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Examination of Microbial Flora Present in Gel Based Cosmetic Creams

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Abstract:
This work was aimed to determine the microbial quality of gel based cosmetic creams. Samples were collected from the local market of Gwalior. Staphylococcus aureus, Pseudomonas aeruginosa, Clostridium tetani and Candida albicans were specifically targeted. A total of 60 samples; 20 each of three different brands was examined. The mean aerobic plate counts obtained were 1.6 x 10^4 cfu/g, 2.3 x 10^4 cfu/g and 4.5 x 10^5 cfu/g while the mean yeast and mould counts were 1.1x 104 cfu/g, 1.4 X 104 cfu/g and 2.7 X 104 cfu/g. Thirty (50 %) of the samples analyzed were contaminated with Staphylococcus aureus, twelve (20 %) were contaminated with Clostridium tetani and four (7 %) were contaminated with Candida albicans. Bacillus sp. was also isolated from four (7 %) samples while Pseudomonas aeruginosa was not isolated from any of the samples analyzed. Aspergillus niger, Apergillus fumigatus, moulds were isolated from the creams.

Keywords: *Microbiological quality assessment, gel based cosmetic creams*

1. Introduction

The microorganisms have the ability to grow and reproduce in cosmetic products also. Microorganisms may cause spoilage or chemical changes in cosmetic products. Cosmetic products are used for various dermatological purposes (MA Duke, 1978). A gel based cosmetic product are used by both men and women and these cosmetics also inhibits the bacterial growth, which may cause unpleasant odours and sometimes skin infections (MA Duke, 1978). Many ingredients such as zinc oxide, titanium dioxide, essential oils are added to provide good effects (R Josh, 2006). Gel based cosmetic creams comes as a packaged container (MSE Ashour et al., 2008). Cosmetic creams have positive effects on a person's appearance which include; reducing wrinkles and puffiness; it hides the blemishes and the dark circles, also helps in reducing the fine lines and provides beautiful and clean appearances to the skin (T Stabile 1984). It was observed that some cosmetic creams are contaminated with moulds and other microorganisms (B Elane, 1989). Cosmetic can contaminated with micro-organisms such as Staphylococcus aureus, Psuedomonas aeruginosa, Clostridium tetani, yeasts and moulds, which can either be from the raw materials or during manufacturing, processing, breakage or damage of the cosmetic container, at the retail market due to the presence of dust, may also be during usage of product (M Pollack 2000) Contamination of cosmetics with micro-organisms such as Clostridium tetani, Staphylococcus aureus, moulds and yeast may cause skin problems (M Pollack 2000). **C**osmetics can be contaminated with micro-organisms when they are not preserved properly (J MA Duke, 1978).

2. Materials and Methods

2.1. Sample Collection

A total of 60 samples, 20 each of three different types of gel based cosmetic creams marked as: sample A, sample B and sample C, were purchased from different areas of Gwalior and transported to the laboratory.

2.2. Media Used

Blood agar was used for the isolation of Clostridium tetani and Pseudomonas aeruginosa. Mannitol salt agar was used for the isolation of Staphylococcus aureus, Plate count agar (PCA) was used to determine the bacterial count present in the cosmetic creams and Sabauroud's Dextrose agar (SDA) was used for the isolation and enumeration of yeasts and moulds. All of the media mentioned above were prepared under sterile conditions.

2.3. Aerobic Plate Count of the Cosmetic Creams

A stock sample of each cosmetic cream was prepared. A fivefold serial dilution was made and aliquots of the last two dilutions were inoculated on Plate Count Agar (PCA) in duplicates using the pour plate method. All the plates were incubated at 37^0C for 24 hours, followed by colony count. Results were expressed as colony forming unit per gram (cfu/gm).

2.4. Yeasts and Moulds Count of the Cosmetic Creams

One ml of the last two dilutions mentioned in the above prepared were inoculated on SDA plates using pour plate method. The plates were then incubated at 25^0C for 24 hours. Colonies were counted after three days. Results of colony count were expressed as yeasts and moulds counts per gram.

2.5. Identification of Bacterial Isolates

All bacterial isolates were identified based on their Gram reaction and biochemical reactions as described by U.S.FDA manual online, 2001.

2.6. Identification of Fungal Isolates

All fungal isolates were identified based on their macroscopic and microscopic appearance with reference to manuals of Barnett and Hunter, (D Ellis, 2006).

3. Results

The results obtained show that the bacterial load of cosmetic creams A ranges from $4.6X10^3$ to 1.3X104 cfu/g with a mean bacterial load of $4.5 X 10^5$ cfu/gm. The bacterial load of cosmetic creams B ranges from $4.6 X 10^3$ to $1.1 X 10^4$ cfu/gm with a mean bacterial load of $2.3 X 10^4$ cfu/gm while that of cosmetic creams C ranges from $4.2 X 10^3$ to $1.3X10^4$ cfu/g with a mean bacterial load of $1.6 X 10^4$ cfu/gm (Table 1). Cosmetic creams C had the highest mean count of yeasts and moulds of $2.7 X 10^4$ cfu/gm followed by cosmetic creams B with a mean count of 1.4 X 104 cfu/gm and the least was Cosmetic creams A with a mean count of $1.1 X 10^4$ cfu/ml (Table 2).

