

# Skin Microbiota and Cosmetics Influence

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Dermatological and cosmetics fields have recently started to focus on the human skin microbiome and microbiota, since the skin microbiota is involved in the health and dysbiosis of the skin ecosystem. Amongst the skin microorganisms, *Staphylococcus epidermidis* and *Cutibacterium acnes*, both commensal bacteria, appear as skin microbiota sentinels. These sentinels have a key role in the skin ecosystem since they protect and prevent microbiota disequilibrium by fighting pathogens and participate in skin homeostasis through the production of beneficial bacterial metabolites. These bacteria adapt to changing skin microenvironments and can shift to being opportunistic pathogens, forming biofilms, and thus are involved in common skin dysbiosis, such as acne or atopic dermatitis.

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## 1. Introduction

Research on human microbiota in dermatology started in the 1950's with Kligman, with the improvement in cell culture [1]. In 2008, the National Institutes of Health launched the five-year Human Microbiome Project with the objective to sequence human microbiome components. Emergence of new technologies, such as next-generation sequencing (NGS), enable the comprehensive study of the human microbiome [2][3]. The microbiome corresponds to the set of genomes of microorganisms in symbiosis with the human host, including microorganisms, alive or dead, and free DNA, whereas microbiota refers to microorganisms living in or on a defined ecosystem [4]. Human microbiota, mainly established on the skin, oral and vaginal mucosa, as well as in the respiratory, urinary and gastrointestinal tracts, have fundamental roles in health and diseases [5][6]. The human adult skin microbiota is comprised of diverse microorganisms, including bacteria, fungi [7][8], yeasts, viruses [9], archaea [10] and mites, principally *Demodex* [11].

The skin, the largest organ of the human body, is a complex and dynamic ecosystem. Its outer epidermal layer, the stratum corneum, is the first physical barrier that prevents chemical substances or pathogenic microorganisms' entrance, fluid evaporation and body heat loss [12]. The stratum corneum is composed of 75–80% proteins, mainly keratins and membrane proteins; 5–15% lipids; and 5–10% unidentified compounds [13][14]. For the establishment of this microbiota, the skin provides essential nutrients, such as amino acids from the hydrolysis of proteins, fatty acids from the stratum corneum, sweat, lipid hydrolysis or sebum and lactic acids from sweat [15]. Microorganisms are based on this stratum corneum and on the deeper cutaneous appendages [16][17]. The skin microbiota, made up of millions of commensal microorganisms, e.g., 1 million/cm<sup>2</sup>, is the second largest microbiota of the human body in mass [18]. Cutaneous bacteria belong to four main phyla among the thirty-six known [17]. The average skin

body distribution of these main bacteria phyla, detected on 20 diverse skin sites of 10 healthy individuals, were found to be Actinobacteria at 51.8%, Firmicutes at 24.4%, Proteobacteria at 16.5% and Bacteroidetes at 6.3% [19][20]. Procurement of the skin microbiota occurs in the early stage of birth. In utero, the skin is sterile, devoid of any microorganism, and is colonized a few minutes after birth by commensal microorganisms of the mother, depending on the childbirth method [21][22][23]. This colonization process in the neonatal stage is essential for the establishment of immune tolerance towards commensal microorganisms [1][24]. Microbiota colonization continues during growth until reaching an equilibrium state in adulthood [1].

An increase in scientific investigations into skin microbiota has inevitably led to the emergence of related studies in the cosmetic industry [25], which has now become unavoidable in the cosmetic market. Indeed, skin microbiota is involved in the maintenance of a healthy cutaneous barrier. Skin and microorganisms live in symbiosis and microorganisms help to maintain the skin barrier, the immune system and limit pathogenic microorganism growth [26]. However, an imbalance in skin microbiota, called dysbiosis, is correlated with skin pathological diseases, such as acne with the loss of phylotype diversity of *Cutibacterium acnes* and atopic dermatitis with the increase in pathogenic *Staphylococcus aureus* and commensal *Staphylococcus epidermidis*, as well as with non-pathological diseases, such as sensitive and dry skins [4].

The principal challenges of such research are (1) the comprehension of each microorganism's role in healthy and beautiful skin; (2) the description of the microbiota of weakened skin with the precise determined cause or consequence of the correlation between dysbiosis and skin pathologies; and (3) evaluation studies of the active ingredients' effect on skin microbiota. The maintenance, protection and restoration of microbiota diversity and equilibrium, as well as prevention of skin dysbiosis, are the emerging claims of cosmetic products. These products may include prebiotics, probiotics, post-biotics or active ingredients with demonstrated effects.

## 2. Influence of Cosmetics on Skin Microbiota

Intra- and inter-individual multiplicities in microbiota and chemical composition highlighted in the study of Bouslimani et al. (2015) are a huge challenge for the cosmetics industry and skincare development. Molecules associated with skincare or hygiene products last on the skin after their use despite several washings and these products might alter molecular and bacterial diversity [20].

