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Effect of lipid-containing, positively charged nanoemulsions on skin hydration, elasticity and erythema—An in vivo study

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Abstract

Dry skin and other skin disorders such as atopic dermatitis are characterized by impaired stratum corneum (SC) barrier function and by an increase in transepidermal water loss (TEWL) leading to a decrease in skin hydration. The possibility that dermatological and cosmetic products containing SC lipids could play a part in the restoration of disturbed skin barrier function is of great interest in the field of dermatology and cosmetics. The aim of the present study was to evaluate the effect of positively charged oil/water nanoemulsions (PN) containing ceramide 3B and naturally found SC lipids (PNSC) such as ceramide 3, cholesterol, and palmitic acid on skin hydration, elasticity, and erythema. Creams of PNSC were compared to PN creams, to creams with negatively charged o/w nanoemulsion and SC lipids (NNSC) and to Physiogel® cream, a SC lipid containing formulation, which is already on the market. The formulations (PN, PNSC, and NNSC) were prepared by high-pressure homogenization. After adding Carbopol 940 as thickener, particle size and stability of the creams were not significantly changed compared to the nanoemulsions. The studies were carried out on three groups, each with 14 healthy female test subjects between 25 and 50 years of age, using Corneometer® 825, Cutometer® SEM 575 and Mexameter® 18 for measurements of skin hydration, elasticity, and erythema of the skin, respectively. The creams were applied regularly and well tolerated throughout the study. All formulations increased skin hydration and elasticity. There was no significant difference between PNSC and Physiogel®. However, PNSC was significantly more effective in increasing skin hydration and elasticity than PN and NNSC indicating that phytosphingosine inducing the positive charge, SC lipids and ceramide 3B are crucial for the enhanced effect on skin hydration and viscoelasticity.

Keywords: Ceramide; Phytosphingosine; Positively charged nanoemulsion; Skin elasticity; Skin hydration; Skin measurement in vivo

1. Introduction

Stratum corneum (SC) lipids such as cholesterol (Feingold et al., 1990), free fatty acids (Mao-Qiang et al., 1993a,b) and especially ceramides (Holleran et al., 1991) have been recognized as playing a major role in the skin barrier homeostasis. It is believed that one cause of dry skin is the reduction in the amount of ceramides within the intercellular lipid lamellae of the stratum corneum (Gaetani et al., 2003; Rawlings, 2003). Thus, it is desirable to be able to successfully replace these depleted SC lipids via the topical route. Ceramides are extremely insoluble compounds, a property directly linked to their intrinsic functionality, i.e. the formation of a water-impermeable barrier. In order to provide this function, ceramides must be able to penetrate the stratum corneum in order to reach the lipid lamellae. A potential problem with the topical application of skin products is finding a suitable dosage form to deliver the active ingredients such as ceramides in sufficient amounts to the active site. The complete dissolution of drug in the formulation and skin permeability enhancement are important considerations for the effective delivery of ceramides via the topical route (De Paepe et al., 2000, 2002).

Oil/water (o/w) nanoemulsions are promising colloidal drug carrier systems for diverse therapeutic applications. Successfully developed, intravenous, oral and ocular delivery systems showed reduced side effects of various potent drugs and prolonged pharmacological effect of drugs in these nanoemulsion formulations (Gershanik and Benita, 1996; Klang and Benita, 1998; Abdulrazik et al., 2001). Since a positively charged delivery system might enhance the permeability of a poorly soluble drug (Piemi et al., 1999), due to the strong interaction between epithelial membranes with positively charged

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solutes (Rojanasakul et al., 1992), the development of positively charged nanoemulsion for the skin penetration enhancement of low soluble biological active compounds such as ceramides is interesting.

Recently, a 6-months stable, positively charged o/w nanoemulsion incorporating ceramides was successfully developed in our labs (Yilmaz and Borchert, 2005). The droplets of the nanoemulsion exhibited their positive charge upon the physiological compound phytosphingosine (PS). PS, a free amphiphilic sphingoid base, is naturally found in the human body and is present at high levels in the SC. Topical application of PS and its derivatives has been shown to increase stratum corneum ceramide levels and barrier function (Rawlings, 2003). PS is also considered to be part of the skin's natural defence system (Wolf et al., 1997; Lambers and Streekstra, 1998; Park et al., 2002). Because of these properties, PS is an attractive candidate for topical use.

