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Analysis of immediate use of sunscreen after microneedling

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Abstract

Introduction: Microneedling promotes skin microlesions that lead to an inflammatory process, increasing cell proliferation, cell metabolism, and synthesis of collagen and elastin, therefore restoring skin integrity.

Objective: This study aims to investigate the differences between the physical and the physical-chemical sunscreen application after microneedling, assessed through histological analysis.

Method: This was a two-phase study. The first phase investigated the physical and physical-chemical sunscreen penetration mixed with India ink through histological analysis. The sunscreens were applied after the microleakage in vivo on the skin of a volunteer who underwent abdominoplasty 24 hours after the procedure. Histological analyses were carried out using optical and electron microscopy. The second phase analysed the skin reactions with the use of physical sunscreen after different microneedling treatments. The sample consisted of 30 volunteers distributed into three groups: G1 received the "Roller" microneedling, G2 received pen micropuncture treatment, and G3 received the fractional radiofrequency treatment.

Results: The histological analyses of the first phase indicated that the physical-chemical protection sunscreen penetrated more deeply, and pigment was found among the collagen fibres and the dermal fibroblast cytoplasm in comparison to the physical protection sunscreen, which had the pigment confined exclusively in the superficial epidermis layer. The second phase results demonstrated that the use of the physical protection sunscreen after the different microneedling techniques showed no adverse reactions such as itching, pain or soreness, and the hyperaemia.

Conclusion: The proposed intervention showed that the use of physical protection sunscreen after different microneedling procedures is safe.

KEYWORDS

collagen, microneedling, sunscreen

1 | INTRODUCTION

The Collagen Induction Therapy (CIT) technique was first introduced in the 1990s, and its purpose was to induce collagen production in skin scars and wrinkles treatments. This technique promotes microlesions on the skin, generating a local inflammatory process that promotes cell proliferation (mainly fibroblasts). Consequently, the tissue's cellular metabolism (dermis and epidermis), increasing the synthesis of collagen, elastin, and other substances present in the skin, restores its integrity.[1]

Several types of microneedle instruments are available in the market, the most popular being the rollers and the microneedle pens. Rollers are small needle-studded rolls (or cylinders) usually made of steel or titanium. Another similar device is the microneedle pen, which is featured in different types/models, either manual (mechanical) or electric. The pen's needles are disposable, and their length can be adjusted from 0.25 mm to 2.5 mm.[2] It is also possible to use the microneedle fractional radiofrequency, a system that allows the heating of a small tissue volume close to the needle tip while the rest of the needle is isolated. With these needles, energy flows only through their tips, resulting in a small point of thermal coagulation in the skin.[3]

Currently, it is possible to associate drug delivery techniques with microneedling, allowing transdermal delivery of selected assets. After skin microneedling, microchannels open, and these pathways facilitate and potentiate the permeation of topical substances through the skin, optimizing the desired results.[4] According to some authors, after microneedling, there is an 80% increase in cosmetic permeation.[5-7] Others estimate increases of up to 500%.[8]

After the microneedling procedure, the use of skin photoprotection is required due to the inflammatory process that results from the mechanical trauma caused by the microneedles in the stratum corneum and its physiological effect.[9,10] Direct or indirect sun exposure should be avoided for about 10-28 days after microneedling when possible, especially when using needles of size greater than 1.0 mm. The use of sunscreen is encouraged for at least 1 week after treatment. However, the literature is not coherent as to the time of sunscreen application after microneedling treatment, and only its recommendation for use is advised.[11,12,13]

Sunscreens are defined as topical preparations that reduce harmful solar ultraviolet (UV) wavelengths penetration. They can be classified into physical sunscreens, also called inorganic sunscreens, composed of minerals such as iron oxide, titanium dioxide or zinc oxide, and are characterized by being unabsorbed by the skin, as the physical filter reflects sun rays. The sunscreens classified as chemical or physicochemical are the most commonly used. They have highly energetic molecules that absorb ultraviolet radiation, creating chemical protection by reacting with solar radiation and preventing it from penetrating the skin.[14]

Despite being essential in post-treatment, the use of sunscreen on broken skin still generates significant divergence, as it is believed that applying sunscreen onto a recently microperforated skin would be considered as chemical drug delivery and, for many, this should be avoided, as they contain substances that could cause serious adverse effects. Thus, this study investigates the differences between the physical and the physical-chemical sunscreen application after microneedling, assessed through histological analysis. This study also analyses skin reactions' responses according to the type of microneedling devices, such as the traditional microneedling (roller and pen) and the fractionated radiofrequency.

