An innovative cosmetic formulation can restore the integrity of the skin barrier and the microbiota of sensitive, irritated and sensitized skin.

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Abstract

Background: A cosmetic formulation containing panthenol, niacinamide, bisabolol, hyaluronic acid, tocopherol and a prebiotic molecule, α-glucan was proposed to repair, soothe and moisturize sensitive, irritated and sensitized skin, also balancing and protecting the skin microbiota. **Methods**: Corneodesmosin expression was evaluated in reconstructed human epidermis (RHEs) after subjection to lactic acid and treatment with the CF using in situ immunofluorescent labeling and semi-quantitative image analysis. Cellular regeneration was evaluated by reflectance confocal microscopy. The facial images of the clinical study participants were obtained before peeling procedure and 14 days after the use of the CF. To assess the restoration and maintenance of the microbiota, metagenomics was studied. **Results**: It was observed a limited diminution of corneodesmosin expression in response to lactic acid (-38% vs. -72% in untreated stressed RHEs). The cellular regeneration studies showed an increase in epidermal thickness (+12%), dermal papilla depth parameter (+20%), keratinocytes morphology parameter (+36%) and in reflectance of the granular layer (+36%), as well as a significant reduction in the skin surface irregularity (-33%) 14 days after the peel procedure associated with the use of CF. Maintenance of the proportions between the main phyla of the human skin resident microbiota and species belonging to them and an increase in alpha diversity in the microbiome was observed 14 days after the peel procedure associated with the use of CF. **Conclusion**: The results obtained suggest that the cosmetic formulation could restore the integrity of the skin barrier and microbiota of a sensitized skin, beyond showing a postbiotic effect.

Keywords: skin barrier integrity; microbiota; sensitive and sensitized skin; postbiotic effect; prebiotic

Introduction.

The skin is the first physical, chemical, microbiological and immunological barrier against external aggressions, having a symbiotic relationship with microorganisms. This mutualistic relationship leads to a well-controlled but delicate balance of the microbiota, which is mandatory for healthy skin. Skin barrier protective abilities vary by region and age, typically creating a distinct microenvironment for microbial growth. Several factors can sensitize the skin, from external factors, such as extreme cold and heat, to the aesthetic procedures [1,2]. One of the biggest challenges of the 21st century is the fight against the aging of the organism, more particularly of the skin, for being more visible [3,4]. Combined, intrinsic and extrinsic aging cause facial skin changes such as: fine lines, deep wrinkles, sagging skin, increased hyperpigmentation, decreased shine and increased roughness. To improve these signs or slow down the aging process, patients have incorporated skin care routines, such as the use of daily cosmetic products associated with aesthetic procedures [4]. Among the aesthetic procedures that can be performed are chemical peels, laser and microneedling, among others, which cause skin damage, with consequent damage to the skin barrier and disruption of the microbiota.[4-6]

Among the chemical peels is the Jessner one, a solution that was developed by Max Jessner composed of 14% salicylic acid, 14% lactic acid, 14% resorcinol in 95% alcohol. Its mechanism of action is based on the keratolytic property of salicylic acid and resorcinol and on the epidermolytic action of lactic acid. The keratolytic agents in Jessner's solution cause corneocyte cohesion within the stratum corneum and subsequently produce intercellular and intracellular edema in the upper epidermis after application. With the application of Jessner's peel, skin signs such as erythema, whitish flocculation, mild to moderate burning can be observed [7]. After an injury, the body needs to start repairing, a process called healing. Healing is a dynamic process that involves three phases: inflammation, proliferation/repair and maturation/remodeling, and several factors can interfere in it, such as UV radiation, skin hydration level, age, among others [1,5,6,8,9]. Simultaneous modulation of different biological processes is the key to ensure fast and efficient recovery. Hence, it is important to use multi-repair agents that help in the process of repairing sensitized skin, either by relieving symptoms or stimulating healing [10]. Restoration and maintenance of the microbiota that was altered during such events is also essential for a good recovery [11,12].

Therefore, a cosmetic formulation (CF) containing a combination of active ingredients that has a synergistic effect in the three stages of the healing process: panthenol, niacinamide, bisabolol, hyaluronic acid, tocopherol and a prebiotic molecule, α-glucan was developed to repair, soothe and moisturize sensitive, irritated and sensitized skin, balancing and protecting the skin microbiota. Its mechanisms of action and its impacts on the skin barrier and microbiota restoration process are explained below.

D-panthenol (panthenol) is an alcoholic analogue of D-pantothenic acid, which has a more stable arrangement for application and permeation of topical products. Promotes skin hydration, due to its ability to retain and attract moisture, keeping the softness and elasticity of the skin. Furthermore, decreases transepidermal water loss from the stratum corneum, maintaining its hydration and improving skin barrier function [13-17]. It also has anti-inflammatory, providing a calming effect for irritated skin and stimulates the healing process epithelialization, increasing differentiation epidermal and aiding in the healing of minor injuries [14, 18-20].