This investigation was focused on two poorly soluble ceramides, ceramide 3 (C3), the most abundant ceramide in healthy human skin, and the non-skin identical ceramide 3B (C3B). By topical application, both ceramides were able to improve skin barrier recovery indicated by increasing of skin hydration (Lambers and Roehl, 1999; De Paepe et al., 2002).

The aim of this work was to study the effect of a positively charged nanoemulsion cream containing phytosphingosine, incorporating ceramides and SC lipids (PNSC cream), on skin properties such as skin hydration, elasticity and skin erythema in healthy female volunteers. In this respect, PNSC cream was compared to (1) a positively charged nanoemulsion cream without SC lipids in order to evaluate the importance of the SC lipids on skin hydration and elasticity, (2) a negatively charged nanoemulsion cream in order to investigate the influence of the phytosphingosine and surface charge on the penetration enhancement, and (3) Physiogel® cream, a SC lipid containing formulation already on the market, in order to classify the effect of PNSC cream. Since ceramides may increase the water content of the skin (Lambers and Roehl, 1999), the skin hydration and elasticity were chosen as parameters to estimate the extent of penetration of ceramides into the skin. Erythema measurements were used as an indication of the skin tolerance to the formulations. Additionally, an ex vivo spreadability study was carried out in order to gain insight regarding the enhanced topical penetration effect of the o/w nanoemulsion as a function of surface charge.

2. Experimental methods

2.1. Materials

Ceramide 3 (C3), ceramide 3B (C3B) and phytosphingosine (PS) were kindly provided by Degussa, Essen, Germany. Lipoid E-80® (LE80; a mixture of phospholipids ex ovo with at least 80% phosphatidylcholine) was obtained from Lipoid KG, Ludwigshafen, Germany. The cosmetic oil, Eutanol G (octyldodecanol), and the preservative potassium sorbate were purchased from Caelo, Caesar & Loretz GmbH, Hilden, Germany and conformed with European Pharmacopoeia specifications. The antioxidant D,L-α-tocopherol and myristic acid were supplied from Synopharm, Barsbüttel, Germany and from Carl Roth GmbH & Co. KG, Karlsruhe, Germany. Tween 80® (T80) was supplied from Uniqema, Everberg, Belgium. Physiogel was kindly provided by Stiefel Laboratorium GmbH, Offenbach. All used ingredients were of pharmaceutical grade.

Table 1
Composition of the nanoemulsions used in the studies

Compounds	Nanoemulsion composition (%, w/w)		
	Positively charged		Negatively charged
	PNSC	PN	NNSC
Oil phase			
Eutanol G	20	20.8	20
Lipoid E-80®	2	2	2
Myristic acid	–	–	0.6
Phytosphingosine	0.6	0.6	–
Ceramide 3B	0.2	–	0.2
Ceramide 3	0.2	–	0.2
Palmitic acid	0.2	–	0.2
Cholesterol	0.2	–	0.2
Vitamin E	0.03	0.03	0.03
Ethanol	–	–	2
Water phase			
Tween 80	2	2	2
Glycerol	2.5	2.5	2.5
Potassium sorbate	0.1	0.1	0.1
Water to	100	100	100

PNSC, positively charged nanoemulsion with stratum corneum lipids; PN, positively charged nanoemulsion without stratum corneum lipids; NNSC, negatively charged nanoemulsion with stratum corneum lipids.

2.2. Production of nanoemulsions and creams

Aqueous and oil phases were prepared separately (Table 1). The aqueous phase, containing T80, glycerol, potassium sorbate, and bidistilled water, was heated to 50 °C under slight mixing. PS, for positively charged nanoemulsion, C3 and C3B were dissolved in Eutanol G above 100 °C and then cooled down to 75 °C. Then, LE80, cholesterol, palmitic acid, α-tocopherol and myristic acid, for negatively charged nanoemulsion, were dissolved in the oil phase and cooled down to 50 °C. Now, ethanol was added in the oil phase of negatively charged nanoemulsions to keep the lipids dissolved. The two phases were merged and prehomogenized with an Ultra-Turrax (Janke and Kunkel GmbH, Staufen, Germany) at 8000 rpm for three minutes and further homogenized with a high pressure homogenizer (Micron Lab 40, APV Systems, Germany) at 50 °C, 500 bar and eight homogenization cycles (Yilmaz and Borchert, 2005). After immediate cooling to room temperature and filtering through a membrane filter (polytetra-fluorethylene filter, Sartorius AG Germany, pore size 1.2 μm), 0.3–0.4% Carbopol 940 was added to the nanoemulsions by stirring with a Unguator® E 100 (GAKO Konietzko GmbH, Bamberg, Germany) for 30–60 min to obtain creams which were stored at a temperature of 2–8 °C.

