A Review of the Range of Effects of Niacinamide in Human Skin

Paul J. Matts[1], John E. Oblong[2] and Donald L. Bissett[2]

Keywords: Niacinamide, nicotinamide, skin, ceramide, ageing, differentiation, barrier

Abstract

Niacinamide (also known as nicotinamide, 3-pyridinecarboxamide) is the physiologically active form of niacin or vitamin B3, the deficiency of which results in the nutritional disease pellagra with distinct cutaneous manifestations. Since its discovery and isolation, a host of dermatological therapeutic benefits and mechanisms have also been ascribed to this essential water-soluble vitamin when used as a topical agent. These include its apparent role as an anti-acne active, an up-regulator of epidermal sphingolipid synthesis, an up-regulator of markers of epidermal differentiation and dermal proliferation (with concurrent stratum corneum barrier benefits), and as a moderator of photoimmunesuppression and accompanying tumor genesis. More recently, fresh evidence points to a role in modifying the cosmetic appearance of skin through suppression of epidermal melanosome transfer with subsequent effect on skin pigmentation and a role in modifying epidermal surface topography. The mechanisms for these cutaneous effects are still unclear. However, since niacinamide is an important precursor of NADH and NADPH, it has been postulated that topical application of niacinamide can promote this reported broad spectrum of activity through local correction of homeostatic balance of these two nucleotide coenzymes. As there has been a dramatic increase in research into and use of niacinamide in recent years, this review will cover the current scope of knowledge of this important vitamin, including mechanistic understanding and cutaneous physiological activity.

History of Niacinamide

Niacinamide is the amide of vitamin B3, also known by the pseudonym »Vitamin PP«, that is, »Pellagra-Preventive«. The name is not without meaning. The first case of pellagra was reported in the U.S. in 1902; four decades of a pellagra epidemic followed during which, in states south of the Potomac and Ohio rivers, some 3 million cases and 100,000 deaths were reported [1]. Pellagra patients presented with a variety of debilitating symptoms including, significantly, a spectrum of cutaneous lesions. Tragically, this led to the public exclusion of thousands of victims, who came almost exclusively from poor, rural, working-class families who fed themselves on a bland staple diet of cornmeal, molasses and fatback. Joseph Goldberger, a Hungarian emigrant who established himself as a renowned clinical epidemiologist, reversed steadfast medical opinion that pellagra was an infectious, communicable disease. He proved that simple dietary supplementation could both prevent and cure pellagra. In 1927, after 13 years of work, Goldberg persuaded the American Red Cross to distribute dried yeast to Mississippi flood victims and, thus, prevented a further devastating epidemic. It was not until 1937 that nicotinic acid and its derivatives (including niacinamide) were shown to be the elusive »PP« factor. By 1945, Goldberger's legacy was permanent; public education had changed forever the poor diet of the South and pellagra was eliminated in the United States.

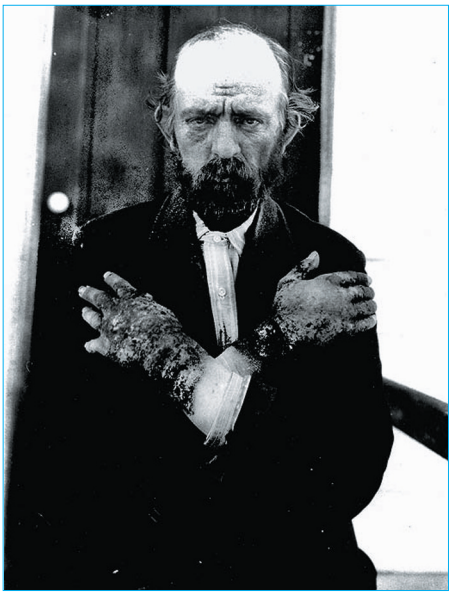

Fig. 1: Pellagra suffer, US South, early 20th century

The authors acknowledge the Waring Historical Library of the Medical University of South Carolina, Charleston, SC