Niacinamide (nicotinamide) is a water-soluble amide form of vitamin B3 [21,22]. Strengthens skin barrier function by increasing the synthesis of ceramides, keratin and other key barrier elements [23-25]. It has anti-inflammatory action, inhibiting important mediators of the inflammatory process, including IL-12, IL-1 and TNF-α [26-28]. In addition, it increases the proliferation of fibroblasts, stimulating collagen synthesis and contributing to the skin remodeling stage [25].

Bisabolol is an active ingredient widely used and known for its calming properties, which has anti-inflammatory action, inhibiting pro-inflammatory cytokines [29-31]. In addition, it promotes antiseptic, bactericidal, antimycotic and healing action [32].

Hyaluronic acid is the largest component of the extracellular matrix and a key player in tissue regeneration [33]. It increases the proliferation and migration of keratinocytes, helping to regenerate the skin barrier [34]. Low molecular weight hyaluronic acid is able to cross the stratum corneum, attracting water molecules and helping to maintain the correct configuration of the dermis fibers. The high molecular weight, on the other hand, creates a protective film, which helps to reduce transepidermal water loss through the skin, favoring its hydration, in addition to its anti-aging properties [35-37].

Tocopherols are antioxidants capable of inhibiting the chain reaction of radical species, such as reactive oxygen species (ROS) [38,39]. α-tocopherol is one of the most predominant in humans, in addition to being the largest non-enzymatic antioxidant source [40,41]. It is responsible for increasing the hydration of the stratum corneum, improving the relief of the skin surface [38]. It also acts on the expression of connective tissue growth factor and on the regulation of gene expression and transcription, thus facilitating the protection of wounds against infections and aiding in the epithelialization process of superficial wounds [42]. It also has anti-inflammatory action, being able to decrease the release of pro-inflammatory cytokines, such as IL-1, IL-6 and TNF-α, and decrease the adhesion of monocytes to the endothelium [43,44].

α-glucan oligosaccharide is a prebiotic molecule that helps in the maintenance and recovery of the skin microbiota and also reinforces the body's natural defenses, as it stimulates the production of antimicrobial peptides, beta-defensins 2 and 3, which are key markers of immunity. It favors the development of saprophytic flora beneficial to the skin microbiota, fighting unwanted, opportunistic or even pathogenic bacteria [11,12]. The prebiotic rebalances the bacterial landscape, strengthening the skin's microbiological barrier [45,46].

Considering the need of a multi-repair solution for a good and efficient recovery of sensitive, irritated and sensitized skin, this research aimed to evaluate preclinical efficacy of a cosmetic formulation (CF) containing panthenol, niacinamide, bisabolol, hyaluronic acid, tocopherol and a prebiotic molecule, α-glucan in corneodesmosin (CDSN) expression and the clinical efficacy in skin regeneration and microbiota maintenance and regeneration.

Materials and Methods.

Evaluation of corneodesmosin (CDSN) expression

Culture and Treatment

Eight-day-old reconstructed human epidermis (RHE) were stimulated or not (non-stimulated control) with the inducer lactic acid, tested at 0.6% (topical application), and the RHE were incubated in culture medium containing or not (control) cosmetic formulation (0.2%)

(systemic application). The RHE were then incubated for 48 hours with a treatment renewal after 24 hours (inducer and CF). All experimental conditions were performed in n=3.
At day 10, RHE culture was stopped and each tissue was fixed. Fixed tissues were dehydrated in multiple baths with increasing concentrations of ethanol and then embedded in paraffin. Transversal sections were performed with a microtome (5 µm thickness, 1 slide per labeling) and maintained at room temperature until immunolabeling.

Immunohistofluorescent labelings

The sections were deparaffinized and incubated at 92°C, pH 6, in a retrieval target solution in order to optimize antigen-antibody interaction. The sections were cooled down to room temperature in the same solution. After PBS-Tween-5% milk saturation, the sections were incubated for 1 hour with the primary antibody (anti-corneodesmosin). After several washes, the binding sites recognized by the primary antibody were revealed with a secondary fluorescent antibody (GAR or GAM-Alexa Fluor™ 568) and the cell nuclei were stained with DAPI solution. The sections were washed in TBS-Tween and assembled in Prolong Mounting Medium (ThermoFisher).

Microscopic observation, image capture and semi-quantitative analysis

The sections were observed using a ZEISS 710 confocal microscope. Images were captured and processed with ZEN software (objective lens x20). One image/replicate was captured. The fluorescence intensity and surface were measured on the captured images using ImageJ software and the fluorescence intensity was expressed related to the epidermis area.

