	NA	NA	NA	NA	NA	NA	NA	NA	-ve
DNase	+ve	NA	NA	NA	NA	NA	NA	NA	NA	NA
Sucrose	+ve	-ve	-ve	-ve	+ve	+ve	-ve	-ve	+ve	+ve
Genera	*S.aureu*	*P. aerog*	*E. coli*	*Ps spp*	*K. neum*	*K.pneu*	*Enter* p.	*P aerog*	*B. ubtilis*	*S. epid*

soap. These bacterial spp. showed resistance to the soap at a very low concentration and were killed at very high concentration. *Staphylococcus epidermidis* was killed at 175 mg/ml that is very low concentration of soap and it showed its MIC at 87.5 mg/ml, the antibacterial agent proved to be efficient against this bacterium but this soap also killed other bacteria like *Enterobacter, B. subtilis* at 350 mg/ml and there was no apparent growth observed on the nutrient agar plate (Figure 4). Lux is a beauty soap and was used against *S. aureus* to check its antibacterial activity. Lux contains *Aloe vera* which might have antibacterial activity. At 500 mg/ml, the Lux killed the bacterium but concentration was high as compared to the antibacterial soap. The MBC was observed at 500 mg/ml and MIC of the soap was 250 mg/ml. This revealed that Lux soap also showed antibacterial activity but not as much, than the other specific antibacterial soaps. It might also be possible that *S. aureus* was sensitive to the Lux soap. This soap showed its MBC at 700 mg/ml and it also showed its MIC at 350 mg/ml against *Bacillus subtilis.* This soap showed its MBC at 250 mg/ml and MIC at 125 mg/ml against *Pseudomonas aeroginosa* 1 that was almost equal to the Lifebuoy red, it might be possible that some natural ingredients such as extract of *A. vera* show-

ed the antibacterial activity. The observation of Lux beauty soap revealed that these might posses' germicidal activity. Palmolive beauty soap: It is beauty soap and was used against *S. aureus*. Although this was not an antibacterial soap but it showed it's MBC at 500 mg/ml and its MIC was observed at 250 mg/ml. This showed Palmolive soap can kill bacteria. At 500 mg/ml the organism did not showed growth on the surface of the nutrient agar medium. This soap showed its MBC at 700 mg/ml and MIC was observed at 350 mg/ml against *Bacillus subtilis* this is equal or almost equal to the Lux beauty soap but it is very high in comparison to the Lifebuoy both red and white this might be possible that due to lack of specific antibacterial agent it did not show its MBC and MIC at low concentration. Lifebuoy white: It also showed the MBC at 350 mg/ml and MIC at 175mg/ml against *B. subtilis*. These values were equal or almost equal to the Lifebuoy red soap and lower than the Dettol and Safeguard soap at 350 mg/ml there was complete absence of bacterial growth on the agar plates and at 175 mg/ml there was no growth. After 24 h of incubation, few colonies were observed. The MBC of soap against *E. coli* was observed at 125 mg/ml and MIC was 62.5 mg/ml that is very low concentration of soap. The *E. coli* showed sensitivity for this antibacterial soap as it was killed at very low concentration. The antibacterial agent of Lifebuoy white soap might be efficient in killing the cells. For *Pseudomonas* and *K. pneumoniae*, the soap showed its MBC at 250 mg/ml and MIC was observed at 125 mg/ml. As this soap showed the MBC at 250 mg/ml and at this concentration no growth of the bacteria was observed. So, the soap is efficient in killing the bacterium at this concentration. For *K. pneumoniae, Enterobacter* spp. and *B. subtilis,* the MBC was seen at 350 mg/ml and MIC was observed at 175 mg/ml. Lifebuoy showed its MBC for *S. epidermidis* at 700 mg/ml and MIC was seen at 350 mg/ml. The organism was isolated from human skin and found Gram positive but it was killed at 750 mg/ml concentration of soap that was very high.

DISCUSSION

Soaps are generally used for the removal of germs and for cleaning purpose. Soaps usage is very common and now a day's especially antibacterial soaps are very popular. According to company's claim their antibacterial soaps are bacteria killers. So with the use of antibacterial soaps we can get dual functions, removal as well as killing of bacteria. The purpose of the study was to determine the bactericidal activity of both the antibacterial as well as of the beauty soaps being used in our daily life. Antibacterial soaps considered to be more effective than beauty (plain) soaps and deodorant (Toshima et al., 2001). This study suggests that antiseptic soaps were more effective against Gram-negative and Gram-positive bacteria than were plain soaps. Present work showed that plain soaps also possessed antibacterial activity although lesser than that of antibacterial soaps. Garner and Favero (1985) studied the hand washing with plain soaps removes millions of microorganisms (Garner and Favero, 1985). Most of the research has been focused on hand washing and hand disinfectants for personnel in health care settings where patients are immune compromised and are at high risk. Bannan and Judge (1965) indicated that hand washing with bar soap reduced bacterial population (Bannan and Judge, 1965). Tierno (1999) response to the Association for Professionals in Infection Control and Epidemiology (APIC) emphasized the use of antimicrobial household products (Tierno, 1999).