Physiological Role of Niacinamide

The substituted pyridine derivative niacinamide is an essential constituent of the oxidoreduction coenzymes nicotinamide adenine dinucleotide (NAD) and nicotinamide adenine dinucleotide phosphate (NADP) **(Fig. 2)**. During glycolysis and the TCA cycle, 10 molecules of NAD^+ (per molecule of glucose) are reduced to 10 NADH by the transfer of a hydride ion to the 4-position of the niacinamide ring. The hydride ion of NADH serves effectively as an energy storage unit, giving up a pair of high-energy electrons to the mitochondrial electron transport chain when needed. In this process of oxidative phosphorylation, electron pairs are transferred from NADH to a final acceptor (oxygen) via a series of electron carriers. This transfer of electrons is thermodynamically

[1] The Procter & Gamble Company, Rusham Park Technical Centre, Egham, Surrey, UK
Email: matts.pj@pg.com
[2] Miami Valley Laboratories, Cincinnati, Ohio, USA

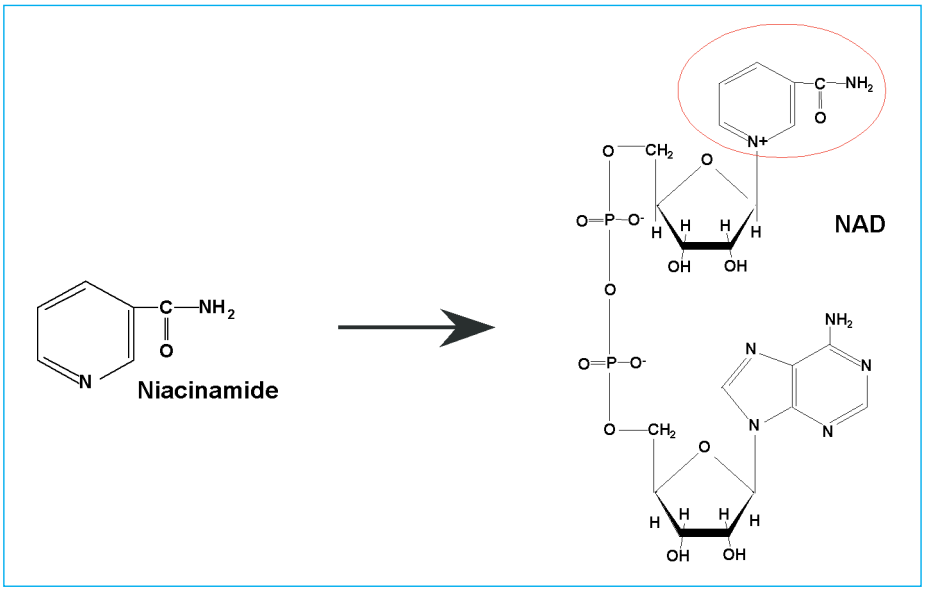

Fig. 2: Structure of niacinamide and inclusion within NAD molecule

favorable, i.e., ΔG is negative, and is coupled to the pumping of protons out of the mitochondrial matrix. The flow of protons back into the matrix, in turn, catalyses the production of ATP by F_0F_1 ATP-synthase. Total energy yield (ΔG') for this process is high (–52.7kcal).

Whereas NADH is involved in catabolism, NADPH tends to serve as an electron (hydride ion) donor in anabolic processes, that is, biosyntheses. For example, NADPH is the reducing co-factor used by fatty acid synthetase in lipid biosynthesis and by desmolases and hydroxylases in steroid biosynthesis.

Nicotinamide Coenzymes in Skin are Depleted with age; Niacinamide can Help Normalize this Imbalance

NADH and NADPH can, thus, be viewed as fundamental energy »currency« units within cells, driving the metabolism of cells involved in both catabolic and anabolic processes. There is an increasing pool of evidence for a decline in systemic and intracellular concentrations of these two coenzymes with age in human and animal models [2,3,4] and recent new data appear to confirm this. Oblong et al. [5] established human dermal fibroblast cell lines from a 7-year old and a 72-year old and used these to measure endogenous NADPH/$NADP^+$ ratios and total NADPH + $NADP^+$ levels. It was found that fibroblasts from the aged donor contained decreased NADP redox ratios and total NADPH + $NADP^+$ levels relative to those from the young donor (51% and 28% respectively). It does appear, therefore, that there is a reduction in nicotinamide coenzymes associated with senescence.

