

Claim substantiation and efficiency of hydrating body lotions and protective creams

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In the present work a practical claim substantiation study is shown by the example of 5 commercially available body lotions. Their efficacy with respect to effects on transepidermal water loss (TEWL) and stratum corneum (SC) hydration of ageing skin has been examined. Results were obtained after single and repeated application (14 days, 2 × a day). The best performing product was then selected and further tested for its potential effects on sodium lauryl sulfate (SLS)-damaged skin. This was done in a younger population and the recovery of the impaired barrier function was followed by TEWL measurements. The selected body lotion had a high efficacy, improving both the TEWL and SC hydration of ageing skin by more than 30%. When applied to SLS-damaged skin, the product was able to improve skin barrier repair in comparison with physiological barrier repair. The results of this study show that a combination of non-invasive objective measurements can be used to substantiate product claims. Claims can be made with respect to protective and preventive properties of products, but also as to effectiveness of topical skin treatment in the case of abnormal barrier function or barrier restoration.

Key words: skin barrier; stratum corneum; claim substantiation; SLS-treatment; TEWL; hydration; bioengineering methods; barrier creams; cosmetics; EU legislation; European Cosmetics Directive. © Munksgaard, 2000.

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Skin dryness associated with a rough and scaly appearance is one of the major problems of ageing skin (1). The characteristic dry feature is often correlated with impaired barrier, decreased stratum corneum (SC) hydration and increased incidence of irritant contact dermatitis (ICD) (2). It is a common phenomenon that the skin becomes drier with ageing. The origin of skin dryness can often be brought back to the level of a disturbed SC. The human SC is composed of corneocytes enriched with keratin and embedded in a lipid-rich matrix. This unique organisation is known as the 'brick and mortar' model (3–4). The lipids, mainly consisting of ceramides, cholesterol, free fatty acids and cholesterol sulfate, are combined in structured bilayers (5–6). Phospholipids, major components of most other biological membranes, are not present. The intercellular lipids are responsible for the permeability barrier properties of the SC. Removal of the upper layers of the skin, as well as of the SC lipids, e.g., by tape stripping or treatment with aggressive surfactants, will lead to a significant increase in transepidermal water loss (TEWL) (7–8).

In several skin pathologies, including atopic dermatitis (9), some forms of ichthyosis (10) and psoriasis (11), formation of the cornified layer deteriorates, passive water flux increases and alterations in specific lipid content occur. It is common knowledge that atopic subjects are more prone to develop dry skin and ICD than other groups.

In all such cases, efficient dermato-cosmetic products could be helpful to protect the skin against external attacks and to prevent it from drying out (12–13). Topical application of hydrating lotions and protective creams to hydrate the SC and prevent or reduce the penetration and percutaneous absorption of harmful irritants is not new (14), but proof of their efficacy is now a new subject due to cosmetic legislation. The 6th Amendment to the European Cosmetics Directive [93/35/EEC] came into force on the 1 January 1997 (15). It includes a demanding clause for claim substantiation of marketed cosmetics. Proof of efficacy can be done in different ways (16) but testing on human volunteers is the preferred one. Human testing on a target group of volunteers may include

in use tests with auto-evaluation (questionnaire) or evaluation by an experimenter (visual, tactile, olfactory, etc.). However, both methods are very subjective. By introducing non-invasive instrumental measuring techniques that are applicable on humans, properties of cosmetic and dermatologic products can be assessed in an objective way, making comparisons between results possible and allowing the detection of subtle changes in skin morphology and function otherwise not detectable by sensorial means.

