Abstracts

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Wissenschaftliche Posterausstellung 2016: Poster 1

In vivo determination of sweat resistance of sun protection products

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Introduction

Sweating may reduce to a great extent the protecting effect of sun protection products. Thus it is important to apply sweat resistant products especially during physical activities or in the summer time. This poster presents an in-vivo method to investigate sweat resistance based on the ISO method for sun protection determination (1). Sweat induction is triggered by means of a sauna.

Material & Methods

Similar to the water resistance method of the Colipa (2) the sun protection factor is determined dry initially. Another day the products are applied on further test areas on the back of the subjects before sweating is induced through a sauna session of 10 to 15 minutes at 80° C. The time in which a significant sweating may appear very much differs from person to person. Hence sweat induction will be stopped once drops of sweat are clearly visible on the back. The subjects then cool down in climatic conditions of 24°C for 20 minutes before the sun protection factor is determined again.

Results & Discussion

Calculation of sweat resistance is accomplished according to the water resistance method of Colipa (2). If the lower confidence interval lies above 50 % sweat resistance, the claim 'sweat resistance' may be applied. In studies with more than 20 test products developed for sweat resistance results showed 50 % until 90 % preservation of sun protection after sweating. This is comparable with the results we achieved with the water resistance method of the Colipa when investigating prospective water resistant products. The variation of the method is low and thus satisfying. With a confidence interval of around 10 % (mean number of subjects n = 12) it is similar to the ISO sun protection method.

Literature

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Wissenschaftliche Posterausstellung 2016: Poster 2

Equilibrium Dialysis with various membranes for API transport studies

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Introduction:
The distribution of the active pharmaceutical ingredient (API) within multiphase dermal formulations with regard to its impact on API-degradation and skin penetration is little understood. This contribution determines the betamethasone dipropionate (BDP) distribution behaviour with the Dianorm® Equilibrium Dialyser initially produced for analysis of protein receptor bindings [1]. BDP shows UV-detectable degradation products, has low water solubility and has been used in approved formulations [2]. For a robust method development buffer-methanol mixtures were used as one-phase systems to examine the impact of various membranes on the diffusion of BDP. In a second step the micellar solubilisation of BDP in aqueous polysorbate 80 (PS) mixtures was studied (which might affect its degradation) [3].

Methods:
Solvent 1 contained citric buffer pH 5 and methanol (70/30 v/v) while solvent 2 was an aqueous 5% PS (w/w) mixture. Donor medium (dispersion of BDP in solvent) and acceptor medium were filled in PTFE-cells (n=5) separated by membranes and equilibrated at 25 °C for 6 h and 24 h, respectively. The process was performed under sink conditions. Membranes made of polycarbonate (0.1 µm, 0.45 µm), PTFE (0.1 µm, 0.45 µm), cellulose (5 kDa), regenerated cellulose (8 kDa, 25 kDa, 50 kDa) and hydrophilic cellulose ester (HCE, 0.5 kDa, 5 kDa, 20 kDa, 100 kDa) with various molecular weight cut offs (MWCO) were used. Membranes were extracted in methanol. Samples were analysed via UPLC with PDA detector. Statistical analysis was performed with one-way ANOVA and an equivalence test to control the equilibrium of the donor and acceptor medium (α=0.05).

Results:
The solubility of BDP varied between the solvents: 0.456±0.010 µg/ml in citric buffer, >37 mg/ml in methanol, 0.023±0.002 mg/ml in solvent 1 and 0.278±0.000 mg/ml in solvent 2 (5% PS).

As 6 h of running time of the dialysis experiment with solvent 1 were too short for equilibration, 24 h were selected. All membranes except those made of regenerated cellulose (8 kDa: p=0.206, 25 kDa: p=0.321, 50 kDa: p=0.268) reached an equilibrium between donor and acceptor media after 24 h ($p<0.05$). The BDP recovery rate was highest for polycarbonate, PTFE and cellulose membranes (111.8±6.9%). Less than half of the initial BDP concentration (49.7±5.6%) was recovered using the HCE membranes. No significant differences were found within the varying MWCOs of the HCE membranes (p=0.057). The recovery decreased with increasing thickness of the membranes.

5% PS aqueous solutions loaded with 22 µg/ml BDP were dialyzed through HCE membranes with MWCOs of 5 kDa and 100 kDa over 24 h. MWCO of 5 kDa is too low for unhindered diffusion of micellar-bound BDP (molecular weight of PS micelles is around 112-127 kDa depending on quality/distributor) [4]. Thus, only low BDP concentration was found in the acceptor medium (1.2±1.1%, p=1.000). With the 100 kDa membrane micellar BDP transport was determined 45.2±1.3% (p=0.012).

