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In Vivo Confocal Fluorescence Imaging of Skin Surface Cellular Morphology: A Pilot Study of its Potential as a Clinical Tool in Skin Research

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The cellular morphology of the stratum corneum was studied *in vivo* using a novel imaging technique that uses confocal fluorescence microscopy in combination with topical application of a fluorescent contrast agent. Images obtained with this method show a strong variation in skin surface cellular morphology among healthy subjects. The results of several clinical studies suggest that cellular morphology is affected by the efficiency of the process of desquamation. As such, cellular morphology shows strong potential to serve as an indicator of skin health that yields mechanistic insight into the origins of skin ailments, such as xerosis, and the effectiveness of their treatments.

all parameters were observed with 2 weeks of moisturizer use on lower leg skin with marked xerosis compared to an untreated control. The same moisturizer had a similar effect on milder xerosis of the forearm, and showed a greater decrease than a moisturizer with lower glycerol content. Increases in skin hydration, as measured with a corneometer, were also seen in both clinical studies, and corresponded well with D-Squame® results. Differences in the degrees of scaling between these two anatomical sites were also detected with this technique. In a previous publication, the same technique was shown to be repeatable and reproducible; in the current article its correlation with clinical observations of scaling or flaking skin has been demonstrated.

Image Analysis of Skin Scaling Using D-Squame® Samplers: Comparison with Clinical Scoring and Use for Assessing Moisturizer Efficacy

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The severity of scaling disorders can be evaluated objectively using the D-Squame® technique coupled with image analysis. The parameters of scaling derived using this approach need to be clinically relevant and should have greater discrimination than visual grading. Improvements to an existing method that fulfil these requirements are presented. Three scaling parameters were calculated using image analysis of digitized video-captured images of obliquely lit D-Squame® samples. These parameters were compared to clinical scores of scaling made by five observers from photographs of the same areas sampled with D-Squame®. In addition, two clinical studies were carried out to assess moisturizer effects on different degrees of xerosis, and to compare two different moisturizer preparations. The three scaling parameters gave correlation coefficients, r , between 0.6 and 0.75 when compared with global clinical scores of scaling. Significant reductions in

Investigating Hair Properties Relevant for Hair 'Handle'. Part I: Hair Diameter, Bending and Frictional properties¹

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The expert working group 'Hair Care Products' of the DGK currently conducts a wide study to contribute to the understanding of how single hair fibre and hair collective properties contribute towards hair 'handle' and 'feel'. During the first stage of this study four hair types were selected from a large group of individual European hair braids, according to either similar or widely different panel ratings for handle. Against the background of the panel test and the state of the literature the working group readily identified the bending properties of single fibres interacting in the tress as a fibre collective and fibre friction as being of central relevance for hair 'handle' and 'feel'. Fibre diameters of the hair types were determined by Optical Fibre Diameter Analyzer and by weighing. From these data mean ellipticity and bending stiffness distributions were calculated. Single fibre friction was determined by the capstan method in the root, middle and tip regions. Significant differences were determined between the hair

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types in diameters, ellipticity, bending stiffness and friction. The results lead to conclude that 'handle' is perceived as inferior when the hair is thick and bending stiffness thus high. For such hair differences in handle rating are related to differences in friction, namely in the tip region. For thin and thus 'soft' hair fibre friction seems to play only a minor role.

A Method for the Determination of N-nitrosodiethanolamine in Personal Care Products – Collaboratively Evaluated by the CTPA Nitrosamines Working Group

C. Flower*, S. Carter†, A. Earls†, R. Fowler‡, S. Hewlins§, S. Lalljie¶, M. Lefebvre**, J. Mavro**, D. Small†† and N. Volpe‡‡

A procedure for the determination of N-nitrosodiethanolamine (NDELA) in personal care products was evaluated in collaborative studies by member organizations of the United Kingdom's Cosmetic Toiletry and Perfumery Association (CTPA) and LGC Limited, formerly known as the Laboratory of the Government Chemist (LGC). Samples were prepared depending on the matrix of the cosmetic product: aqueous samples were prepared by diluting in water followed by solid-phase extraction; emulsions, oils and solid materials were dissolved in dichloromethane and extracted with water. NDELA was separated from the sample matrix using reverse-phase liquid chromatography. The N-nitroso bond was cleaved by photolysis to give nitrite, which was colorimetrically quantified. The nitrite functional group reacted with sulphanilamide in an acid medium to form a diazonium ion which was then coupled with N-(1-naphthyl)ethylenediamine dihydrochloride according to the Griess reaction to give a purple-coloured azo dye that absorbed at 540 nm. Compared with other published methods for NDELA, the method described here is quick and easy to use. It has the required sensitivity and specificity, and can accurately and reliably quantify NDELA in a wide range of personal care product matrices.