2 | MATERIALS AND METHODS

This two-phase study was approved by the Potiguar University Ethics Committee (approval code 3,322,544) and carried out according to the recommendations of the CONSORT TRANSPARENT REPORTING OF TRIALS (CONSORT, 2010). The first phase investigated the collected material through histological analysis using optical and electron microscopy regarding the permeation of physical and physicochemical sunscreens after the traditional in vivo microneedling on human skin. The second phase consisted of a randomized controlled experimental study that analysed skin reactions using physical sunscreen after microneedling treatment performed with different devices.

All volunteers signed an informed consent form. The second phase's simple randomization consisted of a draw using envelopes containing a response card, which indicated the group allocation for each participant. The group allocation sequence was followed according to a list generated by the Research Randomizer Software™.

2.1 | First phase

2.1.1 | Participants

In the first phase, a volunteer who was scheduled for an abdominoplasty surgery was recruited. The participant's abdominal skin would be removed for histological analysis. As inclusion criteria, the participant would have to be in a preoperative period of abdominoplasty surgery and should have preserved comprehension capacity and local body sensitivity. The exclusion criterium would be the presence of any microneedling contraindication.

2.1.2 | Evaluation procedures

After recruitment, the volunteer signed the free and informed consent form in agreement with all study and treatment procedures. The sociodemographic and anthropometric data were collected, and the target area was photographed with a semi-professional camera (Canon, SX530 HS) 24 hours before the plastic surgery.

2.1.3 | Intervention protocol

For microneedling, the skin of the abdominal area was cleaned with a cleansing gel and a topical antiseptic solution with 2%-chlorhexidine

diglycerate with surfactants; then, the two 10-cm^2 areas were demarcated on the right and left infraumbilical region. The microneedling procedure was then performed with the Dr Roller™ device (Moohan Enterprise CO.), which consists of 540 stainless steel microneedles attached to a cylinder that is rolled on the skin surface, causing micro punctures and tissue hyperaemia. Then, two different types of sunscreens were applied to the treated area: the physical, which features a system called "skin cover" and forms a "film" or "physical barrier," preventing its penetration into the skin; and the photoactive SPF 50, a physicochemical protector (both manufactured by Mezzo Dermocosmetics). Before the sunscreens were used, they were mixed with blue India ink (20 mg of sunscreen/20 ml of India ink) to serve as a histological analysis marker. The physical protection sunscreen covered the right side, and the left side received the physical-chemical protection product (Figure 1).

Right after using sunscreens, the micro-pocketed areas were covered with gauze for protection and washed 24 hours later. Then, the volunteer underwent abdominoplasty surgery, in which the abdominal skin flap corresponding to the two treated areas was removed. The collected materials were then sent for histological analysis using optical and electron microscopy.

2.2 | Second phase

In the second phase, the clinical study evaluated 30 volunteers, men and women aged between 25 and 55, of different skin phototypes. As the inclusion criterium, the volunteers would have to present no contraindications concerning the treatment equipment or cosmetics (collagen-related diseases such as keloids, healing and/or protein synthesis problems).

The exclusion criteria were that volunteers could not: be under the effects of drugs that promote physiological changes to the skin; be under dietary restriction (diets, dietary re-education) that could significantly influence skin physiology; have severe metabolic diseases, refrain from signing the free and informed consent form.

The volunteers were randomly distributed into three groups: Microneedling Roller (G1), with 10 participants, mean age 27.5 (±1.2) years; Microneedling Pen (G2), with 10 participants, mean age 26.9 (±2.1) years; and the Microneedle Fractional Radiofrequency (G3), formed by 10 participants, mean age 27.3 (±1.6) years.

2.2.1 | Evaluation procedures

All participants underwent photographic evaluations, which were performed pre-treatment, after 24 hours, and seven days after applying the therapeutic resource (microneedling) and the sunscreen in each group, using a semi-professional camera (Canon, SX530 HS). The photographs were taken with the volunteers in orthostatic position, anterior view, and the same camera was used for all photographs. A tripod was positioned for distance standardization between the camera and the target for all photographs.