It was seen clearly that Gram positive bacteria were killed at low concentration of soaps than Gram negative bacteria. The best of all the soaps used is lifebuoy white (antibacterial) because the calculation of the efficiency of all the soaps revealed that this soap is more efficient than the others used. The most resistant bacterium of all the soaps is *K. pneumoniae* following *P. aeruginosa*. It is proved experimentally that antibacterial soaps kill the bacteria at a specific concentration; they also have bacteristatic activity and can inhibit the growth of bacteria. Beauty soaps contain some natural and plant extracted ingredients in their composition which have the ability to inhibit or kill the bacteria so they also gave some bactericidal activity. Micro-titration plate method is efficient than tube method and easier to perform. This study suggests that selection of soaps should depend on to the working environment. The soap should have good ingredients which have the ability to kill bacteria but not to damage body tissues. Health care workers should use soaps according to criteria of Health and Hygiene. In this way many immuno-compromised or low immunity patients can be protected from transfer of pathogenic or opportunistic pathogens. This area of research requires attention of scientists and people from soap industry, because quality of soaps is very important as they are the need of every home.

REFERENCES

Aulet de Saab (2001). A Comparative Study of Preservation and Storage of *Haemophilus influenzae* Mem Inst Oswaldo Cruz, Rio de Janeiro, 4: 583-586,

Bannan EA, Judge LF (1965). Bacteriological studies relating to hand washing. The inability of soap bars to transmit bacteria, Am. J. Public Health, p. 55.

Cappuccino GJ, Sherman N (1992). Microbiology: A Lab. Manu. 3: 248-251.

Cheesbrough M (2001). District Laboratory Practice in Tropical Countries, Part 2. Cambridge University Press, Cambridge.

Fluit AC, Schmitz FJ, Verhoef J (2001). Frequency and isolation of pathogens from bloodstream, nosocomial pneumonia, skin and soft tissue, and urinary tract infections occurring in European patients. Eur. J. Clin. Microbiol. Infect. 20: 188-191.

Friedman M, Wolf R (1996). Chemistry of soaps and detergents: various types of commercial products and their ingredients. Clin. Dermatol. 14: 7-13.

Garner JS, Favero MS (1985). Guidlines for Hand Washing and Hospi-

tal Environmental Control" NTIS United States, Department of Commerce, Springfeild, pp. 110-115.
Hand-washing Liason Group (1999). Handwashing. A modest measure – with big effects. Br. Med. J. 318-686.
Higaki S, Kitagawa T, Kagoura, M, Morohashi M, Yamagishi T (2000) Predominant *Staphylococcus aureus* isolated from various skin diseases. J. Int. Med. Res. 28: 87- 190.
Horton R, Parker L (2002). Informed Infection Control Practice. Churchill Livingstone, Second Edition. London.
Johnson SA, Goddard PA, Iliffe C, Timmins B, Rickard AH, Robson G, Handley PS (2002). Comparative susceptibility of resident and transient hand bacteria to para-chloro-meta-xylenol and triclosan. J. Appl. Microbiol. 93: 336-344.
Larson E, McGinley KJ, Grove GL, Leyden JJ, Talbot GH (1986). Physiologic, microbiologic and seasonal effects of handwashing on the skin of health-care personnel. Am. J. Infect. Contr. 14: 51-90.
Lucet JC, Rigaud MP, Mentre F, Kassis N, Deblangy C, Andremont A, Bouvet E (2002). mination before and after different hygine techniques: a randomized clinical trial. J. Hosp. Infect. 50: 276-280.
Osborne RC, Grube J (1982). Hand disinfection in dental practice, J. Clin. Prev. Dent. 4: 11-15.
Richards MJ, Edwards JR, Culver DH, Gaynes RP (1999). Nosocomial infections in medical intensive care units in the United States. National Nosocomial Infections Surveillance System. Crit. Care Med. 27: 887-892.
Tierno PM (1999). Efficacy of triclosan, Am. J. Infect. Contr. 27: 71-72.
Toshima Y, Ojima M, Yamada H, Mori H, Tonomura M, Hioki Y, Koya E (2001). Observation of everyday hand-washing behavior of Japanese and effect of antibacterial soap, Int. J. Food Microbiol. 68: 83-91.