In this study, as an example of how to run an

efficiency study on hydrating body lotions or protective creams, 5 commercially available body lotions have been evaluated for their efficacy on SC hydration and barrier function of aged skin using non-invasive methods. Short- and long-term tests have been performed in order to be able to select a well-performing body lotion. The latter was then applied to experimentally induced scaly skin in a younger population. This experiment was designed to investigate whether the product applied was able to assist in a more rapid recovery of a disturbed barrier function than would be the case under nor-

Table 1. Qualitative compositions of the body lotions tested, as listed on the package

Lotions	Ingredients:	
lotion A →anionic o/w emulsion	aqua paraffinum liquidum stearic acid cetearyl alcohol caprylic/capric triglyceride propylene glycol panthenol carbomer triethanolamine sodium PCA sodium lactate fructose	glycine niacinamide urea sodium benzoate inositol lactic acid imidazolidinyl urea parfum methylparaben propylparaben bisabolol
lotion B →non-ionic o/w emulsion	aqua caprylic/capric triglyceride propylene glycol carbomer polysorbate 80 mono oleate sorbitan 80	sodium hydroxide parfum methylparaben propylparaben EDTA
lotion C →anionic o/w emulsion parfum free	aqua 10% urea glyceryl stearate/PEG-100stearate acetylated lanolin alcohol stearic acid glyceryl monostearate SE	isopropyl palmitate petrolatum methylparaben propylparaben carbomer
lotion D →non-ionic o/w emulsion	64.1% aqua (thermal water) 7% glycerin 5% shea butter paraffinum liquidum octyldodecanol PEG-5 glyceryl stearate 3% urea dimethicone cetyl acetate acetylated lanolin alcohol cetyl alcohol	0.5% palmitoyl hydrolyzed milk protein 0.2% bisabolol carbomer 0.2% allantoin triethanolamine steareth-10 parfum phenoxyethanol methylparaben triclosan
lotion E →non-ionic o/w emulsion	aqua glycerin octyl stearate glyceryl stearate decyl oleate ceteareth-20 pentaerythrityl stearate/caprate/ caprylate/adipate behenyl alcohol cetearyl alcohol cetyl palmitate ceteareth-12	allantoin parfum triclosan tetrahydroxypropyl ethylenediamine carbomer PEG-8 2-bromo-2-nitropropane-1,3-diol disodium EDTA tocopherol ascorbyl palmitate ascorbic acid citric acid

mal conditions without any treatment. Positive results can support stronger claims than those based only on hydration measurements. Medical preventive claims can also be supported by these testing methods, as can the most efficient product be selected.

Materials and Methods

Efficacy measurements

The effects on TEWL were measured with a TEWA-meter[®] TM 210 (Courage+Khazaka Electronic GmbH, Köln, Germany). The results were expressed in g/hm². The Corneometer[®] CM 820, also from Courage+Khazaka Electronic GmbH, Köln, Germany, was used for capacitance measurements. Expressed in relative units, these measurements evaluate the hydration state of the upper layers of the skin. The measurements were performed in an acclimatized room with a mean relative humidity of 50.2±6.9% and a mean room temperature of 21.6±0.6°C. They were carried out under standardized conditions as described earlier (17).

Lotions

The qualitative compositions, as indicated on the packages, of all 5 selected commercially available body lotions are summarized in Table 1. The products were renamed by the symbols A, B, C, D and E. All body lotions tested were o/w emulsions and were marketed for their specific lipid components, emollients and moisturisers. Some of the claims made concerned softening of rough and scaly skin types, rehydrating of dry skin and even improvement of an impaired skin barrier. Uniform numbered flasks of the 5 selected lotions were used and the tests were performed double blind.

Volunteers and experimental design

Efficacy measurements on ageing skin. The tested population consisted of 12 healthy female volunteers with a mean age of 58±9 yrs. A dermatologist determined the skin types as normal to dry. All test subjects were asked not to use cleansing or skin care products on the inner forearms for 1 week prior to and during the study. Efficacy measurements were carried out before and after the application of the lotions (short-term test; 1, 2 and 3 h after a single application). This procedure was followed by a long-term test of 2 weeks, when the emulsions (A, B, C, D and E) were applied 2× daily for 14 consecutive days. TEWL and capacitance measurements were then performed 12 h after the last application (day 15). The left volar

forearm skin was exposed to a constant quantity (3.8 mg/cm²) of the 5 products. This procedure was carried out double blind and randomized, while identical non-treated sites on the right volar forearm served as controls.