The data collection instrument used in this research was the Baumann skin characterization questionnaire, 2006,[15] which evaluates the skin using 64 questions and classifies it as: (a) oily or dry; (b) resistant or sensitive; (c) pigmented or non-pigmented; and (d) prone or non-prone to wrinkles. Thus, this classification allows for better identification of the patients' different clinical conditions.

2.2.2 | Intervention protocol

For the therapeutic procedure, the following devices were used: a roller-type microneedling device (Doctor of Aesthetics), consisting of 540 1.5-mm-long titanium microneedles; a Smart Derma Pen microneedle electric pen (Smart Gr™) using 36 1.0 mm needle refills; and a Spectra Medic™ (Tonederm™) fractional radiofrequency device using the stamp-type electrode.

In G1, the microneedling treatment was performed with the Roller device; G2 was treated with the electric microneedling pen; and G3 received the fractionated radiofrequency device application, using the sagging mode as a modulation parameter adjustment of 20 mJ, during 30-minute sessions. The skin was treated with strokes in all directions, as if imitating the shape of a wind rose, with repetitions of 8 to 10 times in each direction. The procedure was performed in a single session. Initially, a topical anaesthetic (Dermomax™, Aché™) was applied (40 mg lidocaine/g), then all volunteers were placed in the supine position, and the treatment was bilaterally applied on the volunteer's face.

In all groups, sunscreen was applied only onto the face's right region immediately after the procedure. The Fotoactive Skin Cover™ (Mezzo Dermocosmetics™) sunscreen lotion was used, which has

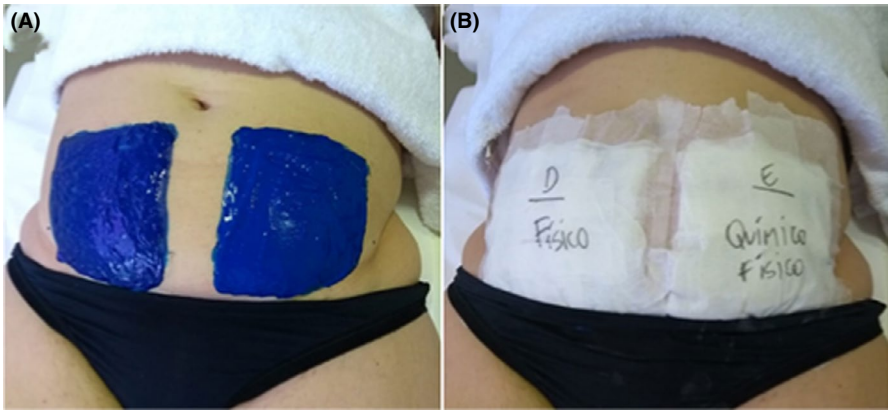

FIGURE 1 A, Application of sunscreen mixed with India ink; B, Treated area protected with gauze after the sunscreen applications

wide UVA, UVB and UV-VIS protection Bioecolia™, copper peptide, Genencare, vitamin E, amino acids, carnosine and Zon-term.

The revaluation was carried out 24 hours and seven days after the treatment session using the same pre-treatment methodology and applying the proposed questionnaires. The study used a large-particle photoprotector in the micronized form, which produces a film on the skin, precisely to prevent this product's permeation in situations of aggressive or damaging skin treatment such as in dermabrasion or any other type of treatment that removes skin layers.

2.3 | Statistical analysis

Qualitative data were described as per pathologist reports (descriptive analysis of histological images) and qualitative analysis of photographic images and quantitative responses to the questionnaires. Data collection and correlation were presented in tables and figures.

3 | RESULTS

3.1 | First phase

3.1.1 | In vivo macroscopic analysis—human skin

After the flaps were removed, they were cut into eight pieces for macroscopic analysis. The results indicated that the flap of the area that received the physical-chemical product mixed with India ink presented more "pigmented" areas (Figure 2B), indicating some degree of mixture permeation through the drug delivery process.