Data management

Raw data were analyzed using Microsoft Excel® software.
Formulas used in this report:
Standard error of the mean: $sem = Sd/\sqrt{n}$
Percentage of viability: $viability\ (\%) = (OD_{sample} / OD_{control}) \times 100$

Evaluation of cellular regeneration by reflectance confocal microscopy

Study design

A 14-days, open-label, randomized, controlled, comparative clinical study was realized to evaluate the cellular regeneration of an injured skin after application of the CF by reflectance confocal microscopy. This study was realized with 7 female participants, aged 30-59 years, phototype (Fitzpatrick) I a IV with intact face skin. This study was conducted in accordance with the principles of the Declaration of Helsinki, applicable regulatory requirements, including CNS Resolution No. 466/2012, and in accordance with Good Clinical Practice (Document of the Americas and ICH E6: Good Clinical Practice).

After medical evaluation for inclusion, the dermatologist performed the clinical efficacy evaluation on both half faces (right and left) of the approved participants (T0). Then, cellular images of the participant's faces were captured with the Confocal Laser Reflectance Microscope (Vivascope® 1500) (T0). After performing this baseline measurement, the Jessner Peel procedure was performed by a dermatologist in all participants. Then the participants were instructed to apply the cosmetic composition 2 times a day (morning and afternoon), on clean and dry skin for 14 days. At the end of the 14 days, the participants returned to the institute for further evaluations and cellular images were captured with the Confocal Laser Reflectance Microscope (Vivascope® 1500) (T14).

Jessner Peel Application

The peel was performed by a dermatologist. Before application, the participant's face was cleaned with 70% alcohol. The peel was applied by the dermatologist with the help of a sterile gauze all over the participant's face, evenly in order to form a thin layer. The application could be made from 1 to 5 layers, the first application being called the first layer and so on. The number of layers applied was defined at the discretion of the dermatologist who performed the procedure, depending on the tolerance of each participant. After the application of the layers, the face of the participant had a white color called frosting and this specific color was an indication that the peel could already be neutralized, that is, the dermatologist should neutralize the action of the peel with sterile gauze and saline solution.

Reflectance confocal microscopy analisys

The Vivascope® 1500 Laser Reflectance Confocal Microscope was used to capture images to evaluate cell regeneration. To obtain the image, a metal ring was fixed to the skin with an adhesive ("adhesive window"). Immersion oil was added to the patch before it is attached to the skin. Then, a digital macroscopic image was captured with dermoscopic quality (VivaCam®), which is used as a guide to obtain microscopic images in the places of greatest interest. Finally, a hydrophilic gel was added for coupling the ring to the device, by magnetic means, which follows the orientation of the macroscopic photo. The images were taken in the malar region on one side of the face of selected participants following randomization.

For the morphological analysis, 4 Vivastacks and 4 Vivablocks (1 per layer) were performed and the following parameters were evaluated: epidermal thickness, dermal papilla depth, keratinocytes morphology, reflectance of the granular layer and skin surface irregularity.

A specialist in microscopy performed the analysis of the images. The thickness of each layer was calculated through the depths indicated in the images. For morphological data, the images were analyzed by classifying each image using a 4-point scale (0 to 3) [47]. For all analyses, scores were assigned to three images from each area of each participant per study time. Thus, generating three values per area per time. The average value of the scores was used in the statistical analyses. The interpretation of scores varies according to the parameter analyzed (Table 1).

Table 1: Parameters, variables and scale used in the assessment of skin hydration by Confocal Reflectance Microscopy.

Parameter	Variable	Scale			
		0	1	2	3
Thickness	Epidermis	Quantitative - Reduction (µm)			
Stratum corneum morphology	Surface irregularity	Regular		Unregular	
Keratinocytes morphology	Morphology	Unregular		Regular	
Reflectance of the granular layer	Radiance	Light		Intense	
Dermal papilla depth	Size	Quantitative - Increase (µm)			

Statistical Analysis

Statistical evaluation was performed by t-Student test. Bilateral hypotheses was used for epidermal thickness and unilateral hypothesis was used for keratinocytes morphology, reflectance of the granular layer (increase) and skin surface irregularity (reduction). The software used were XLSTAT 2021 e MINITAB 14. The values were considered significant for a 95% confidence interval.

Evaluation of the restoration and maintenance of the microbiota

Study design

Microbiological collection of human skin was performed in order to study the metagenomics of the microbiota. The collection was performed on the face of the selected participants. The microorganisms studied are part of the skin's natural microbiota. All human material such as skin surface cells was ignored and not used for analysis. The collection area was standardized with the aid of a flexible, sterile, plastic and individual mold, which limits the collection area to 12.5 cm^2. The swab was gently rubbed over this restricted area of the mold and, after collection, it was immersed in a tube with preservative liquid to avoid degradation and it was cut so that only the tip of the collected material was stored in the tube.

The collection was carried out at the following times:

- T0 – before the peeling procedure and application of the CF;
- T14 – after 14 days applying the CF.