Recovery of SLS-damaged skin. A group of young women ($n=13$, 18–30 years) were selected. The study was approved by the Ethics Commission of the Academic Hospital, VUB-Jette. After visual examination of the skin, the volunteers signed a written consent form and underwent a 24-h patch test with 1.25% SLS aqueous solution. 160 µl of this solution (Sigma, St. Louis, USA, 99% purity) was absorbed on a filter paper (diameter: 20 mm, pore size 200 µm, Sartorius[®], Goettingen, Germany) that was applied on the skin of the ventral surface of the forearms. Occlusion was obtained (Dermalock[®] HDP Medical, Waasmunster, Belgium), and the patches were fixed by self-adhesive Fixomull[®] stretch (Beiersdorf, Hamburg, Germany). 'Blank' patches with 160 µl of pure water were located on the right volar forearm and served as controls. After removal of the patches, the forearms were rinsed with pure water and 24 h after air exposure TEWL measurements were performed. This time interval was respected in order to eliminate the effect of water itself on the skin. In the short-term test, the selected body lotion was applied 1× and TEWL measurements were done after 2 h. During the long-term test of 14 days,

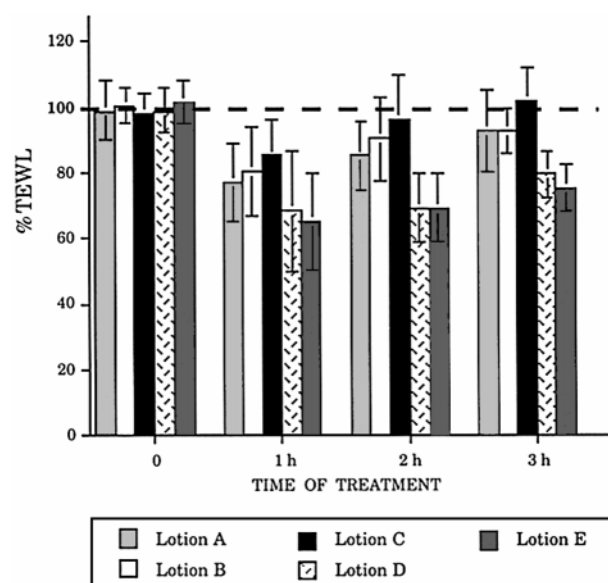


Fig. 1. TEWL values ($n=12$; mean age 58±9 yrs) as measured before and after application of the 5 selected body lotions on the left forearm. For the short-term test of 3 h the values measured are given as mean±SD, expressed as a percentage of the appropriate control values on the right arm, arbitrarily set to 100%.

Table 2. The average TEWL of the forearm after a 14 day-test of 2 applications a day of the 5 selected body lotions

Lotions	Mean $\pm$ SD
lotion A	73.0 $\pm$ 7.8
lotion B	84.2 $\pm$ 9.5
lotion C	92.3 $\pm$ 5.7
lotion D	63.2 $\pm$ 13.2
lotion E	64.4 $\pm$ 7.3

The effect on the barrier function of the skin is given 12 h after the last application of the lotions. The results are expressed as % (mean $\pm$ SD, $n=12$) in comparison with the control values (=100%).

the product was applied 2 $\times$ a day and TEWL measurements were performed after 5, 8 and 15 days, each time 12 h after the last application of the lotions.

Statistical analysis

TEWL and capacitance values of the left forearm were expressed as a % of the values obtained for the appropriate control test sites on the right forearm, arbitrarily set to 100%. They are presented as mean values $\pm$ standard deviation (SD). Analysis of variance (M)anova and Student's paired *t*-tests were performed. Differences are significant if $p<0.05^*$ and highly significant if $p<0.01^{**}$.

Results

In Fig. 1 and Table 2 the TEWL values as measured on ageing skin exposed to the 5 commercially available body lotions are shown. The capacitance values are summarized in Fig. 2 and Table 3. Before any application of emulsions was made, the test sites on the left and right forearms were measured. No significant differences in TEWL ($p=0.243$ for the 5 identical rectangles determined on 1 forearm and $p=0.635$ for the corresponding test sites on both forearms) nor in SC hydration ($p=0.275$ and $p=0.084$, respectively), between the test sites of both forearms of the participating volunteers, could be observed. These results indicated that the left forearm could be used for application of the emulsions to be tested, while the right forearm served as control.