3.2 | Light microscopy analysis

Figure 3(A) showed that through histological evaluation made through haematoxylin and eosin (HE) 400X staining, there was some confined pigment exclusively in the epidermis' superficial layer when it received the physical protection sunscreen. Regarding Figure 3(B), which shows

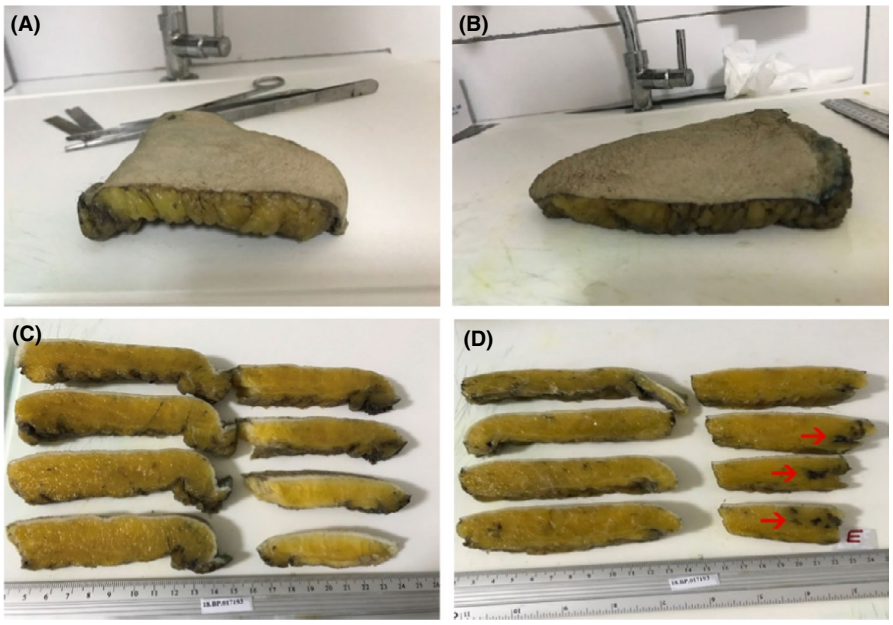

FIGURE 2 A, Surgical flap of the area where the physical sunscreen was applied; B, Surgical flap of the area where the physical-chemical sunscreen was applied; C, Flaps where the physical sunscreen was applied divided into pieces; D, Flaps where the physical-chemical sunscreen was divided into pieces

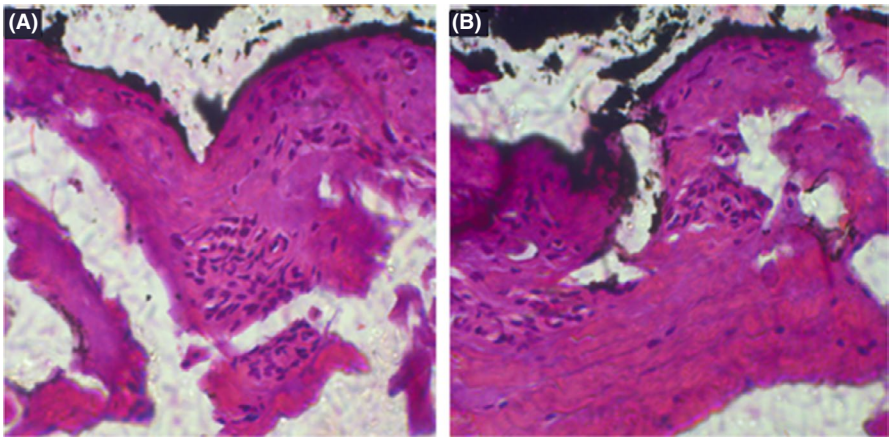

FIGURE 3 Light microscopy analysis Haematoxylin and eosin (HE) 400X staining; A, area where the physical protection sunscreen was applied (identified ("arrow") the presence of pigment in the epidermis); B, area where the physicochemical protector was applied (identified ("arrows") the presence of pigment in the dermis)

