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Skin Rejuvenation with a Biomimetic Peptide Designed to Promote Desquamation

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~ 5 x 10⁶ cells/day

Stratum Corneum Thickness
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Transit Time
~ 14 days

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Introduction

In old skin the turnover time of the epidermis is increased and desquamation is not homogeneous anymore leading to dry, rough and scaly skin. Desquamation requires the degradation of connections between corneocytes that are formed by cell adhesion proteins. A short protein sequence, called cell adhesion recognition (CAR) site, is essential to hold the cells together. A peptide containing the CAR sequence was designed in order to compete with cell to cell binding and thus to promote desquamation. Tested in a placebo-controlled human study, the peptide was found to reduce the epidermal turnover time to values typical for young skin. This kind of peeling effect resulted in secondary skin benefits such as significantly increased smoothness, reduced wrinkle depth and increased hydration.

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Fig. 1 Desquamation: shedding of dead corneocytes at the skin surface

Summary

The stratum corneum, composed of dead, keratinized cells, called corneocytes, provides the barrier against the desiccating effects of the environment. Continual renewal is an essential feature of maintaining the integrity of this barrier. Keratinocytes undergo a process of proliferation followed by differentiation to new corneocytes. To maintain a constant stratum corneum thickness, the rate of keratinocyte proliferation is balanced by shedding of old corneocytes at the skin surface. This shedding is termed desquamation and occurs by proteolytic degradation of protein links between corneocytes (Fig. 1). The turnover time of the stratum corneum is normally two to four weeks.

Between the 3rd and 8th decades the turnover rate of the stratum corneum is decreased about 30 to 50%. The skin in the elderly becomes dry probably because of a decline in the production of sebum and a lack of normal desquamation. The latter could be the reason for the decreased turnover rate but also for the formation of scales because of abnormally high cohesion between corneocytes. In general, the skin of older people tends to be dry, rough and scaly and more prone to winter eczema.

Keratolytic agents used for chemical peeling such as alpha-hydroxy acids (e.g. glycolic acid) or beta-hydroxy acids (e.g. salicylic acid) lead to visible desquamation and to the acceleration of the stratum corneum turnover. But these proce-
dires often cause side effects such as skin irritation and barrier disruption. Especially for elderly skin that is even more prone to irritation they are not recommended. This article describes a peptide that promotes desquamation and thus leads to a reduced turnover time of the stratum corneum. Topical application of the peptide results in a natural rejuvenation effect without concomitant irritation of the skin.

Biochemistry of the desquamation process

Adherence of cells which is a prerequisite for tissue formation is provided by desmosomes. In the stratum corneum they are called corneodesmosomes. It is a complex of different protein families that link the cytoskeletons of neighbouring corneocytes together. The principal link is made between the heterophilic interaction of desmocollin and desmoglein that belong to the cadherin protein family (Fig. 2). Cadherins typically consist of five tandem repeated extracellular domains, a single membrane-spanning segment and a cytoplasmic region (Fig. 3). Cadherins depend on calcium for their function: only when calcium is bound, the extracellular domain has a rigid, rod-like structure. Sequestration of calcium might explain the keratolytic activity of alpha- and beta-hydroxy acids. Corneodesmosomes undergo a maturation process during stratum corneum renewal. Proteolytic cleavage of the extracellular portion of desmoglein and desmocollin by the stratum corneum chymotryptic enzyme is part of the maturation and absolutely required for desquamation.

Adherence of corneocytes was shown to depend on binding of the extracellular domain EC1 of desmoglein to the EC1 domain of desmocollin. The essential sequences for binding, called cell adhesion recognition (CAR) sites, are arginine-alanine-leucine (RAL) on desmoglein (Fig. 3) and tyrosine-alanine-threonine (YAT) on desmocollin (1). The central alanine is assumed to form a hydrophobic cavity that houses the side chain of a conserved tryptophan residue of the partner protein.