Nitrosodiethanolamine in Personal Care Products – Collaboratively Evaluated by the CTPA Nitrosamines Working Group IJCS 28 (1) 21-34 (2006)

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reverse-phase liquid chromatography. The N-nitroso bond was cleaved by photolysis to give nitrite, which was colorimetrically quantified. The nitrite functional group reacted with sulphanilamide in an acid medium to form a diazonium ion which was then coupled with N-(1-naphthyl)ethylenediamine dihydrochloride according to the Griess reaction to give a purple-coloured azo dye that absorbed at 540 nm. Compared with other published methods for NDELA, the method described here is quick and easy to use. It has the required sensitivity and specificity, and can accurately and reliably quantify NDELA in a wide range of personal care product matrices.

The Suppressive Effect of Apricot Kernel Extract on 5 α -Androst-16-en-3-one Generated by Microbial Metabolism*

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Body odours are generated from dead skin cells and secreted materials, such as sweat and sebum, through the metabolism of microorganisms living on the skin. Volatile steroids, key compounds in body odours, are also generated through the metabolism of microorganisms. These volatile steroids strengthen the intensity of the overall body malodour and are sensed differently by males and females. Females are more sensitive than males to volatile steroids, especially 5 α -androst-16-en-3-one (androstenone). To regulate body odours that are especially unpleasant for women, we devised an androstenone-generation model using the metabolism of *Corynebacterium xerosis*, which is one of the bacteria living on the axillary skin. Using this model, we studied the suppressive effect of plant extracts on the generation of androstenone. We found that apricot kernel extract (AKE) had the most positive effect among the plant extracts to which we applied the model. However, although AKE did suppress androstenone generation, it did not show any bactericidal effect. Using the cell-free system, AKE also suppressed the generation of androstenone. In conclusion, we found that AKE suppressed the generation of androstenone, which is especially unpleasant for women, and the mechanism was not bactericidal but metabolic inhibition. The results of these studies provide new understanding of the regulation of androstenone, which, in turn, should lead to the development of novel deodorant systems.

Studies of Compounds that Enhance Sphingolipid Metabolism in Human Keratinocytes¹

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Several products are known to inhibit the biosynthesis of ceramides and glucosylceramides, but very few stimulate this process. We studied the influence of a hydrolysate of potato proteins (Lipidessence[®]) in vitro on the sphingolipid metabolism of normal human epidermal keratinocytes. By measuring growth with the thymidine uptake assay, it was seen that Lipidessence[®], added in the culture medium up to an 8% concentration, did not change significantly the proliferation rate of keratinocytes, but beyond this

concentration a progressive dose-dependent inhibition of growth was noticeable. Following incubation of cells with the product at 5% and 10% concentrations for 2 days, the lipids were extracted. The different lipid classes were separated by fractionation on columns of aminopropyl silica gel and analyzed by high-performance thin-layer chromatography. When keratinocytes were cultivated in the presence of Lipidessence[®], the biosynthesis of cholesterol, phosphatidylcholine, phosphatidylserine and gangliosides was stimulated, and a major increase was noticeable in the biosynthesis of free fatty acids, free ceramides, glucosylceramide and sphingomyelin. Radioactive [¹⁴C]-serine was used as a precursor of sphingoid bases to study sphingolipid biosynthesis. After migration of lipid fractions on thin-layer plates, autoradiography showed that free ceramides and glucosylceramide were labeled, thus suggesting that de novo biosynthesis was accounting for the increased cellular content in sphingolipids.

Adaptive Response of the Skin to UVB Damage: Role of the p53 Protein

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Different adaptation mechanisms like heat shock response, cell cycle arrest and DNA repair, melanin pigmentation and thickening of the epidermis are presentation and thickening of the adverse effects of solar UV irradiation. When DNA damage is beyond repair, cells undergo apoptosis to prevent their replication. We discuss the current knowledge on these different adaptation mechanisms to UVB damage, the most energetic fraction of solar UV that reaches the skin. As p53 protein, the guardian of the genome, plays a key role in protective response to genotoxic damage, its role in this adaptive response of the skin to UV will be further discussed.