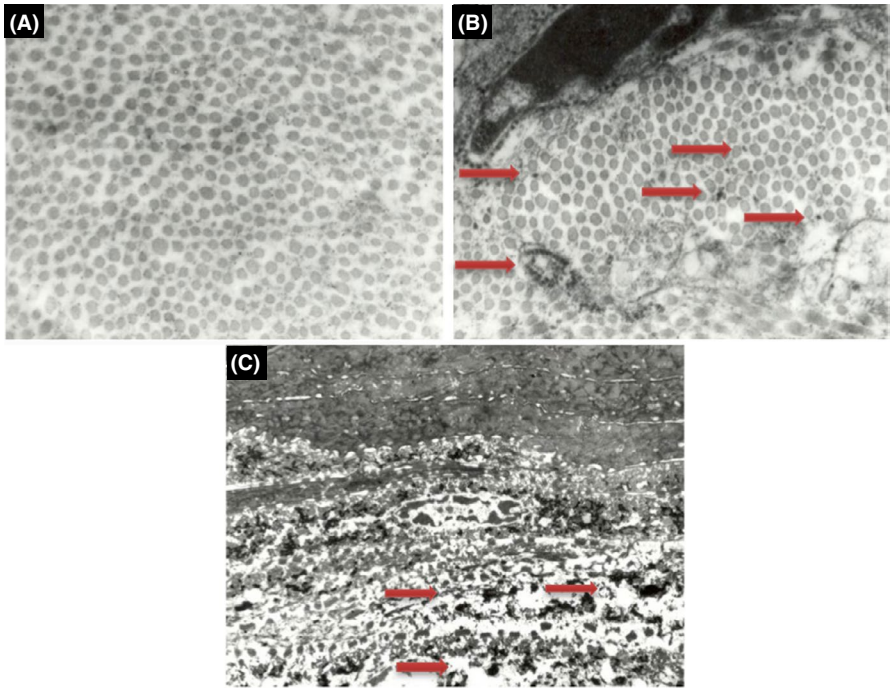

FIGURE 4 Electron microscopy analysis; A, area where the physical protection sunscreen was applied (no product permeation level); B, area where the physical-chemical protection sunscreen was applied protective substance permeation ("arrows"), C, presence of pigments in the collagen fibres and cytoplasm of the dermis fibroblasts in the area where the physical-chemical protection sunscreen was applied

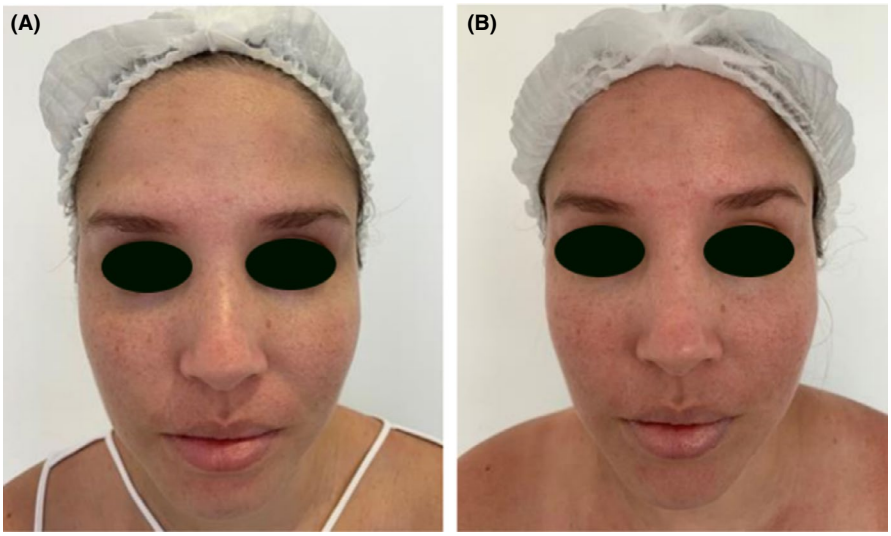

FIGURE 5 G1 (A) before; B, after 24 h

the skin side that received the mixture, a pigment-suggestive area was identified in a blood vessel wall, which led the authors to understand that pigment penetration probably occurred into the dermis.

3.3 | Electron microscopy analysis

An electron microscope was used to analyse any degree of protective substance permeation that could be identified through a histological section of the skin, showing details of the dermis' vascular network with transmission electron microscopy images (increase 31, 5X).

In Figure 4(A), to which the physical protection sunscreen was applied, no degree of product permeation was identified, whereas, in Figure 4(B), there was some visible protective substance penetration after microneedling. This is confirmed in Figure 4(C), which shows some pigment found between the collagen refibres and the dermal fibroblast cytoplasm.

