Topical Probiotics in skin barrier function - report on a double-blinded placebo controlled pilot study in stressed skin with live *Cutibacterium acnes*

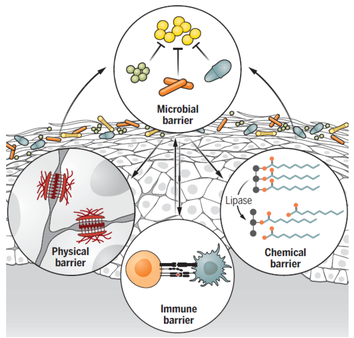
Bernhard Paetzold[1], Ante Karoglan[2], Sven Quist[2 3], Amine Zorgani[1]

[1] S-Biomedic N.V , Turnhoutsweg 30, 2340 Beerse, Belgium
[2] Clinic of Dermatology and Venereology, Otto-von-Guericke University, Magdeburg, Leipziger Strasse 44, 39120 Magdeburg, Germany
[3]Hautarztpraxis Dermatologie Quist

Introduction

SKIN MICROBIOME AND BARRIER FUNCTION

Our skin microbiome form the first barrier that protects us from the exposome through different mechanisms. As highlighted in the figure below by Harris and Grice in their later Science review *(Harris-Tryon et al., Science 376, 940–945 (2022))*, the mcirobial barrier interacts with three other different barriers within the skin: 1) the physical barrier where the skin microbiota also contributes to the differentiation and epithelialization; 2) The chemical barrier where the microbes help into the digestion of the skin's sebum by producing lipases which transform triglycerides into free fatty acids; 3) The immune barrier where the skin microbiome stimulate the innate and the adaptative immune response.



Multiple observational studies showed a significant reduction of the C. acnes population in skin diseases which are associated with skin barrier defects. The microbiome of the skin (most notably *Cutibacterium acnes*) has been identified as an important factor in skin barrier function.

We investigated the application of 2 different topical probiotics (IP-1, IP-2) on the functional capacity of stressed skin in a double-blinded randomized placebo controlled clinical study. The investigational products was composed health associated Cutibacterium acnes strains selected for high production of the antioxidant protein RoxP (Andersson et al. 2019).

Inclusion criteria

Caucasian female volunteers, between 18-55 years old, with all types of skin, having fine to medium wrinkles on the crow's feet, having not applied any products in the last 48hours, ambulatory healthy volunteers, informed and consenting patients who have undergone a general clinical examination attesting to their ability to participate in the study, having signed the consent form for participation, affiliated to a social security system, understanding French language for purpose of understanding study documentation.

Exclusion criteria

Pregnancy or breastfeeding period, showing signs of recent and intense sun or UV exposure, , washing of measurement areas other than with water on the morning of the study, having applied products (exfoliation, peeling, self-tanning, depigmenting or radiance treatment in an institute) for less than a month on the face and mask for less than 2 weeks, having performed cosmetic treatments at a dermatologist (laser, peeling, mesotherapy, injection, etc.) on the face during the last 6 months, suffering from systemic diseases or any dermatosis likely to interfere with the study according to the investigator's assessment, With a chronic or acute condition and / or a topical or general treatment that may influence the results of the study at the discretion of the investigator, Have a history of allergy or hypersensitivity reaction to any component of the study product, Participating in another study or being excluded from another study, Having planned a surgical operation during the study, Cannot be contacted urgently by phone, Being unable to follow the requirements of the protocol, Being deprived of liberty by a judicial or administrative decision, sick in an emergency situation, Being adults protected by law, as well as those admitted to a health or social establishment, provided that the research can be carried out otherwise.

Study Design

The primary goal of this clinical study was to evaluate the safety and efficacy of IP-1 and IP-2 based on clinical signs as well as by measuring the functional capacity of the skin after stripping.

A panel of 20 healthy female volunteers were recruited to the study. The examination was performed in three areas on the forearm of each subject. The functional capacity of the skin was assessed by Spectrocolorimeter®, Corneometer® and Tewameter® measurements. Additionally, the amount of RoxP was determined on the tapes used for stripping.