Importantly, Oblong et al. [5] also found that supplementation of human dermal fibroblast cultures derived from elderly donors with ^{14}C-niacinamide and ^{14}C-nicotinic acid (niacinamide precursor) increased intracellular concentrations of NADPH. It appears that a localized supply of niacinamide, therefore, can be utilized by aged cutaneous cells to restore intracellular nicotinamide coenzyme homeostasis.

It is worth noting, however, that despite the efficacy noted above, nicotinic acid (niacin) produces a well-documented cutaneous vasodilatation (»flushing«) when applied topically. Niacin, therefore, presents a challenge to cosmetic applications; there are no such issues with the use of niacinamide.

Aged Fibroblasts Secrete Less Collagen than Young Cells; Niacinamide can Stimulate New Collagen Synthesis

Oblong et al. [5] used ^{14}C-proline to monitor incorporation of label into collagen protein secreted from cultured human dermal fibroblasts taken from a young (7-year old) and aged donor (72-year old). ^{14}C-hydroxyproline (as a marker of newly-synthesized secreted collagen) and ^{14}C-proline (as a marker of total protein) were extracted, separated and quantified using HPLC equipped with a radiometric detector. Results indicated firstly that dermal fibroblasts from an aged donor secreted significantly ($p<0.05$) less collagen than those from a young donor and, furthermore, that NADPH / NADP redox ratios were also lower ($p<0.05$) in fibroblasts from the aged donor (results were normalized to the cell number from the respective culture well). Secondly, it was found that supplementation of the aged cell culture with niacinamide produced significant increases in total collagen secreted (by 54%), total protein secreted (by 41%) and also in the number of cells (by 20%), relative to a vehicle control. Importantly, there was also a significant 35% increase in the collagen / total protein ratio (relative to a vehicle control), indicating some specificity for collagen biosynthesis and secretion. These data suggest, therefore, that treatment with niacinamide would have a positive impact on the dermal compartment, both in terms of its connective tissue and gel matrix components. These effects would be of particular significance in aged and photodamaged skin.

Niacinamide Up-regulates Epidermal Ceramide Synthesis with Concurrent Epidermal Barrier Benefits

Ceramides are now known to play a central role in the structural and functional integrity of the stratum corneum barrier function. A decrease in ceramide fraction has been reported in aged and atopic skin [6]. Tanno et al. showed that in cultured human epidermal keratinocytes, niacinamide could induce up to a 5-fold up-regulation in ceramide synthesis ($p<0.05$) in a dose-dependent fashion [7]. Further work by the same group [8] showed up-regulation of other sphingolipid fractions (glucosylceramide and sphingomyelin) as well as free fatty acid and cholesterol synthesis (by 2.3 and 1.5-fold, respectively). The workers proposed a mechanism for these observations based on increased levels of intra-cellular acetyl-CoA (the precursor common to epidermal lipid synthesis) and increased expression of serine-palmitoyltransferase. Tanno et al. [8] also showed that these *in-vitro* results had clinical significance *in-vivo*. Topical application of a vehicle containing 2% niacinamide

to dry lower legs over 4 weeks induced a measurable significant increase ($p<0.05$) in recovered stratum corneum ceramide and free fatty acid lipid fractions, vs. a vehicle control. This was accompanied by a significant reduction ($p<0.05$) in TEWL vs. vehicle control (–27%). Similar significant reductions in TEWL vs. a vehicle control were noted also by Ertel et al. [9] after use of a moisturizing vehicle containing 2% niacinamide. Furthermore, they also noted that this positive barrier effect was accompanied by an increase in stratum corneum turnover rate (as measured by dansyl chloride assay). Finally, Draelos et al. [10] treated 48 female subjects with stage I/II rosacea with a moisturizing vehicle containing 2% niacinamide for 4 weeks and demonstrated a significant improvement in global condition (assessed) in 96% of the subjects at week 4. They demonstrated, once again, that this clinical benefit was accompanied by a significant improvement in stratum corneum barrier function.

It appears that topical niacinamide is able to augment the barrier properties of the skin, with accompanying clinically relevant benefits, by up-regulating endogenous biosynthesis of epidermal sphingolipids, in particular, ceramides.