3.4 | Second phase

3.4.1 | Photographic analysis

Photographs were taken before and 24 hours after the intervention. Figures 5 - 7 show the volunteers' skin reactions in each group. The authors found no visible adverse reactions such as irritation, redness (intense erythema), oedema, blisters, or any other dermatological condition on the treated volunteers' facial skin. The area that received the

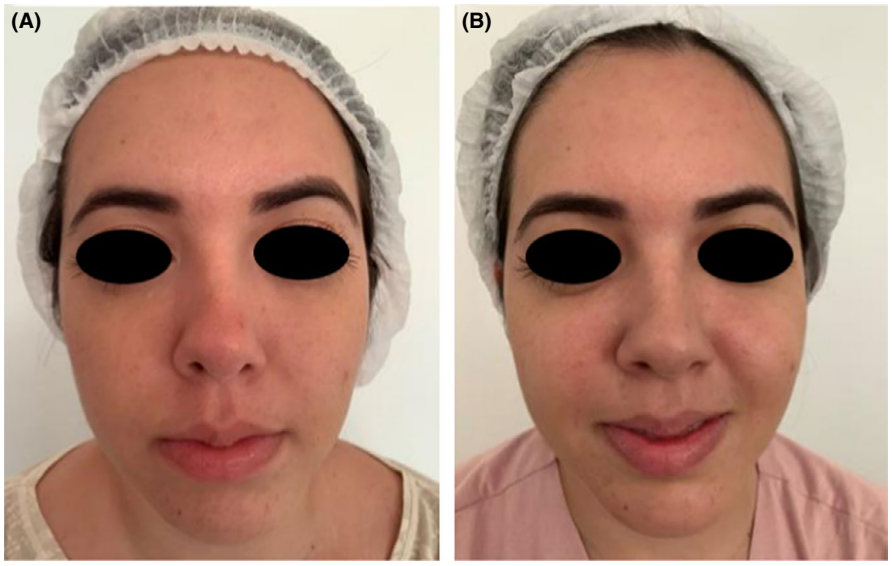

FIGURE 6 G2: (A) before; B, after 24 h

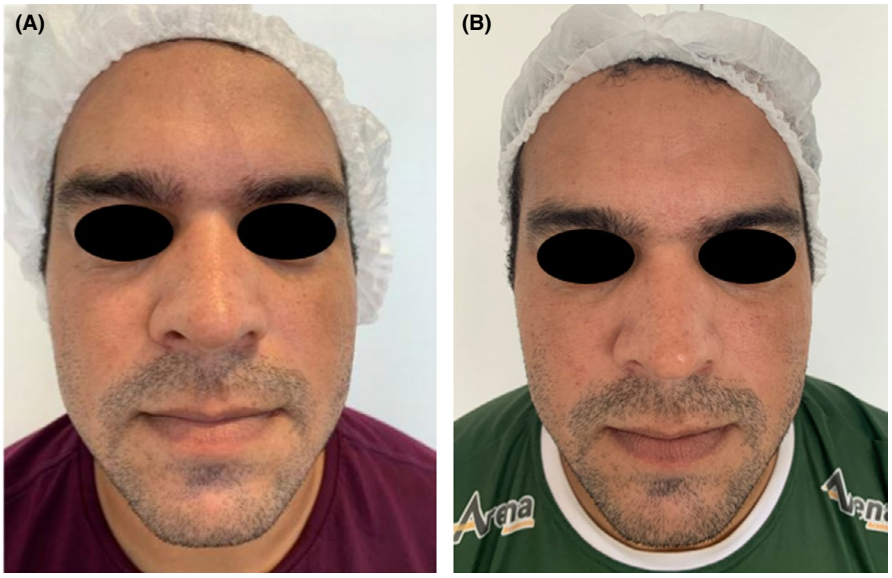

FIGURE 7 G3: (A) before; B, after 24 h

physical sunscreen product with the "skin cover" system presented lower hyperaemia (erythema) than the untreated side.

3.4.2 | Analysis of erythema time according to treatment type

The erythema caused by the needles' physical activity was evaluated according to the time presented by the volunteers' skin and associated with the received treatment type. Table 1 presents the mean times for each group, with highlights to G1, which presented the highest average time.