Conclusion:
Membranes showed varying recovery rates depending on their thickness. Micellar solubilisation of BDP as well as its (micellar) transport through membranes with suitable MWCO is possible. A robust method for determination of API transport with the Dianorm® Equilibrium Dialyser was established. Further research is needed to identify the reasons for the low recovery rate. The behaviour of semisolid formulations has to be investigated in the future.

Literature:
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Wissenschaftliche Posterausstellung 2016: Poster 3

Skin-friendly nanocrystals of miconazole nitrate – synergistic combination with chlorhexidine digluconate

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Miconazole nitrate (MN) is a well-known fungicide used against fungal skin infections. Due to the poor solubility of MN in water (1:3000, ethanol 96% 1:140) and the occurrence of skin irritations while using organic solvents or solubilized solutions, the aim was to develop a skin-friendly formulation based on an aqueous nanosuspension with a skin-friendly stabilizer.

Chlorhexidine digluconate is reported to improve the antifungal activity of MN due to synergistic effects [1]. Combination products on the market are e.g. the veterinary shampoo Malaseb® (1 ml Malaseb® shampoo contains 20 mg miconazole nitrate and 20 mg chlorhexidine digluconate). Thus chlorhexidine digluconate was added to the nanosuspension, however it destabilizes suspensions. Thus a screening needed to be performed to identify a physically stable nanosuspension composition. Finally, a potential superior anti-fungal performance had to be tested.

In contrast to e.g. veterinary shampoos, which are washed off after a certain (unpleasant) incubation time on the fur of dogs/cats, the nanosuspension can be applied and forms a long-lasting depot due to the adhesiveness of the nanocrystals. The fungicide effect was determined against S. cerevisiae in inhibition zone assays.

MN 2 % with the same content of chlorhexidine digluconate in various 0.3 % stabilizer solutions were milled by wet bead milling (PML-2, Bühler AG, Switzerland), using 0.2 mm yttria stabilized zirconium oxide beads at 2000 rpm and 5 °C. As skin-friendly stabilizer poloxamer 407 and Tween 80 (even accepted for i.v. injection!) were used. Optimal milling time depending on stabilizer was 30 minutes for poloxamer 407 and 35 minutes for Tween 80. Size analysis was performed by photon correlation spectroscopy (PCS, Zetasizer Nano ZS, Malvern Instruments, UK) and additionally by light microscopy to detect potential larger particles/aggregates outside the measurement range of PCS. Dermal hydrogel formulations with 1 % MN nanocrystal were prepared by magnetic stirring of a freshly produced MN nanosuspension mixed with 5 % hydroxypropyl cellulose (HPC) for >3 hours while remaining of crystal size was monitored. Inhibition zone assays of the formulations were performed for

48h at 30°C on universal yeast agar plates with 20 µl of formulation. All MN nanocrystal formulations were tested against commercial products (“Malaseb”, “KSK Miconazol Crème” and MN dissolved in DMSO) and also against a microsuspensions.

The surfactants poloxamer 407 and Tween 80 were both identified as suitable stabilizers. After 30 respectively 35 minutes of milling time nanocrystals were obtained. For Tween 80 the main nanocrystal PCS diameter was 237nm/254 nm at 0.20/0.25 PCS polydispersity index (PdI) without/with chlorhexidine gluconate. MN nanocrystal size stabilized with poloxamer 407 was 313/350 nm at 0.27/0.20 polydispersity index, respectively. All PCS results were in good agreement with light microscopy, no aggregations. Incorporation into the HPC gel did not change the size distribution, no aggregation was observed by light microscopy. After one month of storage, PCS results showed negligible particle growth of 10 to 40 nm for the suspensions itself. The stability of the nanocrystals was not affected in hydrogel formulations, very few particles > 1 µm were visible by light microscopy.

Determining the formulations antifungal activity by an inhibition zone assay with S. cerevisiae demonstrated the advantages of nanocrystals. Inhibition zone diameters for all nanocrystal gel formulations, with and even without (!) chlorhexidine gluconate, were higher than the inhibition zone diameters for the commercial products having equal active concentration. The positive effect of the nanocrystals became very obvious when comparing them to MN microsuspension. The inhibition test diameters were almost two times higher for nanocrystals. “Miconazol KSK Crème” (commercial product only with MN) was in the same range as the MN microsuspension.