Niacinamide Up-regulates Biosynthesis of Markers of Keratinocyte Differentiation

Oblong et al. [5] cultured normal human epidermal keratinocytes to near-confluency and then supplemented the medium with niacinamide. Following a 24h incubation, cells were counted, harvested and prepared for assay for involucrin (by ELISA) and filaggrin (by an immunoblot procedure). Results showed firstly a significant increase ($p<0.05$) in the number of niacinamide-treated NHEK relative to a vehicle control. Secondly, niacinamide-treated NHEK showed an up-regulation of both involucrin and filaggrin biosynthesis vs. that induced by a vehicle control (by 45% and 100% respectively). These two proteins are both critical to the differentiation process and the formation of fully integral keratinized corneocytes; filaggrin plays a vital role in aggregation and alignment of keratin tonofilaments in granular cells and involucrin is an essential precursor in the formation of the insoluble cornified envelope surrounding terminal keratinocytes. In other words, niacinamide has been shown to both stimulate basal epidermal keratinocytes and to up-regulate biosynthesis of epidermal intermediates critical to the formation of a fully functioning stratum corneum. This may be due to increased intracellular levels of reduced nicotinamide coenzymes initiated by topical niacinamide. These effects would be expected to have a significant positive impact on ageing epidermal tissue *in-vivo*.

Niacinamide Helps Prevent UV-Induced Deleterious Molecular and Immunological Events

Shen et al. [11] demonstrated the ability of niacinamide to protect cultured normal human keratinocytes against reactive oxygen species induced by UVC irradiation or exposure to hydrogen peroxide. They observed significant ($p<0.05$) dose-dependent attenuation of apoptotic morphological changes, a decrease in p53 induction and a reduction in DNA ladders for niacinamide-treated cells vs. those treated with a vehicle control. These data are consistent with work in animal models [12] demonstrating clearly the ability of niacinamide to significantly reduce both induction of photocarcinogenesis and photoimmunesuppression. The mechanism by which niacinamide exerts these effects is not yet clear.

Niacinamide Inhibits Transfer of Melanosomes from Melanocytes to Keratinocytes

Boissy et al. [13] used co-cultures of human melanocytes and keratinocytes to investigate the ability of niacinamide to reduce pigmentation in human skin. Use of immunolinked dyes specific for each cell type enabled separate counts of keratinocytes, melanocytes and keratinocytes containing transferred melanosomes to be performed. The workers found significant inhibition ($p<0.05$) of melanosome transfer to keratinocytes from melanocytes incubated in the presence of niacinamide (by 25-45%). It was also confirmed that niacinamide had no inhibitory effect on melanocyte tyrosinase activity. These data suggest that treatment of human skin *in-vivo* with topical niacinamide would lead to a reduction in pigmentation with time via this novel, elegant mechanism.

Niacinamide Reduces Human Skin Hyperpigmentation

Hakozaki et al. [14] performed two studies demonstrating the effect of niacinamide on skin hyperpigmentation *in-vivo*. In the first, 18 female Japanese subjects with hyperpigmented facial spots were treated for 8 weeks with a vehicle containing 5% niacinamide vs. a vehicle control in a split-face design. Pigmented spots were qualified and quantified via algorithmic analysis of high-resolution digital images and subjective grading of images. Results of image analysis showed that 5% niacinamide had induced a significant ($p<0.05$) reduction in spot area at the 4 and 8 week time-points (vs. vehicle control), accompanied by a significant reduction ($p<0.05$) in graded visible spot pigmentation at 8 weeks (vs. vehicle control). In the second study, 120 female Japanese subjects with facial tanning were assigned to 2 of 3 treatments (SPF 15 sunscreen moisturizer, 2% niacinamide in SPF 15 sunscreen moisturizer and a vehicle control). Subjects applied treatments split-face for 8 weeks. In this study overall skin lightness was assessed by analysis of digital images and by subjective grading. Results of image analysis showed a significant ($p<0.05$) increase in skin lightness vs. the sunscreen moisturizer and vehicle control at the 4 and 6 week time-points, accompanied by a significant ($p<0.05$) increase in graded visible skin lightness vs. vehicle control at 4 weeks. These *in-vivo* data appear to confirm, therefore, that the inhibitory role of niacinamide in melanosome transfer noted *in-vitro* [13], does indeed translate to a significant effect on hyperpigmentation *in-vivo*.