TABLE 1 Mean time of erythema

	Mean time of erythema after session
G01	20 min to 12 h
G02	5 to 10 min
G03	30 min to 2 h

TABLE 2 Results difference in hemifacial hyperaemia (%)

	Side with no sunscreen	Side with sunscreen
G01	80%	20%
G02	80%	20%
G03	75%	25%

3.4.3 | Hemiface hyperaemia difference (%)

The analysis of the differences between the hemifaces after sunscreen use showed lower hyperaemia on the side that received the physical sunscreen (T1). Table 2 presents the side comparison percentage values (with sunscreen vs with no sunscreen). G1 and G2 had similar results; 80% had hyperaemia on the side without

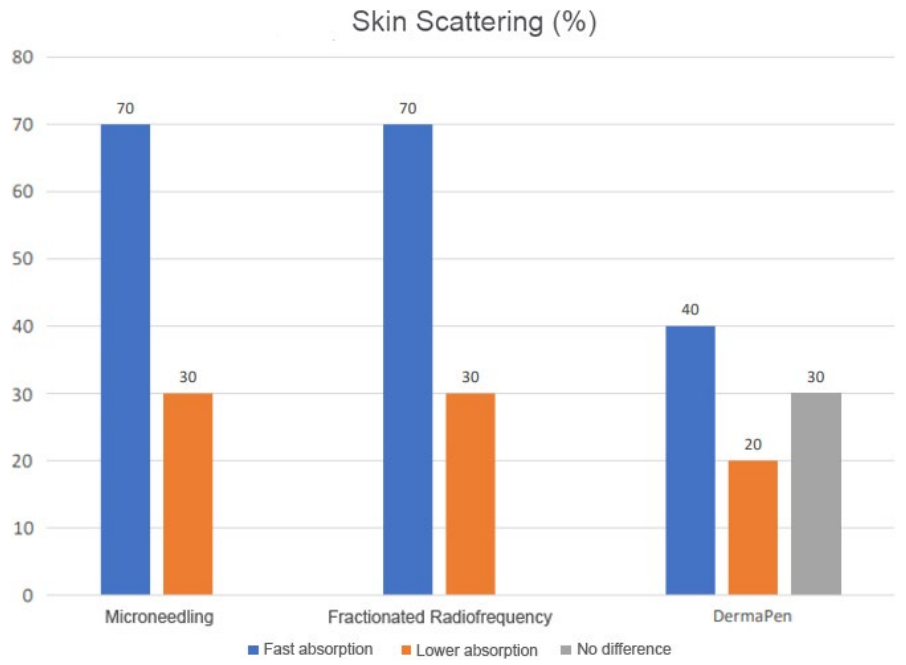

FIGURE 8 Results (%) of the questionnaire on product spreadability on the skin after the microneedling session

sunscreen, whereas only 20% of volunteers had hyperaemia even with sunscreen use. The fractionated radiofrequency group (G3) presented a higher number of volunteers with hyperaemia on the sunscreen-covered side when compared to the other groups.

3.4.4 | Analysis of physical protector spread with skin cover system (%)

According to volunteers' opinions, the subsurface scattering quality was assessed through a questionnaire, and the answers are presented in percentages, according to volunteers' opinions (Figure 8). G1 and G3 showed similar results, 70% of volunteers reported fast product spreading, and 30% reported that product spreading was not as fast after the proposed intervention. For G2, 40% of volunteers noticed spreading was fast, 20% felt spreading was not fast, and 30% found no difference in product spreadability compared to previous sunscreen use.

At the end of the intervention, the volunteers' satisfaction with the sunscreen was verified. In response, 94% reported that they would use the sunscreen after the microneedling procedure, and only 6% would not use the product because due to no adaptation to the use of that particular sunscreen.

4 | DISCUSSION

More pigment was noticed on the evaluated area's flaps to which the physical-chemical protection sunscreen mixed with the India ink was applied. An area suggestive of blood vessel wall pigmentation was also observed. The mixture may have penetrated the dermis, indicating some degree of mixture permeation through the drug delivery process. This may be explained by micro punctures in the dermal tissue caused by the microneedling procedure. The needles open channels that connect the dermis to the external environment, allowing the introduction and absorption of topical assets to be more easily and quickly deposited in the skin's most superficial layer.[16] Thus, with the opening of these channels, the physical-chemical sunscreen, formed by organic molecules are capable of absorbing UV (high energy) radiation, which are essentially aromatic compounds with carboxylic groups, penetrated more in-depth into the skin layers, suggesting that the use of this type of sunscreen after microneedling can be harmful to the skin.[17] In corroboration, a study[18] analysed the organic (chemical) sunscreen permeation in the skin layers with 3-(4-methyl benzylidene), benzophenone 3, and octyl methoxycinnamate with in vivo experiments, as well as their effect on reproductive hormone levels in humans. The results revealed substantial penetration of these topical products into the organism, and residues were identified in the volunteers' urine and blood. However, no changes in female sex hormone levels were detected within the time set for control.