Characterization of the test system

Next Generation Sequencing (NGS) on Illumina MiniSEq platform with genomic DNA library prepared with Illumina's Nextera XT kit in the V4 region of the ribosomal gene of prokaryotes and in the intergenic regions ITS I and II of eukaryotes.

The positive control item preparation was a DNA extracted from a known microorganism (Pseudomonas aeruginosa ATCC 9027) and Standard Zymobiomics (Zymo D6311) that was used to build a control library that was sequenced along with the test samples.

The main steps of the methodology were: DNA analysis and sequencing (PCR1, PCR2, purification, quantification and the combination of test item libraries with control library - PHIX) and Bioinformatics Analysis.

DNA analysis and sequencing

The DNA extracted from the samples underwent a standardization in a PCR reaction (Polymerase Chain Reaction) using the Veriti Thermal Cycler machine from Thermo Fisher with oligos anchored in the ribosomal DNA region of 16S bacteria. After purification with magnetic beads, PCR DNA quantification used the fluorimetry kit with the Qubit 4 Fluorometer – ThermoFischer Scientific equipment. The quantified DNA from the PCRs was used to build genomic DNA libraries with the Illumina Nextera XT kit and these were sequenced on a new generation Illumina MiniSeq pair end 150/150 platform, generating readings that reach 99% of the extracted microbiota.

Bioinformatics Analysis

The metagenomics technique is free from cultivation of microorganisms, as these are collected directly from the samples and their genetic material sequenced, so the terminology for identifying colonies of bacteria becomes readings, because every time a sequence of a given microorganism appears, it is read and quantified. Therefore, microbiota analysis was performed both quantitatively (abundance) and qualitatively (diversity). The microbiota readings generated from each sample were compared against existing public databases to classify the species present and the number of times each of them appears in the collected sample. Note: Negative control does not apply.

Statistical Analysis

The readings generated for each species of bacteria found in each sample of the study were transformed through the logarithmic function as a way to reduce the variation of interspecific data, that is, between samples. After this treatment, the data were analyzed by the Wilcoxon Signed Rank non-parametric statistical method, thus, the population of microorganisms present in the samples was compared with each other and the readings that obtained the 95% confidence level (5% significance, $p<0.05$) and trend at 90% confidence level ($p<0.10$) with

more relevant differences were considered. The software used were XLSTAT 2021 and MINITAB 14.

Results.

Increased corneodesmosin expression

Several factors can sensitize the skin and cause damage, affecting the skin's protective barrier [1,2]. The stratum corneum is composed of corneocytes that form a barrier and provides protection to the skin. Corneodesmosomes are responsible for the cohesion between the corneocytes and the intercellular lipid matrix, mantaining the stratum corneum intact. Corneodesmosin is a glycoprotein present in corneodesmosomes, which contributes to corneocytes cohesion and is a protease target during epidermal desquamation [48-50].

Here we evaluate the effect of the cosmetic formulation in corneodesmosin (CDSN) expression. CDSN was detected mainly at the surface of cells located within the granular layer (Figure 2). In response to the stress with lactic acid, the expression of CDSN strongly decreased (-72%). On the other hand, with 0.2% CF, a limited diminution of CDSN expression in response to lactic acid (-38% vs. -72% in untreated stressed RHEs) was observed (Figures 1 and 2).

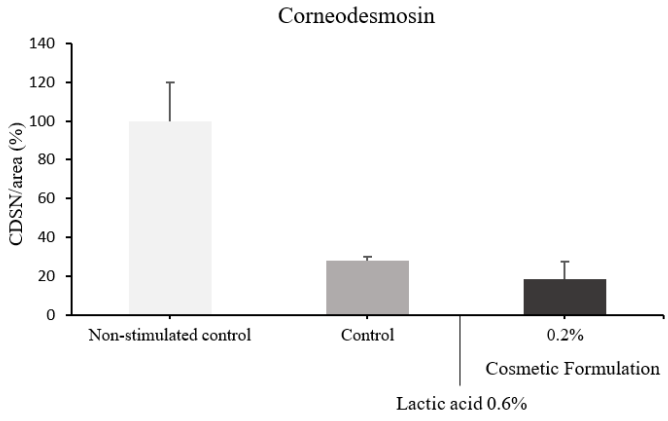

Figure 1: Effect of the evaluated treatment with the cosmetic formulation (CF) on the expression of corneodesmosin in reconstructed human epidermis subject to lactic acid application. The data represent the intensity of corneodesmosin staining related to the epidermis area (mean ± standard error of the mean of three replicates)