Cosmetic ingredients used that are either functional ingredients, such as preservatives, oils and emulsifiers, or active ingredients, impact the skin microbiota and require attention. Indeed, conventional skincare or hygiene products such as soap, gel and cream contain preservatives and natural and synthetic chemicals that impact microbiota even if these effects are not fully investigated in detail for now [26][27]. Preservatives, such as phenoxyethanol, parabens, and methylisothiazolinone, are known to inhibit the survival of skin commensal bacteria such as *S. epidermidis* [28]. This alteration phenomenon depends on the residual activity of the preservative in the cutaneous environment [27]. Functional ingredients like oils, emulsifiers, fatty acids, gelling agents, thickeners and basic cosmetics for skincare, which both improve skin hydration, modulate microbiota diversity [29][30][31][32][33].

Dermo-cosmetic companies must undergo studies on skin microbiota while developing new ingredients or products in order to ensure consumers that their products maintain, improve a healthy microbiome, or restore a healthy skin-microbiome balance in case of a disturbed microbiome [34][35][36]. In 2019, the first certification of being “Microbiome-friendly”, set up by “MyMicrobiome”, appeared for a final cosmetic product. This certification is to validate that the product is contamination-free, that specific bacteria of the targeted area will be unharmed, that the microbiome diversity is preserved and that the skin balance is not disturbed (not by the suppression of commensals nor by the stimulation of pathogenic bacteria). For instance, a study in 2018 evaluated three different face washes, two “everyday” products and one 100% natural product, applied on 32 women’s upper volar forearm, for their effects on skin microbiome diversity, along with skin pH, moisture and trans-epidermal water loss (TEWL), washing twice a day for 4 weeks. Volunteers were divided into three groups according to skin characteristics: skin pH acid/normal/alkaline, very dry/dry/moist skin and very healthy/healthy/normal/stressed/critical skins, and each was assigned one product. All groups exhibited an increase in alpha diversity (species richness via operational taxonomic unit count and species diversity via the Shannon index) over time and their skin moved to a “healthier” state. The study suggested that synthetic ingredients modified the microbiota diversity, especially within the first two weeks [33]. A more recent study (2019) tested the ability of *S. epidermidis* to metabolize the functional ingredients commonly found in dermo-cosmetics formulae with native oils and waxes, fatty acid esters, fatty acid alcohols, fatty alcohols ethers, fatty acids and other [31]. The tested substances were formulated in water-in-oil and oil-in-water emulsions at concentrations commonly used in cosmetics—10% for oils, 5% for emulsifiers and 2% for fatty acids; the gelling agents and thickeners did not affect the *in vitro* bacterial growth of commensal *S. epidermidis* [31], and thus are microbiota friendly for this commensal. Future investigations should be performed to determine the effect of functional ingredients on skin microbiota diversity.

The cosmetic industry targets the potential of skin microbiota with more and more studies conducted on the research of active ingredients targeting skin microbiota and the assessment of their action mode: skin benefits, microbiota balance and bacteria physiology, such as induction or suppression of metabolic pathways, adhesion, biofilm formation, growth kinetics, virulence factors, quorum sensing, etc. [1][37][38]. Active cosmetic ingredients that target skin microbiota can be classified into the following categories:

- active ingredients, algal- or plant-based, and thermal water-based, which are not a nutrient source for microorganism;
- prebiotics: nutrients that confer a health benefit with modulation of structure and functionality of the host microbiota in topical application for the cosmetic sector [26]. Cosmetic prebiotic approaches are to maintain healthy skin microbiota, or improve the skin microbiota composition by limiting or reducing pathogen growth and in the same time preserve or stimulate commensal bacteria growth [39][40][41][42];
- probiotics: fragmented bacteria that confer health benefits to the host. Cosmetic products with “probiotics” or “probiotic ingredients” often contain non-viable bacteria, products of bacterial fermentation or cell lysates, which do not require changes in the preservative ingredient system [26]. Nevertheless, cosmetic products containing fragments of microorganisms as probiotics require care regarding safe production. For now, a strict definition of a probiotic in cosmetic products has not been established and these products should only follow European Cosmetic Regulation 1223/2009 [43];

- post-biotics: bacterial metabolites and or cell wall components released by probiotic microorganisms [27].

The major identified applications of active cosmetic ingredients targeting particularly *S. epidermidis* and *C. acnes* are (1) promotion of commensal metabolism and/or bacterial diversity with the ratio *S. epidermidis*/*C. acnes* for limitation of pathogen invasion; (2) reduction of pathogen growth, virulence and biofilms; and (3) modulation of the skin microenvironment and modulation of the immune responses since the cause–consequence link between the skin pathologies and microbiota dysbiosis is not yet established. These claims in dermo-cosmetics are based on the evaluation methods: “Evaluation Methods of Skin Microbiota Targeting *Staphylococcus epidermidis* and *Cutibacterium acnes* from a Cosmetics Perspective”.

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