Regulation of Sebaceous Lipid and Acne by Niacinamide

Topical niacinamide in the form of a commercial 4% gel (Papulex®) has been shown to provide potent anti-inflammatory activity in the treatment of acne vulgaris. Shalita et al. [15] found that after 8 weeks of usage, 82% of subjects with inflammatory acne showed an improvement in global evaluation, with a significant reduction in papules / pustules (–60%) and acne severity (–52%). Indeed, many practitioners use the treatment citing a combination of efficacy and lack of bacterial-resistance. Shalita et al. [15] and others postulate that niacinamide

may act via its apparent antihistaminic effect, activity as an electron scavenger, or its inhibition of 3'-5' cyclic-AMP phosphodiesterase activity.
Recent data, however, appear to demonstrate an altogether more fundamental role for topical niacinamide in acne treatment. Biedermann et al. [16] used viable human facial biopsies (from face-lift surgery) to measure the effect of niacinamide on sebaceous lipogenesis. Cultured biopsies were treated with niacinamide or trans-retinoic acid (tRA) for 4 days, after which they were incubated with ^{14}C-acetate. Lipid components were subsequently isolated, fractionated and identified using analytical TLC and radiometry. Niacinamide produced significant dose-dependent reductions in total sebaceous lipogenesis (–42% at 25 mM [$p<0.01$]). Furthermore, the reduction induced by 25 mM niacinamide was equivalent to that produced by 1 µM tRA (–32% [$p=0.01$]). When discrete lipid classes were identified and quantified, it was found that niacinamide had produced marked reductions in both triglyceride and fatty acid synthesis vs. the control (–52% and –46% respectively for 25 mM niacinamide [$p<0.05$]). It is now known that triglycerides represent by far the largest proportion of sebaceous gland lipid (50-60%); the observed effect of niacinamide on total lipogenesis is, therefore, probably attributable to triglyceride reduction.
This has important implications for acne pathogenesis. It is accepted that acne is a disease involving the pilosebaceous duct and *Propionibacterium acnes*. Despite the on-going debate as to the exact interplay of these factors, it is without doubt that a significant reduction both in total sebaceous lipid bulk and in the triglyceride fraction would be expected to impact positively acne-form skin.

Niacinamide Exerts Multiple Benefits on the Appearance of Ageing / Photodamaged Skin, *In-vivo*

Bissett et al. [17] studied the effects of topical niacinamide in ageing human facial skin in two double-blinded clinical studies. In the first, 40 female subjects aged 35 - 60 applied a vehicle and vehicle containing 5% niacinamide (randomized split-face) for 12 weeks. High-resolution digital images were taken at baseline, 4, 8 and 12 weeks and texture and hyperpigmentation were evaluated by judges (comparing blind-coded image pairs, baseline vs. another treatment time-point). Judges were able to perceive a significant improvement in skin texture appearance at 4 weeks ($p<0.1$) and 12 weeks ($p<0.05$) and a significant improvement in hyperpigmented spot appearance by 8 weeks ($p<0.05$) **(Fig. 3)**.
In a second study, female subjects aged 35-60 applied blind-coded products (vehicle control and vehicle containing 5% niacinamide; n=88) split-face for 8 weeks. Skin texture appearance was assessed as above. The niacinamide-containing treatment provided a significant improvement in skin texture appearance relative to the vehicle control at the 8 week time-point, confirming the results of the first study.
This effect on skin surface texture is consistent with that noted in a 10-week clinical study where Matts & Solechnick [18] used multiple-angle reflectance spectrophotometry to measure the diffuse component of skin reflection. They noted a significant increase in the diffuse component of 5% niacinamide-treated dorsal hand skin vs. vehicle control after 10 weeks of treatment ($p<0.05$), consistent with significant blind self-rated preferences for texture appearance over vehicle control ($p<0.05$). This change was consistent with a shift in texture distribution towards the finer, anisotropic features characteristic of younger skin.