In conclusion, a physically stable nanocrystal combination product, both as suspension and also as hydrogel formulation, were successfully developed. The inhibition zone assay showed the improved fungicide effects compared to microsuspensions and existing commercial formulations. The developed formulation is thus suitable for developing a superior market product.

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Wissenschaftliche Posterausstellung 2016: Poster 4

smartPearls® – a new dermal micro-delivery system for poorly soluble drugs by amorphization

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The poor solubility of many cosmetic actives and drugs limits their dermal bioavailability, e.g. cyclosporin. A physical strategy is to increase the solubility of such actives, i.e. increasing the concentration gradient to the skin and thus passive diffusional flux. Examples are solubilization with surfactants, microemulsions or co-solvents, but all having several disadvantages (e.g. skin irritation, drug precipitation on skin after solvent evaporation resulting in poor penetration). The alternative is to increase the solubility by changing the crystalline state of the drug, i.e. using higher soluble polymorphs or amorphous drug. The common problem of both approaches is the physical instability, which often prohibits use in commercial products. The smartPearls® technology allows to easily generate the amorphous state and keeps it long-term stable in dermal formulations such as gels and creams. All excipients are GMP materials and regulatorily approved for dermal use, i.e. usable in pharmacy prescription and commercial products.

The delivery technology has been transferred from the oral administration route to dermal application. How does the technology work? Drug is loaded into the mesopores of porous materials, e.g. µm-sized silica particles already used in dermal formulations (e.g. Syloid, company Grace). Loading is performed by soaking the pores with organic solution of the drug, and subsequently evaporating the solvent. The drug precipitates, but due to the small dimension of the pores it cannot form crystals, it stays amorphous. Amorphous stability of these powders was shown over more than 4 years [2]. Solvent-free loading of the silica particles is also possible. For dermal application, the powders are simply admixed to the water phase of gels or creams.

The performance of the system was studied using cyclosporin as model drug, being of interest e.g. for psoriasis treatment. Cyclosporin was loaded onto Syloid SP53D-11920, and then particles incorporated into hydroxypropyl cellulose (HPC) gel. The particles proved to be stable in the formulation during storage. As comparative formulations gels were prepared with amorphous µm-sized cyclosporin powder, and amorphous cyclosporin nanoparticles. Skin penetration was studied in the pig ear skin model, using tape stripping. Despite that a) all the formulations contained amorphous cyclosporin, and b) the reference gels contained higher

drug concentration (5% cyclosporin µm-sized powder, 5% cyclosporin nanoparticles versus only 1% cyclosporin in smartPearls®), the penetration from the smartPearls® was superior. The effect was especially visible when normalizing the penetration profiles (µg drug penetrated/% drug content in formulation). Apart from the penetration, highly essential is the physical stability of the amorphous cyclosporin in the smartPearls®, pre-requisite for a product for the benefit of patients.

The smartPearls® technology can also be used in cosmetics and consumer care products. Considering the increasing reluctance of consumers about nanotechnology, it is important on the long term that smartPearls® are no nanoparticles according to EU definition, they are a microparticulate delivery systems, typical sizes from a few µm to about 50 µm.

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Wissenschaftliche Posterausstellung 2016: Poster 5

smartFilms® – a personalized delivery system for dermal application of amorphous drugs & cosmetic actives

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Films, also known as strips, wafers or patches are established in the market as a versatile delivery system. They can be employed for delivery of actives e.g. to the skin, and also to the mucosa of the mouth cavity. Application areas are pharmaceuticals, but also cosmetics and consumer care products. In general, the production technology is rather complex, the smartFilms® technology is the more straight forward alternative. Apart from loading the smartFilms® with nanoparticles (e.g. nanocrystals etc.), actives can be loaded in the amorphous state, which increases penetration into skin/mucosa.

Usually films as delivery system are prepared by solvent casting or hot melt extrusion [1]. In both methods the film forming process is performed in presence of all ingredients. So especially the active ingredients are exposed to stress (e.g. thermal or mechanical). Additionally the relative loading capacity of ingredients is limited below 30 % based of the total weight [1]. Also subsequent modifications of the films are restricted. These limits are eliminated by the new smartFilm® technology [2].

A film forming polymer, often synthetic, a plasticizer and the active ingredient(s) are at least required to produce film products by solvent casting or hot melt extrusion [1]. In contrast just cellulose based paper, the active ingredient(s) and no plasticizers are essentially required to produce film products by smartFilm® technology.

