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The impact of the new European Cosmetic Regulation on the preservative system

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All around the world, men and women use cosmetic products to promote "cleaning, perfuming, changing appearance, protecting, keeping in good condition or correcting odors" (1) of their "skin, hair, nails, lips or teeth" (1). They must not affect cell structure or cell function, and more than anything, be safe of use for consumers. This is why manufacturers have been using preservatives to assure a good microbiological protection of cosmetic products, when it is necessary, during their life cycle. A chemical microbiological system is not required for some product categories (i.e., product with more than 25% alcohol, product with a water activity <0.6, and anhydrous products).

Several preservatives allowed for cosmetics are available in the market but a small number are frequently used (2). Their efficacy is tested by challenge test (CT), allowing to determine the product stability. It consists of an intentional contamination of a cosmetic product with a fixed inoculum of microorganism. Counts are made at different times to follow the evolution of the contamination and observe the neutralization of microbial growth. Results of this type of test are subject to the *European Pharmacopoeia* EP 5.1.3, *US Pharmacopoeia*—USP51, French standard AFNOR NF T75-611 (3), or SOP (Standard Operating Procedure). A new regulation (CE) No-1223/2009 imposes the use of CT for cosmetic products. But it is not always scientifically necessary. Therefore, this step could be avoided in some cases, when justification such as water activity, percentage of alcohol, pH, manufacturing condition, packaging, etc., has been made. This may save some time and cost within the research and development process, without impacting the protection of health of the consumer.

According to the French standard NF T75-611, some manufacturers may be dispensed with this test on cosmetic products for which a microbiological risk control has been demonstrated (4). For others, they would be easier to rely on factors known and easily available as pH, water activity percentage, or preservative formulation to determine if a CT is useful or not.

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Three factors are essential for microbial growth and their interaction: moisture, temperature, and nutrient. The microorganisms are able to attack and decompose all materials and represent a significant issue for industrialists. These damages can cause material degradation with financial consequences, impairment of reputation, and danger for human health. To avoid this scenario, industrialists have recourse to the use of preservatives routinely. As such, the French standard NF ISO 29621 defines these factors to achieve the first step in microbiological risk assessment, as follows: product composition (water activity [a_w], pH, alcohol content, and raw materials), fabrication conditions, and packaging and factor combination (effect of pH, water activity, alcohol content, etc.).

So, the prediction of a preservation is not mathematical since manufacturers cannot assume the microbiological protection as the more preservatives, the more the formula is protected. This cannot be true. They have to deal with three problems: the product security, product formulation and, not the least, consumer security. Manufacturers have a wide selection of preservatives (5–11), but they must still take into account certain factors such as product type (anhydrous or aqueous product), the population affected by this product, its composition (pH, water activity, surfactants, alcohols, chelating agents, fragrance materials, etc.), and if certain raw materials lead to difficulties in formulating, etc. Antagonism phenomena can occur between the preservatives and other raw materials present in the formula and should be seriously considered for product formulation. Finally, the association of some preservatives may have toxicological effects (12,13). For example, a formulation containing sodium benzoate (0.45%) and potassium sorbate (0.225%) has led to reactions of redness, tingling, and warming of the skin of some subjects. Thus, preservation quality needs to comply with toxicological impacts and finally with the overall consumer security.

The implementation of the upcoming cosmetic regulation (CE) No-1223/2009 forces the investigation of microbiological safety based on formulation criteria and on the realization or not of a CT. For example, the analysis of specific factors mentioned above such as water activity. The packaging is also a criterion that has to be taken into account with the format and mode of administration since the interaction between container content can significantly impact the preservation system. Some difficulties have to be cleared within the process or about data information. Fabrication conditions should follow the good manufacturing practice (GMP) but information is not always given to the safety assessor on this subject. Moreover, the link between GMP and CT decisions is not clear or standardized. Finally, the most difficult to grasp is the combination of factors, their interaction, and consequences. In this context, the European regulation requires microbiological risk assessment of a cosmetic product before its marketing by using the standard technique CT. The regulations governing implementation of the CT can be discussed. Indeed, presentation of results and their format are not standardized. European Pharmacopoeia, French standard AFNOR NF T75 611, and some SOP are used and develop acceptance criteria to validate a CT but none gives a frame to follow in presenting the results. Laboratories performing these tests may therefore present their results in different forms causing a bias in their interpretation. More importantly, the protocol SOP 98 varies completely from the other protocol given in the standard NF T75-611 and European Pharmacopoeia. Indeed, more than five strains are used, mixed, and inoculated into the tested product. The results are presented as an average per strain type (bacteria, yeasts, molds). Moreover, the manipulative effect on this test is important.

All these points lead to an approximation in the exploitation of results. In this cosmetic regulatory context, the use of CT can be avoided if a microbiological risk assessment has

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Table I
Comparaison of Acceptance Criteria

	USP[1] (log reduction)		**EP**[2] (log reduction)				**AFNOR NF T75-611** (log reduction)					
			Bacteria		Fungal and mold		Bacteria		*C. albicans*		*A. brasiliensis*	
Time	Bacteria	Fungal and mold	A	B	A	B	A	B	A	B	A	B
6 h	NT[3]	NT	2	NT	NT	NT	NT	NT	NT	NT	NT	NT
24 h	NT	NT	3	1	NT	NT	NT	NT	NT	NT	NT	NT
7 days	1	NI	NT	3	2	NT	≥3	NT	≥1	NT	NT	NT
14 days	3	NI	NT	NT	NT	1	≥3 et NI	≥3	≥1et NI	≥1	≥0	≥0
28 days	NI[4]	NI	NR[5]	NI	NI	NI	≥3 et NI	≥3 et NI	≥1 et NI	≥1 et NI	≥1 et NI	≥0 et NI

USP: United State Pharmacopoeia;
EP: European Pharmacopoeia;
NT: Not tested;
NI: No increase of microorganisms compared to the previous time; NR: No recovery.

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been demonstrated. The French standard NF ISO 11930:2012, "Evaluation of the antimicrobial protection of a cosmetic product" will attempt to standardize and reduce these differences of interpretation:

The evaluation of the antimicrobial protection of a cosmetic formulation is based on the inoculation of formulation with calibrated inoculum (prepared from relevant microorganisms strains). The number of surviving microorganisms is measured at predetermined time intervals for 28 days. For each time and each strain, the logarithmic reduction rate is calculated and compared with the values minimum requirements for the assessment criteria A and B (see Appendix B).

When used as a reference method, the procedures must be followed scrupulously to avoid variable results (4).

In conclusion, despite bias in the various regulations governing the implementation of CT, our own research departments showed specific factors as pH and water content (aw) factors that significantly impact the CT results (14). We confirmed that spiking a large number of preservatives without specificity in a formula does not ensure the microbiological safety of a cosmetic product. As antiseptics or antibiotics in pharmaceutical domain, it is necessary to better define the target (bacteria, mold, yeast) based on the physicochemical properties of a product, the way it is used and where it is kept, the action spectrum of preservatives and the possible interactions between the different substances present in the formula to allow a better assessment of the preservative system for product preservation and consumer security.

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