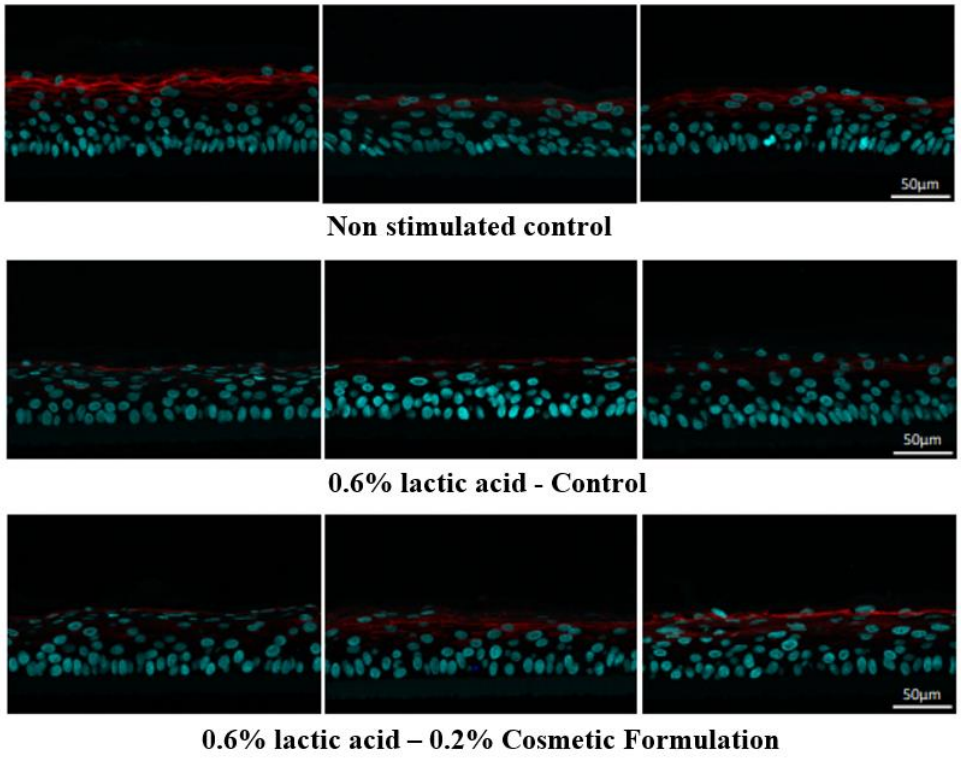

Figure 2: Representative images of the effect of Cosmetic Formulation on corneodesmosin expression in lactic acid-stimulated RHE. Cell nuclei (Blue) and corneodesmosin (red).

Skin regeneration

To evaluate skin regeneration in sensitizes skin the following parameters were analyzed by reflectance confocal microscopy before the Jessner Peel (T0) and after the treatment with the cosmetic formulation (T14): epidermal thickness, dermal papilla depth, keratinocytes morphology, skin surface irregularity and reflectance of the granular layer. The data showed an increase in epidermal thickness (+12%), dermal papilla depth parameter (+20%), keratinocytes morphology parameter (+36%) and reflectance of the granular layer (+36%), as well as a significant reduction in skin surface irregularity (-33%) 14 days after the peel procedure associated with the use of CF (Figure 3).

Figure 4 shows the images of the reflectance microscopy before and after the tretament with the cosmetic formulation. In the region inside the red circle of Figure 4A, it is possible to notice that in T0 there are no well-defined and larger papilla as seen in T14. These changes indicate better structure of the skin's basal layer. In the region inside the red circle of Figure 4B, it is possible to observe in T0 less uniformity of the skin's surface, with structures at

different heights. In T14, it is observed that the surface is more uniform, demonstrating greater cohesion in the stratum corneum. In the region inside the red circle of Figure 4C, it is possible to observe that in T14 the keratinocytes are better delimited, forming a “honeycomb pattern”. The greater delimitation is evidenced by the greater reflectance (lighter regions around the keratinocyte), indicating greater hydration and cellular nutrition.