Out of the 60 samples of cosmetic creams that were analyzed Staphylococcus aureus was isolated from 30 (50 %) samples, Clostridium tetani was isolated from 24 (20 %) samples, Candida albicans was isolated from 8 (7 %) samples (Table 3). Moulds such as Aspergillus niger, Penicillium sp, Aspergillus fumigatus, Rhizopus oligosporus, Mucor plumbeus, Fusarium sp were isolated from the cosmetic creams with Aspergillus niger, having the highest frequency of occurrence of 14 (47 %), followed by Rhizopus oligosporus of 10 (17 %) samples and the least being Aspergillus fumigatus, Mucor plumbeus and Penicillium sp which were isolated from two samples each (Table 4).

Cosmetic Creams	Range of APC	Mean
A	$1.3 X 10^4 – 4.6 X 10^3$	$4.5X10^5$
B	$1.1 X 10^4 – 2.4 X 10^4$	$2.3X10^4$
C	$1.3 X 10^4 – 2.6 X 10^4$	$1.6X10^4$

Table 1: Aerobic Plate Count (APC) of the Cosmetic creams (cfu/gm)
Total Mean 1.6 X 10^5

Other organisms isolated from the cosmetic creams were Bacillus species from 4 (7 %) samples; Pseudomonas aeruginosa which was one of the target organisms was not isolated from any sample.

Cosmetic Creams	Range of APC	Mean
A	$0.0 – 2.0 X 10^4$	$1.1 X 10^4$
B	$3.8 X 10^3 – 2.6 X 10^4$	$1.4 X 10^4$
C	$3.5 X 10^3 – 2.6 X 10^4$	$2.7 X 10^4$

Table 2: Yeasts and Moulds Count of the Cosmetic Creams (cfu/gm)
Total Mean 1.7 X 10^4

Sample No.	Analyzed	No. positive for *S. aureus*	No. positive for *Cl. tetani*	No. positive for *C. albicans*
A	20	12 (60%)	2 (10%)	2 (10%)
B	20	8 (40%)	4 (20%)	-
C	20	10 (50%)	6 (30%)	4 (10%)
Total	60	30 (50%)	12 (20%)	4 (20%)

Table 3: Frequency of the Occurrence of the Target Organisms in the Cosmetic Creams

4. Discussion

On the basis of facts, examined cosmetic creams were found more contaminated with fungal growth than with bacterial growth. In a similar study, (S Hashim, 2003, L Nasser 2008) also reported higher bacterial contamination than fungal. Fungal and bacterial contaminants in unused cosmetic creams were found to be similar for the reason that of the environment in which the creams are manufactured, packed and the ingredients themselves (L Nasser 2008). The guidelines on the microbiological quality of finished cosmetic products have defined cosmetic creams into two categories; Category 1 Cosmetic creams specifically intended for children under 3 years and Category 2 for other cosmetic creams (AOAC International, 2002). The limit for cosmetic creams classified in category 1 is that viable count for aerobic mesophillic micro-organisms should not be more than 10^2 cfu/ml or g in 0.5 gm or mill liter of the creams (AOAC International, 2001). Based on these limits, the aerobic plate count of all cosmetic creams B (a baby creams) analyzed were above the acceptable limit with a total mean count of 2.3 X 10^4 cfu/gm. The high count may be due to environmental contamination during mining or processing of the talc (main ingredient) used or during manufacturing of the baby creams itself. The limit for cosmetic creams classified in category 2 is that the total viable count for aerobic mesophillic micro-organism should not be more than 103 cfu/gm or ml in 1 gm or millitre of the product (AOAC International. **2001**). Based on this information, the aerobic plate count of both cosmetic creams A and B were above the acceptable limiting which also agrees with the findings of Ashour et al., (MSE Ashour, 2008). The isolation of Staphylococcus aureus as the most predominant contaminant tallies with the findings of Ashour et al. (MSE Ashour, 2008). Clostridium tetani was isolated from 20% of the cosmetic creams analysed which also agrees with the findings of Ashour et al. (MSE Ashour, 2008) who also reported the isolation of Clostridium spp. in cosmetic creams. The presence of the organism poses a serious danger to the user, because the tetanus toxin produce by the organism is lethal to human (at dose of approximately 1 ng.kg) especially neonatal cases. A serious tetanus neonatorum outbreak from talcum creams contaminated with Clostridium tetani in New Zealand was reported by Brazier et al. The organism gain entrance into the body through cuts in the skin thereby causing infection. The organism is an inhabitant of the soil, which may contaminate the main raw material (talc) of the talcum cosmetic, creams (K Todar, 2005). The isolation of Candida albicans from the cosmetic creams though at low frequency is also of concern because when these creams contaminated with Candida albicans are used on genital areas and sanitary napkins it may lead to vaginal candidiasis and also oral candidiasis when the creams mistakenly gets into the mouth of the user (AD Hitchin, 2001). Contamination of cosmetic creams may derive from a variety of sources such as raw materials, manufacturing, storage and packaging or use. Cosmetic creams are not expected to be aseptic; however, they must be completely free of high – virulence microbial pathogens, and the total number of aerobic microorganisms per gram must be low (AD Hitchin, 2001, S Hashim, 2003).

5. Conclusion

It can be concluded from the findings of this research work that all the cosmetic creams analyzed are of poor microbiological quality since their bacterial counts are found to be higher than the limit. The presence of organisms such as Clostridium tetani, Staphylococcus aureus and Candida albicans in the cosmetic creams implies that they can serve as vehicles for the transmission of these pathogenic organisms.

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