The study protocol foresaw that each area is stripped with tapes until a TEWL value of 20 is reached and that the number of strips needed is recorded, to protect the subjects a maximum number of 20 strips was set. Unfortunately, none of the volunteers reached the TEWL 20 although the maximum number of strips was performed. Ten volunteers who had after the tape stripping a strong increase in TEWL were included in the Per protocol population , while all volunteers are represented in the ITT population.

At baseline measurements (skin redness, skin hydration and TEWL) were taken on the 3 areas. Afterwards, the areas were stripped, the test products applied and after 1h the measurements were repeated. The topical probiotics and the placebo were applied once daily for 11 days. At day 12 the measurements and striping were repeated.

Figure 01. Different application areas on the volar forearm. Area 1: Investigational product 1 (IP-1), Area 2: Investigational product 2 (IP-2), Area 3: Placebo

Study workflow

Results

In the TEWL measurement we observed a significant better recovering effect after skin stripping in the area treated with IP-2 compared to Placebo. This improved recovery was both after 1 hour of application (p = 0,016 / 0,047 PP/ITT) and also after 12 days of application (p = 0,006 / 0,036 PP/ITT). For IP-1 we did not observe significant differences on these parameters compared to Placebo.
After 1 hour of tape stripping both the areas which received the live probiotics and the placebo returned to redness levels as before the stripping. Indicating that no skin irritation was induced on the damaged skin by the application of live probiotics. No significant differences neither at day 1 nor day 12 were found in regard to skin redness or skin hydration when comparing IP1/2 to the placebo.

The improvements seen with IP-2 on the TEWL measurement correlate very well with a significant increase of RoxP concentration on the treated area of the skin after 12 days.

In conclusion, both investigational products were well tolerated and increase the functional capacity of the skin. We show for the first time that application of carefully selected live C. acnes strains as topical probiotic supports the functional capacity of the skin by strengthening the skin barrier function. We hypothesize that this is due to an increased secretion of the antioxidant RoxP.

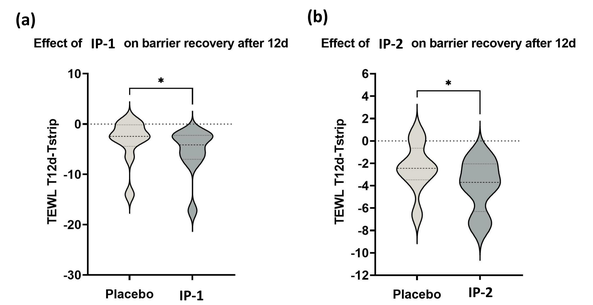


Figure 02. Skin functional capacity after acutely damaging the skin barrier through tape stripping. **(a)** variation of TEWL value between IP-1 (dark grey) and placebo (light brown) after 12 day of products application. **(b)** variation of TEWL value between IP-2 (dark grey) and placebo (light brown) after 12 day of products application.

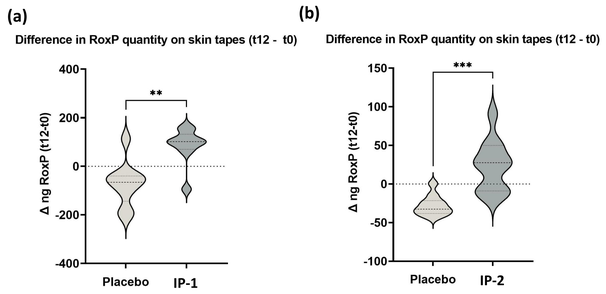


Figure 03. Variation of RoxP concentrations (a) between IP-1 (dark grey) and placebo (light brown) after 12 days of products application. **(b)** between IP-1 (dark grey) and placebo (light brown) after 12 days of products application. RoxP was measured directly from the taps used for the stripping using a monoclonal antibody ELISA.

Conclusions

In conclusion, both investigational products were well tolerated and increase the functional capacity of the skin. We show for the first time that application of carefully selected live C. acnes strains as topical probiotic supports the functional capacity of the skin by strengthening the skin barrier function. We hypothesize that this is due to an increased secretion of the antioxidant RoxP.

Acknowledgments and funding

Sencyr

AGENTSCHAP INNOVEREN & ONDERNEMEN