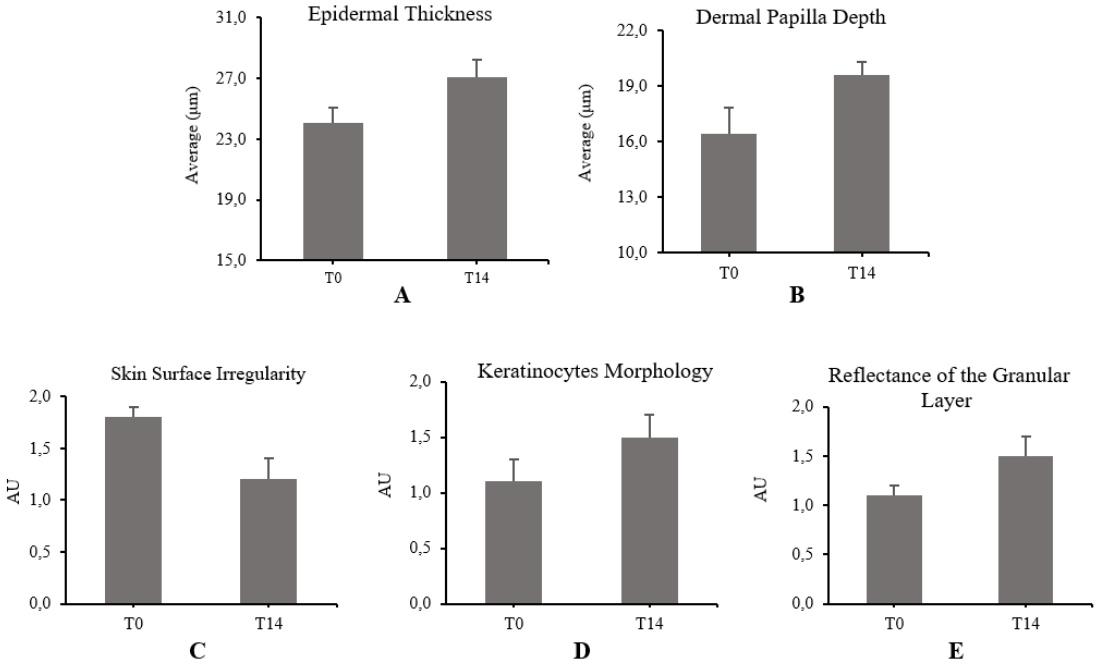

Figure 3: Effect of the evaluated treatment with the cosmetic formulation (CF) on the epidermal thickness, dermal papilla depth, keratinocytes morphology, skin surface irregularity and reflectance of the granular layer of the skin participants subject to Jessner peel. The data represent the mean ± standard error of seven participants. The values were considered significant for a 95% confidence interval. * $p < 0.05$ in relation to the T0 group.

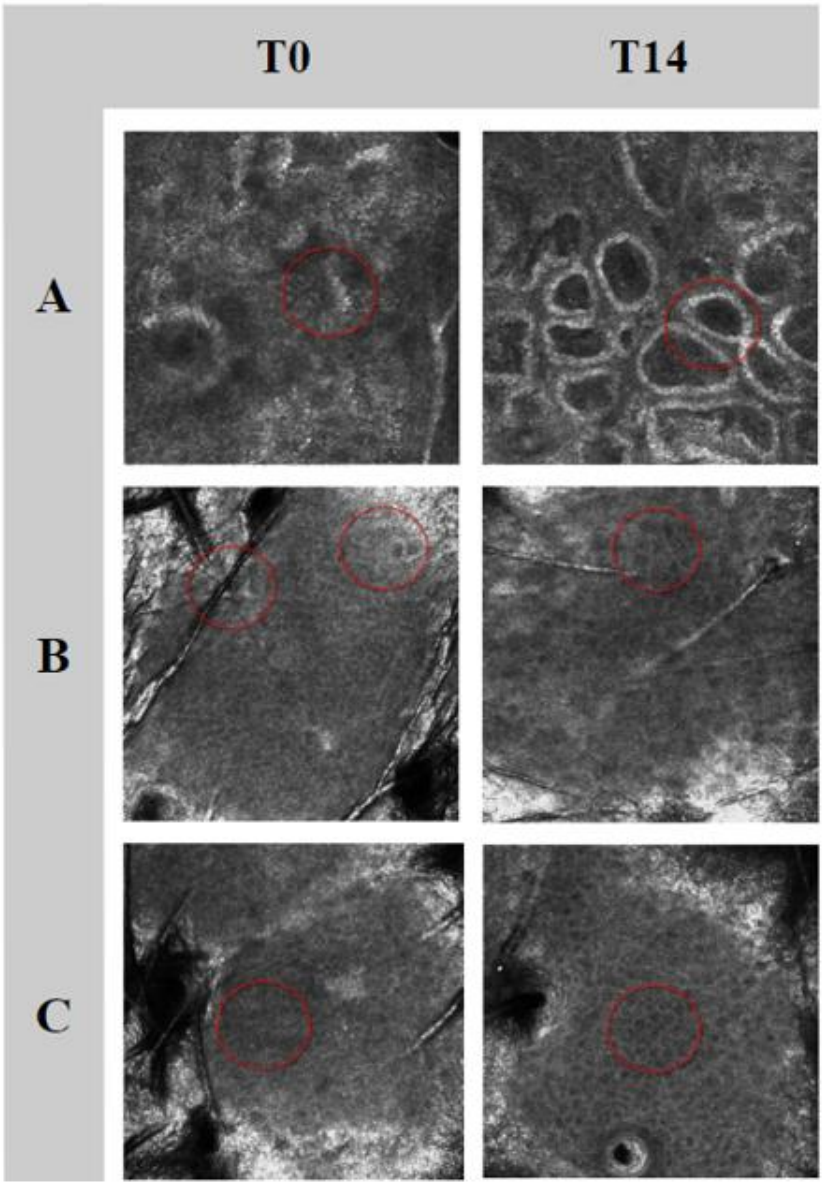

Figure 4: Reflectance Confocal Microscopy images. (A) Dermal papilla. (B) Surface uniformity. (C) Brightness of the granular layer and keratinocyte morphology.

