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Research Article

Should we be Concerned about Non Sterile Pharmaceutical Products?

Mariana Nunes de Menezes[1], Loren Fernanda Ghidini[2], Ana Carolina Kogawa[2]* and Hérida Regina Nunes Salgado[2]

[1]Pontifícia Universidade Católica do Rio Grande do Sul - PUCRS, Porto Alegre, Brazil

[2]São Paulo State University (UNESP), School of Pharmaceutical Sciences School, Campus Araraquara, Araraquara, SP, Brazil

***Corresponding Author**: Ana Carolina Kogawa, São Paulo State University (UNESP), School of Pharmaceutical Sciences School, Campus Araraquara, Araraquara, SP, Brazil.

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Abstract

Many pharmaceutical products commercialized are not required to be sterile, such as creams, gels and shampoos. The microbiological quality of these products is ensured by Microbiological Examination of Nonsterile Products described in the official pharmacopeias. They determine the total number of microorganisms present and the presence of pathogens such as *Salmonella* sp, *Escherichia coli*, *Staphylococcus aureus* and *Pseudomonas aeruginosa*. Nonsterile products They can present microbiological contamination due to poor hygiene in manufacturing and/or poor stability of the formulation constituents. This work shows the microbiological analysis of 81 samples of non-sterile products. 9.88% were disapproved due to the total count of bacteria, molds and yeasts above the specification. Thus, non-sterile products can still be found in the market despite the dissemination of legislation that discusses Good Laboratory Practices and Good Manipulation Practices.

Keywords: *Non-Sterile Products; Total Number of Micro-Organisms; Pathogenic Micro-Organisms; Good Laboratory Practices; Good Manipulation Practices*

Introduction

The increased consumption power of the classes C and D additionally the industry investment in innovation and technology, more accessible prices reached the market and consumption of non-sterile products such as creams, gels and shampoos increased significantly.

Non-sterile products are those in which the presence of microbial charge is conceptually admitted, although limited, taking into account the characteristics of its use, as is the case of topical and oral cosmetic and pharmaceutical products, which come into contact with areas bearing natural microbial flora [1].

Due to its aqueous composition, care must be taken from its manipulation to the daily handling, during its use, because of the components that can favor microbial development.

Attention in the quality control of non-sterile products ensures that the microbial charge present in the product does not compromise the final quality or patient safety, either in the qualitative or quantitative aspect, that in ordinary conditions of use will have contact with areas bearing natural microbial flora, composed of high numbers of saprophytes [2-5].

Like many other pharmaceutical products, the gels, creams and shampoos come into contact with areas bearing natural microbial flora, being of extreme importance the control for microbial charge present aiming at the final quality of the product and the patient well-being [6-9].

Various types of microorganisms could be present, such as bacteria, molds, yeasts coming from several sources of contamination, such as raw materials, adjuvants and water, besides the environment, the conditions of preparation and manipulation, storage and, later, for use by users.

In this context and with the concern of the quality of non-sterile products, this study aimed to determine the total number of microorganisms present in non-sterile preparations and the presence of pathogens such as *Salmonella* sp, *Escherichia coli, Staphylococcus aureus* and *Pseudomonas aeruginosa* in 81 commercial samples.

Experimental

The samples were analyzed by the method present in the official compendium [10] and described in figure 1.