2.3. Viscosity

The viscosity of the formulations was measured using a rotational viscometer (Haake RH101, Thermo Haake Gmbh, 76227 Karlsruhe, Germany) with a parallel plate and cone (20 mm in diameter and 4° angle) at a constant shear rate and temperature of 20 min^{-1} and 25 °C, respectively.

2.4. Particle size and zeta potential

For measurement of the mean droplet size, polydispersity index (width of the size distribution) and the surface charge by measuring zeta potential (ZP), a Malvern Zetasizer 4 (Malvern Instruments, Worcestershire, UK) was used. Prior to particle size and size distribution analysis, the formulations were diluted with double-distilled water to weak opalescence.

2.5. Skin humidity (skin hydration)

The measurement of the skin humidity was carried out by a Corneometer® 825, which was mounted on a Multi Probe Adapter® MPA5 (Courage and Khazaka, Electronic GmbH, Cologne, Germany). The measurement was performed by the capacitance method. This makes use of the relatively high dielectric constant of water ($\varepsilon_r = 81\ C^2/Nm^2$) compared to the ones of other substances in the skin ($\varepsilon_r < 7\ C^2/Nm^2$). The front surface of the measurement sensor contains the measuring condenser. If the measurement head is pressed onto the skin for one second, the horny layer comes into the scatter range of the condenser field. Depending on the water content, different capacitance changes can be measured, which are converted into a digital measured value (arbitrary units) being proportional to the skin humidity. Because of the short measurement time, errors due to skin deformations or evaporative build-up were excluded. Five measurements were performed in each testing area at different points of the volar forearms.

2.6. Skin elasticity

The measurement of the elastic property of the skin was conducted by a Cutometer® SEM 575 (Courage and Khazaka Electronic GmbH, Cologne, Germany) using a non-invasive suction- and elongation-method. During the measurement, the skin surface was pulled into the aperture (2 mm internal diameter) of a special probe by an applied vacuum with a pressure of 450 mbar. The depth of evagination of the skin into the probe was sensed optically, friction-free and without mechanical action. Each measurement consisted of five suction cycles (3 s of suction followed by 3 s of relaxation) and was performed three times in each testing area on different points of the volar forearms. The relative parameter U_a/U_f (relation between maximum deformation during the first cycle and back formation directly after the first cycle = gross elasticity) was evaluated.

2.7. Skin erythema and skin irritation

Skin irritation was observed visually. Any kind of changes of the colour or the constitution of the skin surface was recorded. The pathological parameter, erythema (=content of haemoglobin), was measured photometrically by a Mexameter® 18 (Courage and Khazaka Electronic GmbH, Cologne, Germany) based on remission principle. The special probe of the Mexameter® 18 emits light of two defined wavelengths. One of these wavelengths (568 nm) corresponds to the spectral absorption peak of haemoglobin. The other wavelength (660 nm) was chosen to avoid other colour influences (e.g., bilirubin). A receiver measures the light reflected by the skin. The erythema index (EI) was calculated by the instrument according to the formula: EI = 1000 log (red-remittance/green-remittance). Each measurement was performed in quintuplicate on different points of the volar forearms.

2.8. Spreadability

The higher the extent of spreading of a liquid on a solid is, the higher is the attraction or adhesion between solid and liquid (Young–Dupre equation). Therefore, the spreadability could be used as a parameter in order to clarify the extent of interaction between the liquid o/w nanoemulsion, and the solid skin surface, which would affect the penetration behaviour of the nanoemulsions. The Young and Dupre equations, which are relationships of fundamental importance in interfacial thermodynamics, can be transformed into the following equation:

$$S = \gamma_{LV}(\cos\theta - 1)$$

where S is the spreading coefficient, γ_{LV} is the surface tension of the formulations and θ is the contact angle between the liquid formulation and the skin. It can be deduced that any negative value of S approaching zero will indicate enhanced spreading on the skin, since at $S = 0$ complete wetting occurs.