Paper as a porous carrier for loading is formed by a renewable and environment-friendly cellulose fiber network. An adjusted to the ingredients loading liquid (solution, suspension or emulsion) with the desired amount is added once or in multiple to the preformed carrier. Then the loading liquid is absorbed by the carrier. During drying the solvent or dispersion medium is evaporated and the ingredients are remained embedded in the pore matrix. Relative loading capacities of over 80 % related to the total weight were achieved. After loading and drying, the smartFilms® are cut into the final size and form. Both the film forming and the loading process are separated to give high flexibility in dosing. Various papers were used whereby tissue paper, filter paper and blotting paper were preferred due to their high absorbing capacity.

Besides other results, a mainly amorphous state of the loaded ingredients was found after loading with solutions. This amorphous state was investigated over 12 month by now after stor-

age under room conditions. A conversion into a crystalline form is suppressed by the porous structure of the carrier. Based on the amorphous state of the ingredients the bioavailability is increased.

smartFilms® are suitable for different kinds of dermal products for example cosmetics (e.g. face mask for wrinkle treatment), pharmaceuticals (e.g. glucocorticoid treatment) or medical devices (e.g. antiseptic treatment). The effect can be local and/or systemic. A reactivation after wetting the smartFilm® leads to a good adhesiveness to the skin. smartFilms® can be removed anytime from the application spot.

smartFilms® offer a simple delivery system for a dermal application of amorphous ingredients. The smartFilm® technology can be implemented on industrial scale based on the model of the newspaper industry with a conceivable output of millions. On the other hand smartFilms® can be manufactured flexibly at the point of sale e.g. in a pharmacy or a drugstore. So individual, personalized doses in user accepted appearance and taste are available in only little time.

References:

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Wissenschaftliche Posterausstellung 2016: Poster 6

BergaCare smartLipids® – commercial lipid submicron particle concentrates for cosmetics, consumer care & pharma

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BergaCare smartLipids® are commercial submicron solid lipid particles for delivery of cosmetic and pharmaceutical actives to the skin, being in size > 100 nm and < 1 µm. They are a specialized version of the NLC delivery system [1, 2]. In contrast to the "standard" NLC, they are composed of a multiple mixture of lipids (preferentially 5-10). This complex "chaotic" lipid mixture gives them special properties such as increased drug loading and increased physical stability [3, 4]. In addition, based on the size distribution they are no nanoparticles according to the EU cosmetic regulations, i.e. no labelling as "nano" on the final product is necessary. These submicron lipid particles are ideal for the incorporation of lipophilic or amphiphilic actives. The BergaCare smartLipids® are produced with industrial quality under "cosmetic/pharma GMP" and can be purchased as concentrates (10/20-30%) for admixture to dermal formulations such as gels, creams and lotions, or being incorporated into decorative cosmetics. This is the translation of an academic, science-based development into commercial products.
The BergaCare smartLipids® combine the advantages of the previous generation of lipid particles (NLC) with the special feature of the specialized complex lipid mixture. The overall key features can be summarized:

1. High loading & increased physical stability: The complex lipid particle matrix has a low ordered crystalline state (α, ß′) and many imperfections, thus allowing high loading with actives (e.g. instead just 5% retinol in standard NLC now 15%). The actives are firmly enclosed/protected in the particle matrix during storage.

2. Increased dermal penetration: The particles adsorb onto the skin, forming an occlusive film (so called "invisible patch") which promotes penetration. For example, coenzyme Q10 can be made more active compared to existing traditional dermal formulations, allowing a market differentiation. The BergaCare smartLipids® are therefore a delivery system for all lipophilic actives on the market heaving need for penetration/bioavailability improvement.

3. Restoration of natural skin barrier: By adhesion to the skin, an impaired, stressed lipid film of the stratum corneum (= natural barrier) is repaired and re-enforced. The enforced barrier protects the skin against environmental hazards (UV, pollution, etc.). Thus also unloaded

BergaCare smartLipids® are promoting the generation of normal, healthy skin.
4. Protection of chemically labile actives: In contrast to liquid o/w emulsions, a solid particle matrix can more effectively protect actives against degradation, e.g. retinol. Thus these particles are a carrier system for labile actives.