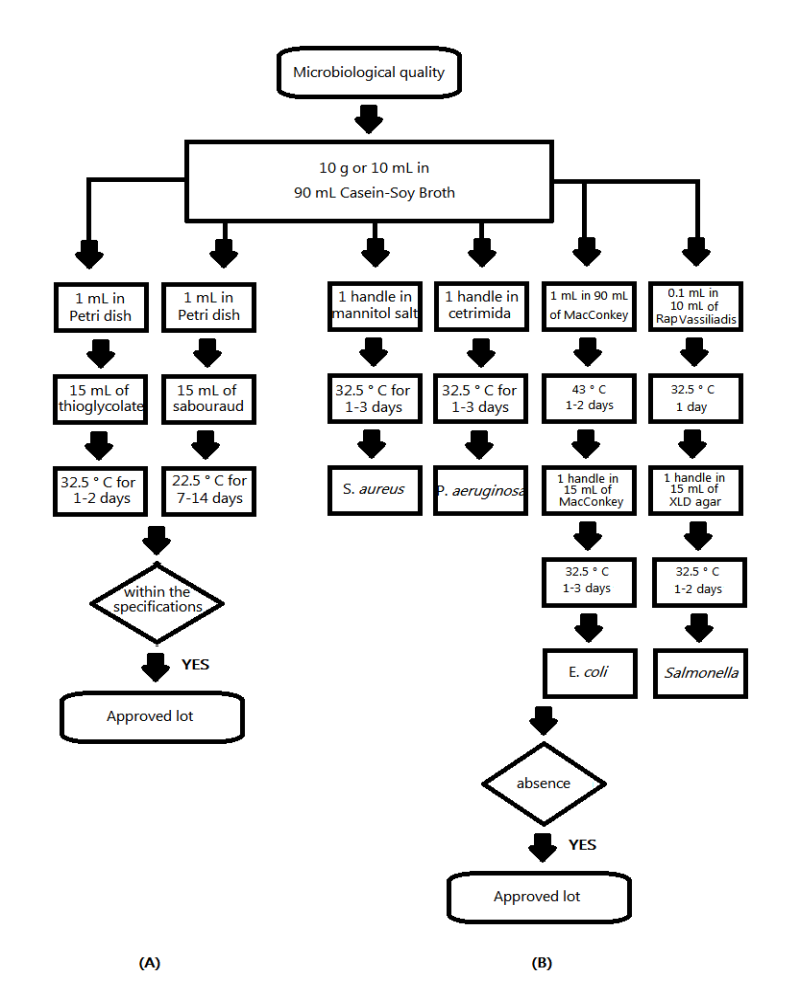

Figure 1: *Microbiological evaluation of non-sterile products by (A) total number of microorganisms using the Pour Plate technique and (B) presence of pathogens such as Salmonella sp, Escherichia coli, Staphylococus aureus and Pseudomonas aeruginosa.*

Material and Method

Material

81 samples of gels (40 units, 49.38%), creams (32 units, 39.51%), sunscreen (5 units, 6.17%), syrup (2 units, 2.47%) and shampoos (2 units, 2.47%) were collected from 5 pharmacies in São Paulo State, Brazil.

Method

Total number of microorganisms

External disinfection of the packages was performed using 70% ethanol.

10g of each sample was transferred to erlenmeyer containing 90 mL of sterile Casein-soy broth.

After homogenization of the sample in the Casoy broth, 1 mL of the dilution was transferred to sterile Petri plate.

Thioglycollate agar or Sabouraud agar was added at the temperature of 46°C ± 1°C according to the Pour Plate technique.

After agar solidification, the plates were incubated at 32.5°C ± 1°C for 2 days and at 22.5°C ± 1°C for 14 days, to verify the development of bacterial and fungal colonies, respectively.

The assay was performed in triplicate.

Presence of pathogens

The sample with Casoy broth was incubated at 32.5°C for 24 hours for the verification of the *Salmonella* sp., *Escherichia coli, Staphylococcus aureus and Pseudomonas aeruginosa*.

Search for *Salmonella* sp

After 24 hours, 0.1 mL of the incubated broth was transferred to the Rappaport Vassiliadis broth which was incubated at 32.5°C for 24 hours. After this period, a handle was transferred to XLD agar which was incubated at 32.5°C for 24 hours.

Search for *Escherichia coli*

After 24 hours, 1 mL of the incubated broth was transferred to 90 mL of MacConkey broth which was incubated at 32.5°C for 24 hours. After this period, a handle was seeded on EMB agar which was incubated at 32.5°C for 24 hours.

Search for *Staphylococcus aureus*

After 24 hours, a handle of incubated broth was transferred to Johnson mannitol salt agar which was incubated at 32.5°C for 24 hours.

Search for *Pseudomonas aeruginosa*

After 24 hours, a handle of the incubated broth was transferred to cetrimide agar which was incubated at 32.5°C for 24 hours.

The tests were carried out with strict care using positive and negative controls, guaranteeing the viability and the sterility of the media and the processes and avoiding doubtful results.

The results were evaluated according to microbiological specifications.

Results and Discussion

The official tests described in the literature can indicate the absence or limited occurrence of specified microorganisms which can be counted [10]. These tests are designed primarily to determine if a product complies with the established specification for microbiological quality.