The surface tension of the positively and negatively charged o/w nanoemulsions (γ_{LV}) was measured by the Wilhelmy plate method using tensiometer K100 (Krüss GmbH, Germany). All surface tension results were the mean values of six-fold determinations.

The contact angle (θ) measurement was investigated by the drop shape analysis system DSA10D04 (Krüss GmbH, Hamburg, Germany) using the static, sessile, optical contact angle drop method. One droplet of the formulations (5 μl) was placed on the with 2% Tween 80 solution previously treated skin (freshly excised human abdomen skin after plastic surgery) with a cannula. The image of the droplet was recorded by a camera and the contact angle between the baseline of the droplet and the tangent at the drop boundary was measured by the drop shape analysis system using the ellipse-fitting method. The plateau value of the contact angle was reached after 5 min. Each measurement was performed in sextuple.

Table 2
Study design

Study	PNSC creams	Physiogel® creams	PN creams	NNSC creams
1	√	√		
2	√		√	
3	√			√

PNSC creams, creams of positively charged nanoemulsion with stratum corneum lipids; PN creams, creams of positively charged nanoemulsion without stratum corneum lipids; NNSC creams, creams of negatively charged nanoemulsion with stratum corneum lipids.

2.9. Study design

The three studies (Table 2) were designed as one-sided blind, placebo controlled studies with intraindividual comparison of two formulations and two untreated test areas on the volar forearms. For each study, 14 healthy caucasian females (age 25–50) with healthy skin who had previously given oral consent applied two test formulations (Table 1, Physiogel® cream included) twice daily over a period of 28 days guided by an instruction protocol. The skin properties were measured approximately at the same time on day 0 (basic value, prior to application of the creams), 14, 28, 29 (1st day after last application) and 31 (3rd day after last application). On days 14 and 28, the creams were not applied prior to the measurements. The results were calculated with the following equation in order to be able to make valuable statements on the effects of the formulations:

$$\text{changes in skin property (\%)} = \frac{\bar{Q}_{tv}}{\bar{Q}_{0v} - 1} \times 100$$

where $\bar{Q}_{tv}$ is the mean of the quotients of the measured values of treated and untreated skin area after application time t of all volunteers v, and $\bar{Q}_{0v}$ is the mean of the quotients of the measured values of treated and untreated skin area before application time of all volunteers v. The volunteers were accommodated in an air-conditioned room at $21 \pm 1\,^{\circ}C$ and $50 \pm 5\%$ relative humidity. The compliance was verified and controlled by a detailed instruction of the application, by an application protocol for home, in which the volunteers had to record the application time, and by weighing the samples before and after the study.

2.10. Statistical assessment

Statistical differences were determined by the software package STATeasy, Version 2002 using the Wilcoxon test for paired observations with $P < 0.05$ as a minimal level of significance.

3. Results and discussion

3.1. Evaluation of the formulations

The good stability of the positively charged nanoemulsions with and without SC lipids (PN and PNSC, respectively) and the negatively charged nanoemulsions with SC lipids (NNSC) was among other parameters (Yilmaz and Borchert, 2005) due to the presence of the cosurfactants phytosphingosine (PS) and myristic acid, respectively, providing zeta potential values of $+35 \pm 4$ mV for PNSC creams, $+38 \pm 5$ mV for PN creams and -43 ± 5 mV for NNSC creams (Table 3), leading to strong repulsion of the nanodroplets. This repulsion supported the prevention from aggregation, flocculation and coalescence of the nanosized droplets. Even though Carbopol 940 was used as gelling agent, the positively and negatively charged nanoemulsions were stable during the study indicated by no significant change of the mean droplet size and the viscosity.

3.2. Compliance and tolerance

All creams were regularly applied with application rates of more than 97% (Table 4). There were no statistically significant differences in the consumption of the creams for both forearms throughout the three studies. The erythema measurements after 2 and 4 weeks of application revealed no statistically significant differences to the basic values on day 0 and to the untreated control. The formulations did not induce any visual skin irritation. Thus, it could be concluded that all formulations were well tolerated and the compliance was given.