5. Controlled release: The controlled release allows tailor-made particles, for fast action (e.g. lidocaine) to prolonged action, or even avoidance of release in case of actives which should preferentially remain on the skin and not penetrate (UV sunscreens). Due to controlled and prolonged release, skin irritations can be minimized/avoided due to avoidance of too high concentrations on the skin (e.g. as shown for tretinoin).

6. Delivery system for sunscreens: Incorporation into lipid particles increases UV absorption efficiency of molecular sunscreens (= reduction of concentration for same efficiency), and the retarded release reduces skin penetration compared to o/w emulsions (= reduced side effects), e.g. avobenzone.

The first product in launch preparation are BergaCare smartLipids®Q10 and retinol.
Incorporation into products: after production of dermal products the BergaCare smartLipids® concentrate is simply admixed under gentle blending. BergaCare smartLipids® formulations are also offered customized. Developments can also be made for pharma actives (drugs). A pharmaceutical contract manufacturer is available.

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Wissenschaftliche Posterausstellung 2016: Poster 7

BergaCare smartLipids® lidocaine – increased dermal anesthetic potency & duration of action at reduced risk of serious adverse reactions

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Lidocaine ointments with a high drug content of up to 20% are commonly used to anesthetize the skin sufficiently prior a painful and invasive skin treatment such as tattooing or lasering. Two formulation problems occur: Firstly, the lidocaine ointment has only a short shelf life of less than one week since such a large quantity of lidocaine cannot be completely dissolved even in a highly lipophilic ointment such as petroleum jelly. Consequently Ostwald ripening progresses leading to formation of sharp needle-like crystals irritating the skin and impair the penetration. Secondly, lipophilic ointments cannot be entirely removed from skin without soap or alcohol. Because both should be avoided prior to an invasive skin treatment, non-removed lidocaine can enter the blood circulation unhindered increasing the risk of serious adverse reactions e.g. tremor, dizziness, respiratory depression and cardiac arrhythmia. To overcome this problem a hydrophilic, better removable lidocaine formulation is required. However, just dissolving the lidocaine directly in a hydrophilic base at skin pH would form single charged cations which only poorly penetrate the skin, thus decreasing distinctly the bioavailability. Therefore lidocaine was loaded into smartLipids® and then incorporated into easily removable hydrogels.

smartLipids® are the optimized 3rd generation of lipid nanoparticles after the solid lipid nanoparticles (SLN) and nanostructured lipid carriers (NLC). Strictly speaking, they are a specialized version of the NLC. The special about smartLipids® is their clearly increased drug loading capacity with firm drug inclusion during storage. The difference is in the lipid composition. The lipid matrix of the SLN typically consisted of only 1 solid lipid, the NLC of 1 solid lipid combined with 1 liquid lipid. In contrast, the smartLipids® matrix is composed of multiple lipids, e.g. 5 to 10 lipids causing a “chaotic” particle matrix structure. The many imperfections and less ordered lipids allow a higher drug loading, which was exploited for formulating lidocaine. The lidocaine showed an initial maximum loading of 15% in SLN and 25% in NLC, but the lidocaine was expelled from the NLC matrix after only 2 weeks of storage at room temperature. In contrast, lidocaine at 40% could be loaded in the optimized smartLipids® matrix consisting of Polyglyceryl-2 Dipolyhydroxystearate, Octyldodecanol, Carnauba (Carnauba Wax), Candelilla Cera (Candellila Wax), Cera Alba (Beeswax), Cetearyl Glucoside, Cetearyl Alcohol and MCT (medium chain triglycerides).

A particle suspension composed of 20% lipid-drug matrix, 2% Plantacare 810 UP, 1% Sisterna PS 750C and 77% distilled water was produced by high pressure homogenization (500 bar, 3 cycles, at 75°C). A small skin-adhesive particle size of 143 nm was achieved and the formulation showed no drug expulsion with stable particle size over 3 months up to now.

To transfer the suspension into a dermally applicable form, hydroxypropyl cellulose was used to from a hydrogel containing 8% lidocaine. Physical stability of the particles in the gel was shown at both 5°C and room temperature over 3 months up to now.