Nonsterile Products can accept a number of organisms and absence of pathogenic organisms according Microbial Limits Test, described in guidelines and official literature [11]. In the last years there was the harmonization of this test by pharmacopeias such as American, British, European, Japanese and Brazilian Pharmacopoeias [10-14].

From the total of 81 samples, 7 gels samples (8.64%) were reproved for total bacterial count above the specification and 1 cream sample (1.23%) did not meet the specifications for the presence of molds and yeasts. 73 samples (90.12%) were approved indicating the importance of quality control to ensure the microbiological safety of a non-sterile product for the patient and consumer.

Figure 2 shows the results of the microbiological analyzes of the 81 commercial samples.

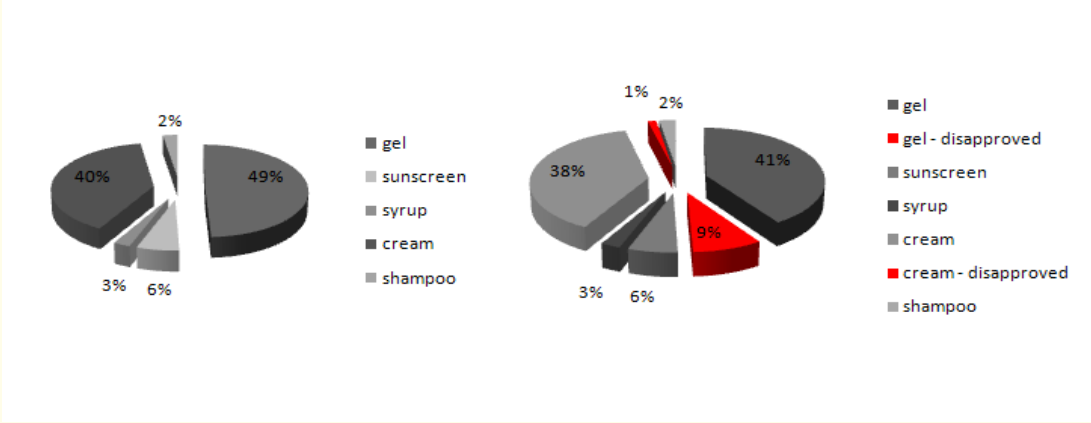

Figure 2: *Results of the samples analyzed according to Microbiological Test for Non-Sterile Products. (A) Samples before analysis (B) Status of samples after analysis.*

The control of microbial contamination of non-sterile products has fundamental importance during the development of a formulation as well as throughout the storage period. The low microbial quality can occur mainly due to the microbial contamination coming from the lack of hygiene in the manufacture and the low stability of the constituents of the formulation.

Therefore, the use of appropriate personal protective equipment such as lab coat, mask, cap, gloves are recommended as well as correct washing of the hands, face and forearms immediately before the operators correct wearing to prepare the material. In addition, the use of laminar flow chamber is also advocated.

This test is suitable for the qualification of non-sterile products. It has basic characteristics such as ease of handling, fast execution and low cost of material and solvents used.

Based on the results of the 81 samples, it is possible to conclude that the handling of non-sterile products is still not performed in a careful and clean way, since 9.88% of the samples were microbiologically reproved.

More attention should be given to the manipulation of gels, in which almost 9% of the samples were reproved. On the other hand, the samples of sunscreens, syrups and shampoos analyzed were 100% microbiologically approved and only 1 cream sample was disapproved.

Conclusion

The total number of microorganisms and the investigation of the presence of *Salmonella* sp., *Escherichia coli*, *Staphylococcus aureus* and *Pseudomonas aeruginosa* allow the evaluation of the microbiological quality of non-sterile products.

The analysis of non-sterile products presented can be easily adopted and routinely used for quality control in manipulation pharmacy and research laboratories, a fact that is in line with the concept of quality in the production of pharmaceuticals and the requirements demanded in the Good Laboratory Practices and Good Manipulation Practices.

However, non-sterile products can still be found in the manipulation pharmacy. In them the Good Laboratory Practices and Good Manipulation Practices, as well as the concern with the quality of its products unfortunately is marginalized. Then, in this case, the reliability and history of the manipulation pharmacy come into play.

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Declaration of Interest

The authors report no declarations of interest.

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