Microbiota maintenance and regeneration

In sensitized skins, there are also changes in the microbiota, making the skin more exposed to external aggressors. Restoration and maintenance of this microbiota are essential for a good recovery [11,12]. To assess the restoration and maintenance of the skin's microbiota, metagenomics was studied. The term metagenomics was first described by Handeslman in 1998 to access the metabolism of unknown microorganisms in the soil [51]. In mid-2012 the

National Institute of Human Health and the European Meta Hit consortium began publishing the results of Human Metagenome studies that aimed to characterize the human microbial communities present at specific sites in the body. These efforts in recent years have resulted in an extensive map of the microbes (microbiota) that live in and on us. Overall, the study of the metagenomics of microbial communities is an effective approach to identifying the microorganisms or microbial metabolic characteristics of any non-culturable sample. Analysis of metagenomics data typically seek to identify specific organisms, genera, operational taxonomic units, or relative abundances that differ between two or more groups of samples, and various characteristics of microbial communities have been proposed as potential biomarkers to address diseases and be applied in the design of new products [52].

Here we found that after 14 days of use of the CF, the proportion of species for the 4 predominant phyla present in skin was similar to time T0 (Figure 5B), suggesting that the product used must have acted in the rebalancing and repopulation of the skin after the treatment. In general, astringent treatments such as peel, even if mild, lead to unbalance of the skin's microbiota population as it removes the protective barrier removing the resident microbiota [7]. However, as can be seen in the graphs of Figure 5A in absolute numbers, the number of microbiota readings at time T14 is twice the number of readings at initial time T0, corroborating the fact that the CF acted in order to preserve the return of a microbiota resident in addition to having brought more diversity to the participant's microbiota. This can be evidenced by the number of alpha diversity measured in the microbiota of each participant before and after of using the CF.

The Shannon index (alpha diversity) takes into account the uniformity and richness of species in a community, that is, the variety and relative abundance of species in a community in a certain population. The higher the index, the greater the diversity of species in that population. From this index it was possible to calculate the uniformity (E) that goes from 0-1, and the closer to 1 the greater the diversity in that population [53]. In this study, 60% of the participants showed greater diversity alpha after the use of the CF. Species of the genera Streptococcus and Veilonella of the Phylum Firmicutes also showed a statistical difference of 5% ($p < 0.05$) between times T0 and T14 for each of the study participants, showing an increase in their readings.