The potency and duration of action of new developed hydrogel was compared with a 20% lidocaine petroleum jelly formulation in a double blind and triply performed in-vivo orientating study. 50 mg formulation was applied on 2cm x 8cm sized area at the inner side of the forearm and in short intervals the treated area was pricked with a blunt needle. The numbness had to be evaluated from 0% to 100%, where 100% represented the totally numbness. Regarding the potency lidocaine smartLipids® hydrogel was clearly superior with a maximum numbness of 60% reached already after 15 minutes, were the standard formulation never exceed 20% during the whole test period although having a 2.5 times higher lidocaine concentration. With regard to the analgesic duration, the hydrogel showed again considerable advantage with a long lasting numbing effect of 35 minutes compared to the standard formulation with duration of only 20 minutes. After 40 minutes of exposure time the hydrogel dried out completely forming a thin and elastic film which was removed by pulling off in one piece. In contrast, the standard formulation was wiped off with a tissue conventionally done in laser centers. Afterwards, the relative reminding lidocaine content on the skin surface was investigated. Even after removing the standard formulation very carefully from the skin, 2% of applied lidocaine amount remained, whereas less than 0.1% and thus 20 times lower amount of lidocaine was detected on the skin surface treated with the lidocaine smartLipids® hydrogel.

To sum up, lidocaine smartLipids® hydrogel was developed having a 12 times prolonged shelf life with three times increased potency. The duration of action almost doubled compared to the standard formulation, which had even 2.5 times higher lidocaine concentration. In addition, the removability of the formulation from skin is 20 fold improved and thus the safety of the formulation regarding serious adverse reactions is distinctly increased.

Wissenschaftliche Posterausstellung 2016: Poster 8

Dermal smartCrystals® – new model for better understanding of nanocrystal penetration mechanism

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smartCrystals® are nanocrystals, which consist of pure active, being in size in the nanodimension (< 1,000 nm = < 1 µm). Due to their small size they have nanoproperties such as increased saturation solubility (Cs), and thus increased penetration into the skin. They are an ideal formulation to increase skin penetration of poorly soluble actives (e.g. flavonoids), but also medium soluble compounds such as caffeine. Curcumin is a very interesting active for dermal delivery, e.g. anti-inflammatory, anti-oxidant and anti-cancer properties. In addition, it is fluorescent, easy to detect, and thus it was used as model molecule. Curcumin has low water solubility, low penetration into the skin, and yellowish color of limited consumer acceptance. Thus the aim was to develop a formulation with distinctly increased bioavailability compared to existing commercial products, and curcumin concentration as low as possible to minimize color effects. For this, penetration was investigated as function of nanocrystal concentration in the dermal formulation. From the findings, a new model of penetration enhancement was derived. By now, the sufficient concentration of nanocrystals in dermal formulations was roughly estimated. The new model allowed for the first time to give a scientifically well-based concentration recommendation for nanocrystal formulations.

smartCrystals® were produced by bead milling (PML 2, Bühler, Switzerland) and subsequent high pressure homogenization (Micron LAB 40, APV Deutschland). The suspension consisted of 5% curcumin, 1% Plantacare 2000 UP (all w/w %) and distilled water. Characterization was performed by photon correlation spectroscopy (PCS), laser diffraction, light microscopy and electron microscopy. For penetration studies, aqueous nanosuspensions and nanocrystals in hydroxypropyl cellulose (HPC) gels were applied to pig ear skin in a Franz cell, and after certain times (1-20 hours) skin slices were taken with a microtom, and analyzed by confocal laser scanning microscopy (CLSM). Nanocrystal concentrations investigated were 2%, 0.2%, 0.02% and 0.002%. As comparison, a commercial dermal curcumin product with 0.0001% dissolved curcumin (= saturation solubility) was used.

From the nanocrystal theory by now, only the saturation solubility of nanocrystals was considered as the dominating factor, leading to a distinct penetration driving concentration

gradient (Cs-Cskin). Based on this, the penetration should be the same for 2% to 0.002% nanocrystal formulations (all have identical Cs). However, similar good penetration was found for 2%, 0.2% and 0.02%, but very little penetration for 0,002%. Obviously the density of nanocrystals on the skin surface plays also an important role. The density needs to be so high, that the "diffusional coronas" of dissolved active around the nanocrystals on the skin surface overlap. This provides a continuous, skin surface covering layer of highly concentrated dissolved active. As a rule of thumb, a concentration as low as 0.02% can now be recommended for dermal nanocrystal formulations. This reduces costs for the nanocrystals, and at the same time reduces color effects of colored actives such as curcumin.

The commercial product with dissolved curcumin (= solution) showed practically no detectable fluorescence in the skin, in contrast to nice fluorescence of the 0.02% nanocrystal formulation. This confirms the "old galenic rule" of better penetration from "suspensions", and the superiority of the nanocrystal technology.