Statistical analysis of the results shown in Fig. 1 revealed a significant effect on the barrier function from all 5 lotions. The decrease in TEWL was identical for lotions D and E, but was significantly higher than the effects observed for lotions A, B and C. After a single application of the lotions, the effect of lotion A was also significantly higher than for lotion C. Similar observations could be made after application of the emulsions for 14 days (Table 2). For lotions D and E, a positive effect on

the barrier function of more than 30% could be observed, which was significantly better than for the other 3 lotions.

In Fig. 2 the results have been summarized for the effect of the 5 lotions on SC hydration. Lotion C lost its hydrating capacity after 3 h, in comparison to the non-treated control arm. Paired Student *t*-tests showed that lotions D and E had a significantly greater effect on skin hydration than lotions A, B and C. Lotions A and B were in the latter case also more efficient than lotion C in hydrating the skin after a single application of the lotions. From the results of the 14-day test a similar conclusion could be drawn (Table 3). 12 h after the last application, lotions D and E still sustained an improvement in SC hydration of more than 30%, in comparison with the appropriate values on the right control arm.

At that moment, it was clear that both products could serve in the study with SLS-damaged skin. Product E, however, was preferred since it was cheaper than lotion D and its qualitative composition available in detail. In further work, SLS patches were applied on the skin of the left forearm of younger volunteers to induce ICD and a rough and scaly appearance of the skin. In Fig. 3, the TEWL values before ($t_1=0$) and after ($t_2=0$) SLS treatment are given, showing the expected increase in TEWL values after SLS damage. 2 h after a single application of lotion E no improvement in

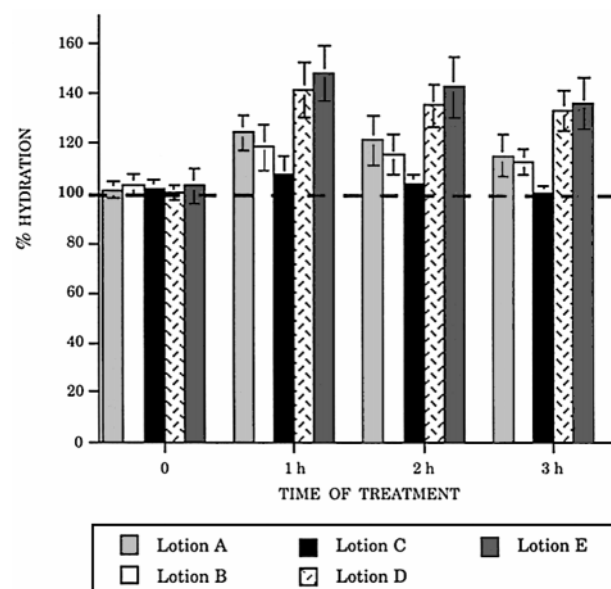


Fig. 2. Effect on skin hydration of the 5 selected o/w emulsions after topical application on the left forearm. Mean values $\pm$ SD are given for each data point during a short-term test of 3 h ($n=12$; mean age 58 ± 9 years). The results are expressed as a % of the values measured on the right arm on which no lotions were applied, arbitrarily set to 100%.