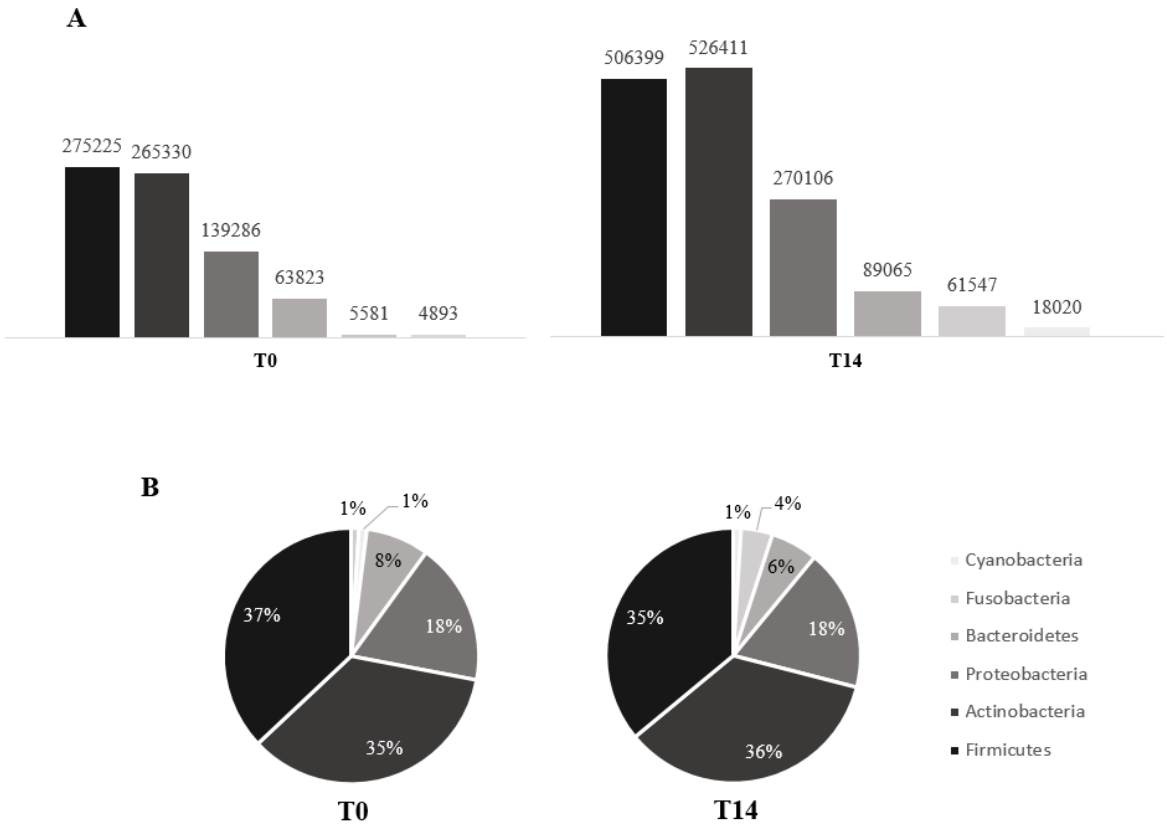

Figure 5: Microbiota composition of participants at times T0 and T14 for the main Phyla in absolute numbers of readings (A) and percentage (B).

Discussion.

The data obtained by reflectance confocal microscopy analysis suggest that the use of CF promotes cell proliferation, in addition to an improvement in the pattern of the dermal-epidermal junction and a better organization of the keratinocytes, resulting in a better structuring of the basal layer and cellular renewal. In addition, the increase in the granular layer reflectance indicates deep hydration and greater nutrition of the region, while the significant reduction in the surface irregularity parameter represents a more cohesive and, therefore, more uniform stratum corneum, corroborating with the increase in corneodesmosin expression after treatment with CF.

As expected, the stress with lactic acid resulted in the aggression of the skin barrier, demonstrated by the reduction in expression of cornedesmosin, which confers adhesive property and plays a fundamental role in conferring resistance by the primary cohesive force and preventing cellular desquamation, maintaining skin hydration [49,50]. In the presence of

the cosmetic formulation, a limited reduction of corneodesmosin expression was observed, demonstrating the capability of the CF in limit the damage in skin's barrier.

The increase in the thickness of the epidermis (Figure 3A) indicates greater hydration of the skin, an increase in the number of cells (cellular proliferation) and cell renewal. The morphology of keratinocytes in the granular layer is measured by evaluating the "honeycomb pattern" generated by the distribution of keratinocytes (Figure 4C). In aged skin, the morphology and size of keratinocytes tends to be pleomorphic and the pigmentation pattern is also irregular [54]. The improvement in the parameter (increase in the parameter, Figure 3D) indicates stimulation of cell renewal, with more organized and more homogeneous keratinocytes. The granular layer reflectance is the degree of reflectance in the area between the edges of the keratinocytes and the cytoplasm. The increase in reflectance (Figures 3E and 4C) indicates an improvement in the parameter. Dry and/or aged skin tend to have low reflectance. A better contour of the keratinocytes and, therefore, greater reflectance may be related to an enrichment of the inter-keratinocyte space with lipids and proteins, which indicates more nourished, hydrated and functional keratinocytes [47]. The size of the papilla is a quantitative measure where it is measured how many micrometers the papilla has. With aging or any damage, there is a loss in the number and definition of the papilla, which become flattened and shorter, which leads to a reduction in the thickness of the epidermis [54]. Thus, the improvement in the parameter (increase in size, Figure 3B) indicates better structuring of the basal layer and improvement in the pattern of the dermal-epidermal junction (Figure 4A).

The metagenomic study showed the maintenance and restoration of the skin microbiota after the use of CF, ensuring the return of the resident microbiota, in addition to bringing more diversity to the microbiota, showing also a postbiotic effect. Species diversity denotes balance and health due to the increase in beneficial metabolites that are produced. Species of the genera Streptococcus and Veilonella of the Phylum Firmicutes showed a statistical difference of 5% ($p < 0.05$) between times T0 and T14 for each of the study participants, showing an increase in their readings. Species of the genus Streptococcus produce lactic acid from the fermentation of sugars in the host's diet. Lactic acid, like glycolic acid and mandelic acid, are alpha-hydroxy acids (AHA), natural organic acids that bring benefits to our body and our skin. One of the main benefits is the humectant property, which provides the skin with greater water retention, contributing significantly to skin hydration. These two genenera

(Streptococcus and Veilonella) are found in several places in the human body and are known to act in coordination with each other due to the lactic acid produced by the species of the genus Streptococcus, which is an essential source for the growth and development of the species of the genus Veillonella, and the species of the genus Veillonella, in turn, create an environment that is suitable for the development of other bacterial species, thereby allowing the repopulation of the place where they reside [55]. Therefore, favoring these two genera possibly brought benefits to a skin that needed to be repopulated after treatment that affected the integrity of the physical barrier and the balance of the microbiome.

Conclusion. The results obtained suggest that the cosmetic formulation containing panthenol, niacinamide, bisabolol, hyaluronic acid, tocopherol and a prebiotic molecule, α-glucan could restore the integrity of the skin barrier and the skin's microbiota of a sensitive, irritated and sensitized skin, beyond showing a postbiotic effect.

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Conflict of Interest Statement.

NONE

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