Wissenschaftliche Posterausstellung 2016: Poster 9

3rd generation technology – BergaCare smartLipids® submicron carrier for improved retinol formulation

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The solid lipid nanoparticles (SLN®) [1] are the first generation, the nanostructured lipid carriers (NLC®) [2, 3] the second generation of "lipid nanoparticles with solid particle matrix". In 2014 the third generation was developed, the smartLipids® [4, 5], being technically a specialized version of the NLC.

Basic difference is the lipid composition for particle production. The SLN consist typically of 1 solid lipid, the NLC of a blend of a 1 solid lipid and 1 liquid lipid (oil).The 3rd generation consists of a "chaotic" mixture of 5-10 lipids, either solid lipids only, or with limited admixture of oils. The chaotic mixture leads to more imperfections in the particle matrix, thus to an increased loading capacity for cosmetic and pharmaceutical actives. Retinol was incorporated into smartLipids® particles. The loading, reported in the literature, with firm incorporation was 1% for SLN, 5% for NLC, but 15% could be incorporated into smartLipids®. In addition, localization in imperfections inside the particle should protect the actives against degrading influences, i.e. increasing the chemical stability.

The complex lipid mixture delays/avoids polymorphic transitions of the lipid particle matrix to more ordered ß modification, which can lead in SLN (and partially in NLC) to expulsion of loaded actives from the particle matrix (in more highly ordered particle matrix is no space anymore for foreign molecules). The smartLipids® approach increases firm inclusion during storage.

The particle size produced is preferable >100 nm and <1,000 nm (= 1 µm), i.e. being in the "submicron range" and legally the particles are no nanoparticles (no declaration according to EU guidelines as "nano" on products). However, due to the size in the nanodimension (< 1,000 nm), these submicron particles possess still special skin beneficial nano-properties, but being legally no nanoproducts. The mean sizes of the bulk populations of the produced retinol smartLipids® was in the range 112 nm to 400 nm (determined by photon correlation spectroscopy - PCS).

Retinol was incorporated into smartLipids prepared from medium to highly chaotic lipid mixtures (i.e. 7 or 8 lipids of different complexity, e.g. mono- to triglyceride content, at simultaneously very different fatty acid chain lengths). To study additionally the influence of

the stabilizer on chemical stability, particles were prepared with different stabilizers, e.g. anti-irritant sodium cocoamphoacetate. The stabilizer affects the stability via its influence on the crystalline structure of the particle matrix (i.e. delay or acceleration of polymorphic transitions, subsequently reduction of imperfections for protective localization of active). Particle suspensions were stored at room temperature and at 40 °C. Stability data were compared with previously published stability data by Jenning [6] and Hommoss [7].

A very pronounced effect of the stabilizer was found, sodium cocoamphoacetate was most protective for retinol. Both high but also the medium chaotic lipid mixture (only solid lipids) had a good stabilizing effect on retinol (e.g. 94-97% both after 3 months at room temperature, 48% (medium) and 70% (highly chaotic mixture) at 40 °C). After 6 months at room temperature the stabilities of retinol in smartLipid mixtures (solid and liquid lipids) were about 88%, clearly higher than [7]. It should be pointed out, that the mixtures did not contain added anti-oxidants (e.g. BHA, BHT etc.), the pure protective effect of the carrier was investigated. The combination of protective effect and long-term firm inclusion into the particle matrix by absence of polymorphic transitions makes the smartLipids® a highly attractive carrier not only for retinol, but generally for chemically labile cosmetic and pharmaceutical actives.

For the final formulation, in addition to the protection by the lipid matrix, antioxidants were added for perfect protection. A study was performed adding different antioxidants, e.g. BHT, Tinogard TT etc., and the formulations were stored for 6 months at stress condition of 40°C. Even under this stress, the best combinations had a remaining retinol content of about 67%. Thus, long term stable retinol in dermal formulations is possible by effective combination of NLC/smartLipids and anti-oxidants.

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Wissenschaftliche Posterausstellung 2016: Poster 10

Extemporaneous Formulations – Communication between pharmacists and physicians in “Hautapotheke”-Pharmacies

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Introduction:
Due to the pharmacy practice order in 2012, the requirements for prescribing and manufacturing extemporaneous formulations have increased. Magistral formulations should be favored over individual formulations. There is a high need for a good cooperation between pharmacists and physicians to gain optimal results.

In Germany, about 8 million extemporaneous formulations compounded by pharmacists for topical therapy are produced yearly. To evaluate the requirements on this topic in pharmacies, we created a standardized questionnaire sent to every pharmacy which is part of the network “hautapotheke.de“ by the “Gesellschaft für Dermopharmazie e.V.”.

