**Introduction**

Nicotinamide is widely used in cosmetics and skin care products. The vitamin has been shown to be able to reduce skin pigmentation and to increase lipid biosynthesis in the stratum corneum. Hakozaki et al., showed that 1 mM nicotinamide is able to suppress melanosome transfer from melanocytes to keratinocytes in co-cultures in vitro (35.3-67.6 % reduction). Tanno et al., demonstrated in vitro that lipid biosynthesis was increased by concentrations of nicotinamide in the range 1-30 µM.

**Aim of the work**

The aim of this work was to determine the concentration of nicotinamide in the dermal and epidermal layers of the skin after topical application of a gel.

**Objectives**

To compare the accumulation in rabbit and human skin.
To study the influence of nicotinamide concentration in the gel on human skin accumulation.
To study the effect of application time.

**Experimental Methods**

**Permeation experiments:**
- Franz type diffusion cells (area 0.6 cm²).
- Barrier: rabbit ear skin or human skin.
- Donor (0.5 ml): Nicotinamide gel (pH 6.0).
- Receptor solution: saline (4 ml).

**Extraction:**
- Epidermis-dermis heat separation.
- Nicotinamide extraction with mobile phase.

**Nicotinamide HPLC analysis:**
- µBondapack Waters C18 column.
- UV detection @ 261 nm.
- Mobile phase: methanol (15.8 % v/v) 0.1 M sodium phosphate containing 0.01 M TBA to pH 5.0 (84.2 % v/v), @ 1.2 ml/min.

**Results & Discussion**

**Validation of Extraction Method**

The recovery from epidermis and dermis was complete, thus the use of an internal standard could be avoided.

<table>
<thead>
<tr>
<th>Added (µg)</th>
<th>Recovered (µg)</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Epidermis</td>
<td>7.94±0.85</td>
<td>8.32±0.80</td>
</tr>
<tr>
<td>Dermis</td>
<td>7.94±0.85</td>
<td>8.04±1.27</td>
</tr>
</tbody>
</table>

**Rabbit vs. Human Skin**

<table>
<thead>
<tr>
<th>Concentration</th>
<th>Epidermis</th>
<th>Dermis</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0 % NAD</td>
<td>0.0 ± 0.0</td>
<td>0.1 ± 0.0</td>
</tr>
<tr>
<td>5.0 % NAD</td>
<td>0.5 ± 0.5</td>
<td>0.5 ± 0.5</td>
</tr>
<tr>
<td>10 % NAD</td>
<td>1.0 ± 1.0</td>
<td>1.0 ± 1.0</td>
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**Effect of Nicotinamide Concentration**

- Rabbit ear skin resulted more permeable to nicotinamide, since the amount accumulated at 20 min (5% gel) was 5-6 times higher than in human skin.
- Nicotinamide accumulation in the epidermis was insensitive to application time.
- The amount of nicotinamide recovered in the receptor compartment was lower than 1 µg/cm² after one hour of 3.5 % gel application.
- Nicotinamide molar concentration in the skin was calculated by dividing the amount recovered by the weight of tissue, assuming its density was one. The results for human epidermis are in the range 4 mM (5 % gel applied for 20 min) to 10 mM (10% gel applied for 20 min), well above the values reported as sufficient to suppress melanocyte transfer and to stimulate lipid biosynthesis in vitro.

**Discussion**

- Nicotinamide accumulated in the skin already after 20 min of gel application; the higher concentration was found in the epidermis.
- Nicotinamide concentration in the donor gel modified to some extent the amount accumulated in the epidermis and dermis.
- Increasing the application time from 20 to 60 min had no effect on epidermis concentration, albeit increased dermis accumulation (p<0.05).
- Overall, these results indicate that nicotinamide accumulates in the skin after topical application of a gel for 20-60 min, without producing systemic absorption (despite infinite dose conditions).

**Conclusions**

2. O. Tanno, Y. Ota, N. Kitamura, T. Katsube, S. Inoue, Nicotinamide increases biosynthesis of ceramides as well as other stratum corneum lipids to improve the epidermal permeability barrier, Br. J. Dermatol., 143, 524-531, 2000

**References**