3.3. Study 1

In the first study, PNSC cream was compared to Physiogel® cream because of their similar compositions regarding the content of ceramide and because of its proven positive effect on skin humidity and elasticity (Derma Consult GmbH, 1998). For both formulations, the maximum increase in skin humidity was reached after 2 weeks with $33.3 \pm 5.2\%$ and $32.7 \pm 7.6\%$ for PNSC cream and Physiogel® cream, respectively (Fig. 1). Three days after the last application the increase of skin humidity was $13.6 \pm 4.5\%$ and $14 \pm 5.4\%$ for PNSC cream and Physiogel®

Table 3
Physicochemical characterization of the formulations used in the studies

Formulations	PNSC creams	Physiogel® creams	PN creams	NNSC creams
Type	Nano emulsion	Derma-membrane-structure	Nanoemulsion	Nanoemulsion
Zeta potential (mV)	+35 ± 4	–	+38 ± 5	−43 ± 5
Mean droplet size (nm)	200 ± 38	–	180 ± 34	187 ± 34
Gelling agent	Carbopol 940	i.a. Carbopol 940	Carbopol 940	Carbopol 940
pH	5.0 ± 0.2	–	5.3 ± 0.3	5.0 ± 0.2
Viscosity (mPa s)	17000 ± 2500	22000 ± 2800	17000 ± 1900	19000 ± 2000

PNSC creams, creams of positively charged nanoemulsion with stratum corneum lipids; PN creams, creams of positively charged nanoemulsion without stratum corneum lipids; NNSC creams, creams of negatively charged nanoemulsion with stratum corneum lipids; all values are expressed in mean ± S.D. with $n = 14$.

Table 4
Compliance and skin tolerance

Study	Formulations	Consumption of the creams (g)	Application rate (%)[a]	Skin erythema (%)[b]	
				2 Weeks of application	4 Weeks of application
1	PNSC creams	9.57 ± 4.98	98.2 ± 1.3	0.7 ± 3.9	1.4 ± 4.3
	Physiogel® creams	10.83 ± 4.95		1.3 ± 3.5	0.4 ± 4.8
2	PNSC creams	10.71 ± 1.65	97.2 ± 2.0	0.1 ± 4.2	0.7 ± 3.2
	PN creams	9.10 ± 1.68		0.3 ± 4.5	0.7 ± 4.6
3	PNSC creams	10.70 ± 3.82	97.4 ± 1.5	0.3 ± 2.8	0.5 ± 3.6
	NNSC creams	9.88 ± 4.32		0.2 ± 3.4	0.3 ± 3.0

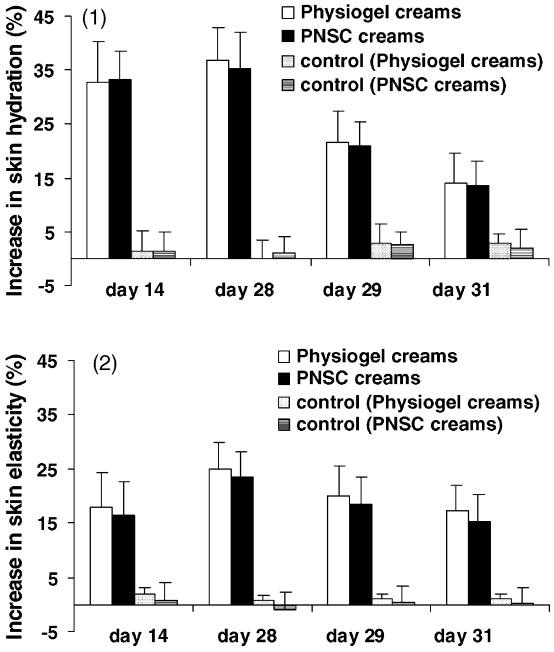
[a] Ratio of real application time to declared application time of 56 times (two times per day for 28 days).
[b] Relation value with regard to day 0 and to the untreated control all values are expressed in mean ± S.D. with $n = 14$.

cream, respectively, showing the sustained effect of the formulations on skin hydration. Regarding the effect on skin elasticity (Fig. 2), there was a similar trend. The maximum increase of 23.6 ± 4.7% for PNSC cream and 25.0 ± 4.9% for Physiogel® cream was reached after 4 weeks, which was at least 2 weeks later than the increase of the skin humidity. The skin humidity was influenced by the water content of the upper layer of the skin, i.e. the stratum corneum. The elasticity was affected by the elastin and collagen fibres, which are located in the deeper layers of the skin, in the dermis. Their influence occurred later after an increase in water content of the skin took place, which then affected the deeper layers (Cua et al., 1990). This might explain the different increase kinetics in skin humidity and elasticity.