Niacinamide is Delivered Effectively from a Range of Vehicles and Demonstrates Excellent Skin Compatibility

Franz [19] determined niacinamide absorption *in-vitro* through full-thickness abdominal skin mounted in flow-through diffusion cells. Franz used acetone as a carrier and found 28.8% of the starting dose in the receptor medium at 24 h. Delivery of niacinamide (2-20%) from a range of cosmetic formulae (including moisturizers, foundations and lipsticks) was studied using a modified *in-vitro* Franz flow-through cell technique [20]. For formulae containing 2% niacinamide, approximately 10% of the starting dose was detected in the receptor medium at 48 h. Importantly, these studies highlighted the apparent independence of niacinamide penetration rate from diverse vehicle matrices.
The Cosmetic Ingredient Review Expert Panel report for niacinamide [21] details a very wide range of cutaneous tolerance studies confirming the excellent profile of niacinamide as a cosmetic skin care ingredient.

Conclusion

Niacinamide, therefore, has been shown to be a cosmetic ingredient with an extraordi-

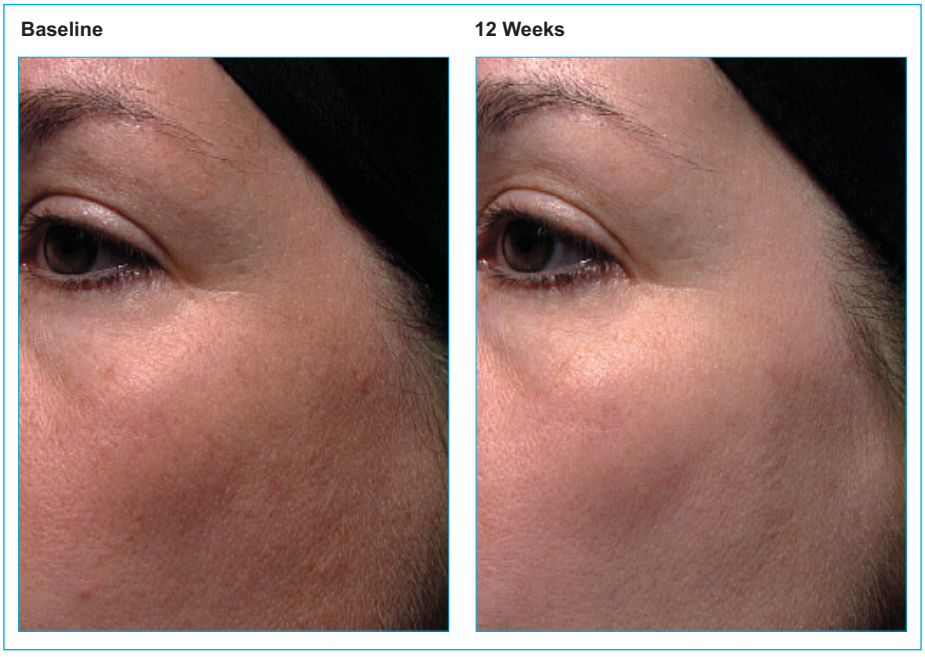

Fig. 3: Same subject at baseline and after 12 weeks of topical treatment with 5% niacinamide

nary breadth and history of cutaneous benefits. It is thought that its fundamental role as a precursor of reduced nicotinamide coenzymes such as NADH and NADPH is pivotal to its observed effects. It displays distinct advantages over other ingredients with similar benefits, such as retinol, in that it is well tolerated, and is not subject to oxidation or photolysis. In short, the multiplicity of effects and formulation benefits seen with niacinamide make it an ideal choice for a variety of cosmetic products targeting young and old skin alike.

References

[1] Rajakumar, K., Pellagra in the United States: a historical perspective, *Southern Med. J.*, **93** (3), (2000) 272-277.

[2] Jongkind, J.F., Verkerk, A., and Poot, M., Glucose flux through the hexose monophosphate shunt and NADP(H) levels during *in vitro* ageing of human skin fibroblasts, *Gerontology* **33** (5) (1987) 281-286.

[3] Gilchrest, B.A., and Yaar, M., Ageing and photoageing of the skin: Observations at the cellular and molecular level, *Br. J. Dermatol.*, **127 Suppl 41** (1992) 25-30.

[4] Seitz, H.K., Xu, Y., Simanowski, U.A., and Osswald, B., Effect of age and gender on *in vivo* elimination, hepatic dehydrogenase activity and NAD^+ availability in F344 rats, *Res. Exp. Med.*, **192** (3) (1992) 205-212.