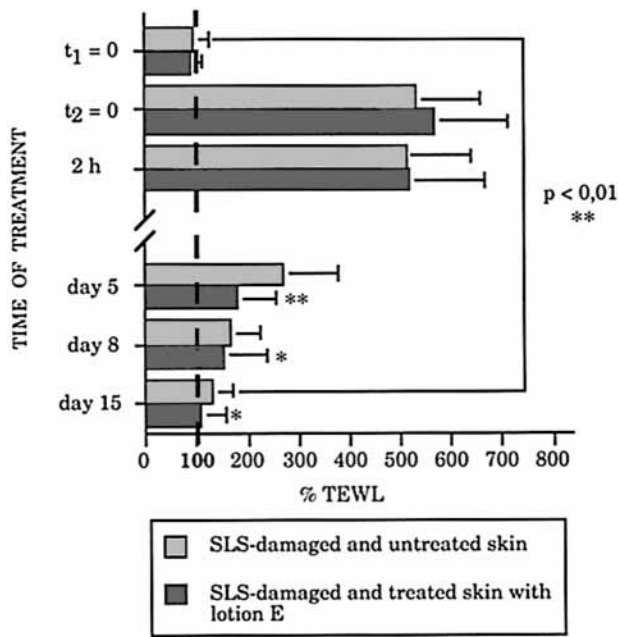


Fig. 3. The effect of topical application of lotion E on the TEWL values of SLS-damaged forearm skin in comparison with physiological barrier repair ($n=13$, 18–30 years). $t_1=0$: TEWL measurements before application of SLS patches; $t_2=0$: TEWL values after SLS treatment. Each bar represents mean TEWL $\pm$ SD, expressed as a % of the values of the 'blank' control sites on the right forearm, arbitrarily set to 100%. A short-term (2 h) and a long-term test were carried out. In the latter case, measurements were performed 12 h after the application of lotion E on days 5, 8 and 15. Statistical evaluation was made by comparison using the Student paired t -test; $p < 0.01^{**}$, $p < 0.05^{*}$. After repeated application of lotion E, significant effects on TEWL could be detected in comparison with the TEWL values of damaged, but untreated, skin. On day 15, there was no longer any difference between the TEWL values of the treated damaged test area and the untreated undamaged control skin ($p > 0.05$). On the contrary, the damaged untreated skin still showed an increased TEWL ($p < 0.01$).

TEWL could be observed, when values of treated skin were compared to those of damaged but untreated skin.

During the long-term test, however, clear differences could be measured, namely in comparison to the values observed for SLS-damaged, untreated skin, repeated application of lotion E significantly improving the TEWL values found for damaged and treated skin. Decreases in TEWL were statistically significant on days 5, 8 and 15. On the last day of the experiment, it was observed that repeated application of lotion E supported the SLS-damaged skin to complete recovery. Indeed, no significant differences could be detected anymore by Manova between the TEWL values obtained at day 15 for damaged treated skin and the non-treated control skin, measured at $t_1=0$.

The normal physiological barrier repair of an SLS-damaged but untreated control area was also

Table 3. Hydrating capacity of the 5 selected lotions

Lotions	Mean $\pm$ SD
lotion A	117.7 $\pm$ 11.1
lotion B	115.3 $\pm$ 7.9
lotion C	106.6 $\pm$ 3.7
lotion D	127.9 $\pm$ 9.2
lotion E	132.0 $\pm$ 7.3

The results are expressed as % (mean $\pm$ SD, $n=12$) in comparison with the untreated control values arbitrarily set to 100%. The measurements were carried out 12 h after the last application of the lotions following a long-term test of 14 days.

investigated. At day 5, this control area already showed a barrier recovery of 50%, together with clinical improvement in the scaly appearance and redness of the skin that had almost disappeared. However, on day 15 the TEWL values of the SLS-damaged and untreated skin were still statistically increased, when compared to the undamaged control values.

Discussion

According to the new cosmetic legislation [93/35/EEC], proof of claims made for cosmetic products must be provided in the technical dossier and kept ready for inspection by the competent authorities (15). This concept is not really new in the sense that Council Directive 84/450/EEC already dealt with misleading advertising of cosmetic products. The novelty lies particularly in the fact that proof of efficacy is now an integral part of the technical dossier and can form the subject of discussion between the parties concerned, e.g., consumers and industry, authorities and industry, as well as between firms themselves.

Although it is clear that efficacy testing is asked for by European and national regulations, no provisions have been made in the law as to how this should be done. Some private initiatives have been taken, resulting in the foundation of EEMCO (European Group for Efficacy Measurements on Cosmetics and Other Topical Products), a group of experts providing guidance documents with respect to efficacy measurements to all parties involved (18–24). Colipa (European Cosmetic, Toiletry and Perfumery Association) too has published practical guidance with respect to claim substantiation but this document keeps to quite general terms (16).

How should one now proceed, practically, in the case of body lotions and protective creams claiming positive effects on SC hydration and barrier function? When hydrating properties need to be substantiated, the simple short- and long-term test, based on the measurement of SC hydration

by changes in its electrical properties, as used in this work, can be carried out. When claims with respect to barrier function need to be substantiated, the same short- and long-term tests as just mentioned could be applied, but measurements are then based on quantification of the TEWL. The short-term test allows in most cases high discrimination, whereas the long-term test usually shows better the real effects the products have on the upper layers of the skin when the product is no longer present (22).