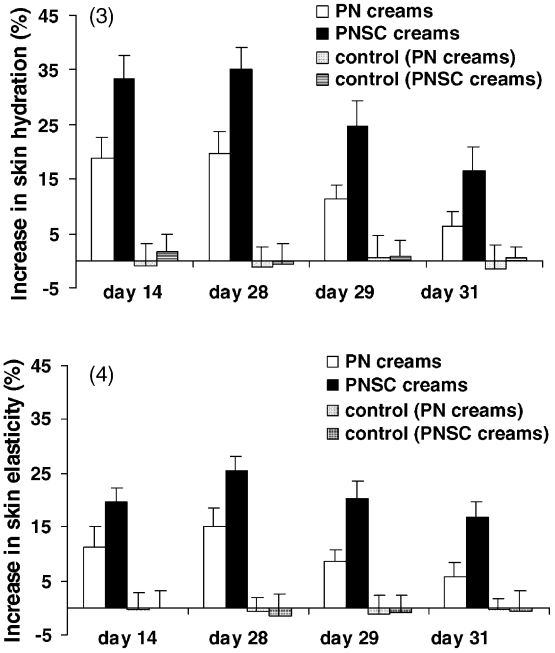
Throughout the study, there were no statistically significant differences in the values of PNSC and Physiogel® cream, demonstrating the comparable efficacy of both formulations on skin humidity and elasticity, deducing the excellent effect of the new developed PNSC cream on skin properties.

3.4. Study 2

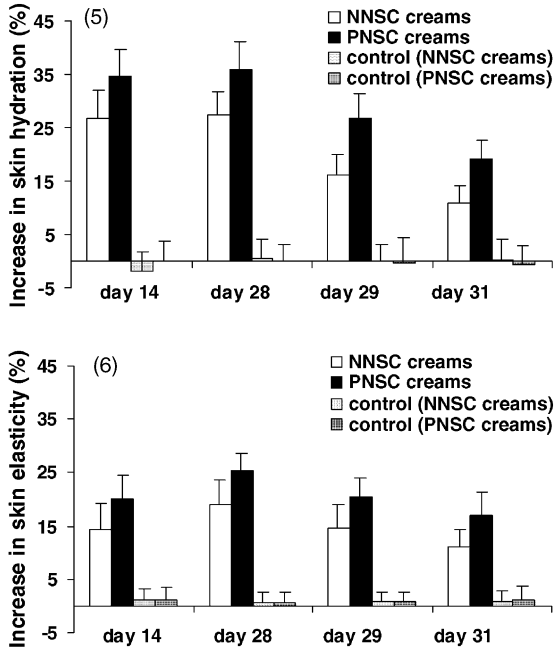
After 4 weeks both formulations showed an increase in skin humidity and elasticity of 33.4 ± 4.3% (PNSC cream), 18.9 ± 3.6% (PN cream) and 19.7 ± 2.7% (PNSC cream), 11.4 ± 3.7% (PN cream), respectively (Figs. 3–4). Additionally, a sustained effect 3 days after the last application of 16.5 ± 4.4% (PNSC cream), 6.3 ± 2.7% (PN cream), and 16.8 ± 3.0% (PNSC cream), 5.9 ± 2.4% (PN cream), similar to study 1, could be observed. The effect of the PN creams were less pronounced than the one of PNSC creams demonstrating the importance of the SC lipids and ceramide 3B on skin properties. All values of PNSC creams were significantly higher than the one of PN creams, indicating the requirement of SC lipids in order to prolong the effect on skin properties and in order to improve the barrier function of the skin by leading to an increase of the skin humidity and thus to an increase in skin elasticity. These results are in conformity with Mao-Qiang et al. (1995), who stated that cholesterol,



Figs. 1–2. Effect of Physiogel® and PNSC creams on skin hydration (top) and skin elasticity (bottom); all results are expressed as values related to the measured values on day 0 and the untreated control (mean ± S.D.; $n = 14$).