However, when analysing the physical protection sunscreen penetration, it was macroscopically observed that the flaps were less pigmented, and the optical microscopy analysis showed that the pigment was confined into the skin's superficial layer. This occurred due to the oxides (titanium dioxide and zinc oxide) present in this product formulation. When they are incorporated into the formulations, they form a particle film on the skin. Inorganic (physical) protection sunscreens are made of particles, and depending on their size, protection can occur not only through reflection but also through absorption. The size of these particles is of great importance not only in the sunscreen effectiveness but also in the product's cosmetic appearance. [17,19] This is the case with the "skin cover" system used in this study, which has minimal particles and features low skin irritation potential and good spreadability.

The characteristics of titanium dioxide (TiO_2) and zinc oxide (ZnO) are similar. Apart from protecting against UVA radiation, they do not have significant skin irritating properties or sensitization potential. However, the ZnO is more efficient as far as this protection is concerned. In vivo and in vitro studies[20,21,22] showed no penetration of titanium dioxide with product use; however, limited zinc oxide penetration was identified in the skin. This corroborates with the present study, in which the India ink mixed with the physical protection sunscreen was exclusively confined to the epidermis' superficial layer, indicating that the use of this type of product is safe after microneedling sessions. Based on the studies mentioned above, it is noteworthy that the early application of sunscreen after this type of procedure seeks to protect the skin against solar radiation harms, especially hyperpigmentation, without causing health risks to the treated patient with the penetration of harmful substances into the skin.

Given the safe use of physical protection sunscreen, the second phase of the study was carried out, in which it was verified that after the application of the different microneedling types, the area that received the sunscreen with skin cover showed less hyperaemia (erythema) compared to the side with no sunscreen, with around 80% the difference between the sides. Also, pruritus reactions or pain in the treatment region were not reported through answers to the satisfaction questionnaire. The justification for the reduced hyperaemia is found in the formulation of the physical sunscreen with the skin cover system, as one of the present substances is the Bioecolia™, considered a soothing prebiotic. The substance does not have an anti-inflammatory action, which could interfere in the response to microneedling.[23]

The technology called "skin cover" acts by reinforcing the natural defences of the tissue through the Bioecolia's™ prebiotic property, stimulating the growth of the beneficial flora that is naturally found on the skin reducing pathogenic flora.[23] It acts by accelerating recovery due to the presence of copper peptides, which are growth factor fragments that act on cell differentiation, and on the healing process due to dermal fibroblasts proliferation and stimulating extracellular matrix production and angiogenesis.[24] Given these facts, the authors found the use of this product to be efficient as, in addition to not presenting percutaneous absorption after microneedling, its properties acted in blocking solar radiation and were beneficial for skin regeneration.

Physical (inorganic) sunscreens are considered non-toxic, stable, do not react like the chemical (organic) sunscreens, and are usually clinically safer. For these reasons, physicists consider it the first choice among photoprotection for individuals with a history of allergy. Nevertheless, there are some adversities for the cosmetic area, such as the appearance of whitish opaque spots on the skin after application, favouring comedogenesis, and the transfer of the product to the clothes' fabric, jeopardizing the cosmetic's photoprotective efficacy.[20,21,25] However, the results proved to be satisfactory regarding product spreadability and coverage, and the best results for skin scattering were observed within G1 and G3.

5 | CONCLUSION

The authors concluded that sunscreens with physical characteristics could be safely applied after microneedling to protect the skin against solar radiation harmful effects with no health risk. With no penetration of the physical protection sunscreen after microneedling, the sunscreen remained at the epidermis level only with the India ink mixture. It was also demonstrated that the "skin cover" physical protection technology is safe and can be applied after invasive procedures such as microneedling (roller and pen) and fractional radiofrequency, effectively and satisfactorily acting to protect and recover scattering skin, regardless of their physical characteristics. Chemical (organic) protectors are not recommended for immediate use after ablative procedures due to their penetration, as they could represent a risk to skins that have suffered any layer loss, in addition to their greater allergy index.

DATA AVAILABILITY STATEMENT

I declare to make my data available.

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