In both types of test, it is advisable to work with a target group of elderly volunteers. Indeed, it is known that aged skin is fragile and changes in barrier properties and hydrolipidic film make it difficult to cope with unbalanced external circumstances, such as temperature changes, cold, wind and relative humidity (2). Moreover, frequent use of soaps and aggressive detergents is often one of the main causes of the induction of an ICD (25). The feature of an altered epidermal barrier function, as seen by higher TEWL values, is typical for such persons. For these reasons, it is clear that hydrating and barrier protective properties of cosmetic products will be better and more easily seen in elderly people than in a young population with normal skin.

In this work, the approach has been chosen to use the combination of both non-invasive methods, namely the measurement of SC hydration, based on electrical properties of the skin, and the quantification of TEWL. Both techniques were applied according to EEMCO guidelines (17, 22, 24). A product is considered as well formulated when its application increases in a significant way SC hydration while the TEWL is not concomitantly increased, but, on the contrary, remains the same or, even better, is significantly decreased. By using such a combined approach, interpretation of the results obtained is possible.

Products D and E both gave high hydration of the SC of elderly volunteers. This increase in water content could, however, be due to different reasons (26), including: (i) the effect of specific hydrating ingredients such as natural moisturizing factors or humectants capable of retaining or binding the water of the applied emulsion or coming from the deeper skin layers; (ii) the occlusive or semi-occlusive properties of lipid components providing a 'shielding' effect and preventing the loss of water coming from the deeper layers; (iii) the effect of some specific active ingredients on the intercellular cement. This effect can be either positive, e.g., in the case of lipidation of the intercellular cement by lipid-rich liposomal preparations, or negative, e.g., keratinolytic effect, weakening of the bilayer structure, etc. When SC hydration increases and the

TEWL measured remains constant or decreases, as is the case in our study for products D and E, the experimenter can be confident that the increase in SC hydration is not due to a damaged or unbalanced skin barrier. The latter would result in an increased loss of the user's own skin water content. Rather, it proves that the formulated product has very good hydrating properties.

Stronger claims for product E can be made than for products A, B and C. In an additional experiment, it was shown that cosmetic product E was able to improve the recovery of damaged skin in comparison to physiological barrier repair. The volunteers were a homogeneous group of young people, though they could have been a group of any other age. The important factor is that the experimental design concerned damaged skin. The anionic surfactant SLS was used to damage the skin, since it is widely known as a model irritant (25, 27). Topical treatment with SLS induces perturbation of skin barrier function, leading to increased percutaneous penetration of other chemicals applied to the skin (28–29). It could be clearly shown that repeated application of product E improved the TEWL of SLS-damaged skin and supported complete recovery before normal physiological repair was completed. Product E is thus not only able of providing a good hydration of the SC but also has healing properties, by improving the recovery of damaged barrier function and re-equilibration of skin properties.

A new generation of barrier and protective creams is now coming onto the market. The concept of 'shielding the skin' has changed during the last few years to a new approach of re-establishment of the proper lipid components of the SC barrier. It is suggested that such preparations should include the disturbed and missing lipid substances which will, after 'penetration', be able to restore the physiological bilayer structures (12, 30). The published data on the effects of such formulations, however, are very conflicting. Whereas some publications claim a positive effect of ceramide, cholesterol and fatty acid-containing mixtures on the normalization of TEWL of surfactant (31, 32) or solvent (32–34)-induced damaged skin or after tape stripping (32, 35), these results could not be confirmed by our group (36–38) though promising effects are now seen with ceramide-containing liposomal formulations (39). The increased availability of skin-identical lipids now makes it possible to formulate emulsions with a lipid phase similar to the lipid bilayers found in the intercellular spaces of the SC. However, the claims made for such preparations also need to be substantiated in an acceptable way. The combined strategy, as proposed in this paper, has already been successfully

applied to this new generation of products (39) and shown to be a useful tool in claim substantiation of hydrating and protective topical products.

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