[5] Oblong, J.E., Bissett, D.L., Ritter, J.L., Kurtz, K.K., and Schnicker, M.S., »Niacinamide stimulates collagen synthesis from human dermal fibroblasts and differentiation marker in normal human epidermal keratinocytes: Potential of niacinamide to normalize aged skin cells to correct homeostatic balance, 59th Annual Meeting American Academy of Dermatology, Washington, 2001.

[6] Imokawa, G., Abe, A., and Jin, K., Decreased level of ceramides in stratum corneum of atopic dermatitis: a factor in atopic dry skin? *J. Invest. Dermatol.*, **96** (1991) 523-526.

[7] Tanno, O., Yukiko, O., Kitamura, N., and Inoue, S., Effects of niacinamide on ceramide biosynthesis and differentiation of cultured human keratinocytes, 3rd ASCS Conference, Taipei, Taiwan, 1997.

[8] Tanno, O., Ota, Y., Kitamura, N., Katsube, T. and Inoue, S., Nicotinamide increases biosynthesis of ceramides as well as other stratum corneum lipids to improve the epidermal permeability barrier, *Br. J. Dermatol.*, **143** (3) (2000) 525-531.

[9] Ertel, K.D., Berge, C.A., Mercurio, M.G., Fowler, T.J., and Amburgey, M.S., New facial moisturizer technology increases exfoliation without compromising barrier function, 58th Annual Meeting of the American Academy of Dermatology, San Francisco, 2000.

[10] Draelos, Z.D., Ertel, E., Berge, C., and Amburgey, M.S., A facial moisturizing product as an adjunct in the treatment of rosacea, 59th Annual Meeting American Academy of Dermatology, Washington, 2001.

[11] Shen, S.C., Yoshii, T., Chen, Y.C., Tsai, T.H., Hu, C.H., and Lee, W.R., Niacinamide reduces DNA damage caused by reactive oxygen species, 60th Annual Meeting American Academy of Dermatology, New Orleans, 2002.

[12] Gensler, H.L., Prevention of photoimmunesuppression and photocarcinogenesis by topical niacinamide, *Nutr. Cancer*, **29** (2) (1997) 157-162.

[13] Boissy, R.E., Minwalla, L., Bissett, D.L., Zhuang, J.C., and Chhoa, M., Niacinamide inhibits transfer of melanosomes from melanocytes to keratinocytes, 59th Annual Meeting American Academy of Dermatology, Washington, 2001.

[14] Hakozaki, T., Matsubara, A., Miyamoto, K., Hillebrand, G.G., and Bissett, D.L., Topical niacinamide reduces human skin hyperpigmentation, 60th Annual Meeting American Academy of Dermatology, New Orleans, 2002.

[15] Shalita, A.R., Smith, J.G., Parish, L.C., Sofman, M.S., and Chalker, D.K., Topical nicotinamide compared with clindamycin gel in the treatment of inflammatory acne vulgaris, *Int. J. Dermatol.*, **34** (6) (1995) 434-437.

[16] Biedermann, K., Lammers, K., Mrowczynski, E., Coombs, M., Lepp, C., El-Nokaly, M., and Burton, E., Regulation of sebum production by niacinamide, 60th Annual Meeting American Academy of Dermatology, New Orleans, 2002.

[17] Bissett, D.L., Oblong, J.E., Saud, A., and Levine, M., Topical niacinamide provides improvements in ageing human facial skin, 60th Annual Meeting American Academy of Dermatology, New Orleans, 2002.

[18] Matts, P.J., and Solechnick, N.D., Predicting visual perception of human skin surface texture using multiple-angle reflectance spectrophotometry, 59th Annual Meeting American Academy of Dermatology, Washington, 2001.

[19] Franz, T.J., Percutaneous absorption: On the relevance of *in vitro* data, *J. Invest. Dermatol.*, **64** (1975) 190-195.

[20] P&G internal data, cited in »Safety assessment of niacinamide and niacin; tentative report of the Cosmetic Ingredient Review Expert Panel«, September 17, 2001.

[21] »Safety assessment of niacinamide and niacin; tentative report of the Cosmetic Ingredient Review Expert Panel«, September 17, 2001