Design of a functional peptide

Peptides corresponding to the desmocollin or the desmoglein CAR site regions were found to specifically prevent desmosomal adhesion (2). These peptides compete with binding between desmoglein and desmocollin (Fig. 4) leading to reduced corneocyte cell to cell adhesion and finally to better desquamation. The active cosmetic ingredient PerfectionPeptide P3 contains the tripeptide with the amino acid residue sequence of the CAR site of desmoglein. The precise chemical structure of the peptide is hexanoyl-Arg-Ala-Nle-NH2. The length of the peptide was restricted to the CAR site in order to get penetration of the peptide into deeper stratum corneum. Again to increase uptake and penetration the N-terminus was linked to hexanoic acid and the resulting amphiphilic peptide derivative was encapsulated into lecithin liposomes. To increase further hydrophobicity and also resistance against proteolytic degradation the residue leucine was replaced by the unnatural amino acid norleucine.
Results and Discussion

Two human studies have been performed to show the efficacy of the ingredient PerfectionPeptide P3. In the first study a test product with 1% of the peptide ingredient was applied twice daily on the forearm during 17 days. Smoothness of the skin was analyzed by laser profilometry (Fig. 5) and by scanning electron microscopy (Fig. 6). Laser profilometry was done on silicon rubber prints of the relevant skin areas. The median depth of the microrelief furrows decreased from 53.9 µm at the beginning of the study to 42.9 µm at day 17 and the complexity value, representing depth and number of wrinkles, from 13.2% to 10.3%. In the non-treated zone the complexity did not change and the median depth even increased slightly. For scanning electron microscopy stratum corneum material was sampled by tape stripping. A rough skin surface is characterized by eruptive layers and broad distance between layers. The skin treated with PerfectionPeptide P3 was at the end of the study clearly smoother than placebo treated and untreated skin.

In another human study gels with 1% or 2% PerfectionPeptide P3 were tested over four weeks on 20 female subjects of 41 to 58 years old. The test products were applied twice daily. Skin hydration and roughness and epidermal turnover time were measured on the inner side of the forearm. Wrinkle depth was recorded in the crow feet area where only the gel with 2% ingredient was applied. The same area of the other eye was treated with the control gel. Skin roughness and wrinkle depth were measured with the PRIMOS device, a non-contact optical 3D measurement system. Skin hydration clearly improved after two weeks by 20% with the 2% test product and by 10% with the 1% test product and stayed at the same level until the end of the study. After four weeks application the skin treated with both concentrations of the test ingredient was compared to the untreated area significantly smoother (Fig. 7). The wrinkle depth in the crow feet area was clearly reduced compared to the zone treated with the placebo after four weeks application (Fig. 8). The epidermal turnover time decreased significantly from 22.7 days in the untreated area to 20.5 days in the zone treated with the 1% test product (Fig. 9). The gel with 2% PerfectionPeptide P3 showed about the same effect (20.8 days).

Reduction in the turnover time proves the interference of the peptide hexanoyl-Arg-Ala-Nle-NH2 with the desquamation process. This leads to a peeling effect with the mentioned skin benefits such as increased smoothness, reduced wrinkle depth and increased hydration. Neutralization of the interaction of the cadherin proteins desmocollin and desmoglein is the principal mechanism of action of PerfectionPeptide P3. Comparison of gene expression in cells of young and aged skin showed that the gene for desmocollin is clearly overex-
pressed in old skin (3). Skin biopsies from a 28 and a 69 years old donor were analyzed by two different techniques, SAGE-Analysis and Micro-Array technology. Desmocollin was found 10 times overexpressed in old skin analysed by the SAGE method and 1.33 times overexpressed when analysed by the Micro-Array method. This gene expression analysis identified desmocollin as ageing marker. An ingredient for skin rejuvenation, whose mechanism of action is based on the inhibition of the activity of desmocollin and desmoglein, is therefore very plausible. Peptides are normally of limited value as actives for skin care because they can hardly penetrate into deeper skin layers. But for interference with desquamation, a process that happens at the skin surface, peptides are the perfect actives.

References


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