Figs. 3–4. Effect of PN and PNSC creams on skin hydration (top) and skin elasticity (bottom); all results are expressed as values related to the measured values on day 0 and the untreated control (mean ± S.D.; $n = 14$).



Figs. 5–6. Effect of NNSC and PNSC cream on skin hydration (top) and skin elasticity (bottom); all results are expressed as values related to the measured values on day 0 and the untreated control (mean ± S.D.; $n = 14$).

ceramides and fatty acids are required for skin homeostasis, but the individual application of these lipids impeded rather than facilitated barrier recovery. Moreover, all three species needed to be applied together for improving the barrier function of the skin; incomplete mixtures delayed skin barrier homeostasis. Further increase in the amount of one lipid led to an accelerated skin barrier repair (Mao-Qiang et al., 1996). Thus, beside the better solubility compared to C3, C3B was combined with C3 in the formulations to improve skin barrier function more effectively.

The placebo PN cream also had a minor effect on skin humidity and elasticity due to the presence of glycerol (Thau, 2002) and the occlusion effect (Berardesca and Maibach, 1988) induced by the oil phase of the creams.

3.5. Study 3

The increase kinetics of skin humidity and elasticity were comparable to the first and second study (Figs. 5–6): there was a maximum in increase of skin humidity after 2 weeks of 34.6 ± 5.0% and 26.9 ± 5.2% for PNSC cream and NNSC cream, respectively, and a maximum in increase of skin elasticity after 4 weeks of 25.4 ± 3.1% and 19.0 ± 4.5% for PNSC cream and PN cream, respectively. Moreover, a prolonged effect of skin humidity and elasticity 3 days after the last application of 19.0 ± 3.7% (PNSC cream), 11.0 ± 3.0% (PN cream) and 16.8 ± 4.4%(PNSC cream), 11.0 ± 3.2% (PN cream) could be observed, respectively.

All values of PNSC creams were significantly higher than the ones of NNSC creams (Fig. 3), indicating that PS, inducing the positive charge, was crucial for the enhanced efficacy on skin humidity and elasticity. However, this difference was less pronounced than in study 2. The increase might be explained

Table 5
Physicochemical properties and spreading ability of the positively and negatively charged nanoemulsion without stratum corneum lipids on excised human skin surface

Properties	Nanoemulsion	
	Positively charged	Negatively charged
Viscosity (mPa s)	1.011 ± 0.042	0.984 ± 0.050
Surface tension (mN/m)	30.9 ± 0.6	31.3 ± 0.9
Contact angle (°)	40.3 ± 5.1	55.9 ± 6.8
Spreading coefficient (mN/m)	−7.3 ± 1.7	−13.8 ± 2.3

All values are expressed in mean ± S.D. with $n = 5$.

by a stronger interaction of the phytosphingosine induced positively charged nanodroplets in PNSC cream with the surface of the skin, leading to enhanced penetration of the lipids into the stratum corneum. To prove this, ex-vivo spreadability studies were carried out (Table 5). The surface tension and the viscosity of the positively (PN) and negatively (NN) charged nanoemulsions did not significantly differ from each other. However, the spreading coefficients of both formulations were significantly different with values of −7.3 ± 1.7 mN/m for PN and −13.8 ± 2.3 mN/m for NN, caused by different contact angles of 40.3 ± 5.1° (PN) and 55.9 ± 6.8° (NN). Any value approaching zero showed enhanced spreading ability on the skin. Consequently, these results indicated that PN had a higher spreading ability on the human skin surface than NN, caused by stronger interaction of the positively charged nanodroplets with the surface of the skin, leading to improved spreading properties on the skin surface and to the enhanced penetration of the SC lipids and ceramide 3B into the skin. Similar results were obtained by Klang et al. (2000) using the eye cornea of rabbits as a barrier.

4. Conclusion

Because of the need to prove the efficacy and safety of cosmetics, non-invasive and biophysical measuring methods increasingly gain in importance. In this study, methods easy in handling were used to estimate the effect of positively and negatively charged nanoemulsions with stratum corneum lipids on skin properties. It could be shown that the stratum corneum lipids and the phytosphingosine inducing the positive charge, were crucial for the improved effect on skin humidity and elasticity.

These results clearly indicate that phytosphingosine-induced positively charged nanoemulsions are promising carrier systems for the dermal application of low soluble drugs like ceramides.

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