Methods:
The questionnaire with 16 questions was sent out to 140 “hautapotheke”-pharmacies in Germany: 3 questions to answer with free text, 7 to answer by ticking boxes and 6 - a combination of both question-types.

Results:
Overall, 41 of 140 (29,3%) questionnaires were returned via fax, E-Mail or postal way. 33 (82,5%) stated, that extemporaneous formulations are an “important therapeutic alternative” in daily practice. None of the participants think that extemporaneous formulations are “obsolete”. Every second (n=22 (55%)) complained about “the excessive work with poor reward”. 15 (37,5%) confirmed, that the extemporaneous formulations are “necessary but not always practicable”.

65% of all extemporaneous formulations are prescribed without following any magistral standards, 13% are “magistral formulations exclusive NRF” and 20% are formulations compounded according to the NRF.

To evaluate the cooperation, the participants were asked to state on the communication practice between pharmacists and physicians concerning the topic “extemporaneous compositions”. Only 5 (12,2%) stated “very constructive”. The majority (24; 58,5%) think that the communication is “associated with anger” and that the communication is “constructive, but too costly in terms of times”(23; 56%). Interestingly 12 (29%) complained that “the communication is not taking place or takes place on a too small scale”.

Conclusion:
Considering our findings, extemporaneous formulations are necessary and helpful tools in treating patients with dermatological diseases. Beside the improvement of communication between physicians and pharmacists, the prescription behavior should change by increasing prescriptions with magistral extemporaneous formulations. Further research should explore and compare the questionnaires with physicians in Germany.

Wissenschaftliche Posterausstellung 2016: Poster 11

Improved quantification of skin erythema pattern by color analysis of highly standardized photographic images

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Introduction
Visual scoring is the time–honored approach for quantification of erythema. With appropriate training visual scoring can be done efficiently, however with only moderate precision and considerable inter-grader variability. Measurements with chromameters are device-based alternatives with greater precision and objectivity. They have, however, their own limitations. First, they are not truly operator-independent. Second, the measurement area is limited to approx. 1 cm². Third, complex pattern of erythema as present in most diseases cannot be quantified satisfactorily with chromameters. Usually chromameters integrate over the entire area measured and the intense redness of patchy areas or teleangiectasia are averaged with skin parts without any erythema. Erythema evaluation by quantitative image analysis on highly standardized photographs can overcome these limitations.

Objective & Methods
For advanced color measurements on images the photographic device must provide correct and highly reproducible illumination in terms of color temperature and light intensity. Further all geometric parameters as distance to skin, magnification and angle of illumination must be kept constant.

In this investigation we compare measurements on images obtained with a handheld professional camera system with a distance holder and flash illumination (Canon EOS 5D Mark II with Elinchrom RQ Ringflash ECO) to color measurements with a chromameter (Minolta Chromameter CR400) and visual scoring. Erythema spots of 1 cm² of different intensity were induced on volar forearm skin of six subjects with a solar simulator. Erythema was measured in the L*a*b*- color system with the chromameter and in parallel by quantitative image analysis in photographs.

In a second step facial skin of Rosacea Type I patients displaying teleangiectasia was evaluated by visual scoring, chromametric measurements and image analysis. After that an unobtrusive camouflage product was applied and the assessments were repeated.

Results & Conclusions
The erythema data obtained from the photographs on 1 cm² erythemal spots induced by UV-light showed a good correlation (r > 0.9) with visual scoring as well as with chromameter

measurements. These results showed that the erythema quantification from photographs had a satisfying precision.

The measurements of camouflage effects on teleangiectatic areas revealed the advantage of the photographic method. The camouflage efficacy derived from the chromameter measurements were not well in line with the visual clinical observation. In the color assessment from the photos not only the average redness but also the variation of red blood vessels and the unaffected surrounding skin were taken into account and the blood vessels were measured separately from the surrounding skin. As a result the measurements from photographs matched much better with the clinical observations.

In summary, erythema quantification from photographs showed a good correlation with chromametry when the area of interest showed homogenous erythema. In areas with uneven color distribution the color quantification of photographs had the clear advantage because erythema intensity, could not only be over averaged over the whole area but also quantified selectively and hence with greater sensitivity in areas/structures of interest, e.g. teleangiectasias.

Wissenschaftliche Posterausstellung 2016: Poster 12

Differential scanning calorimetry on human nail clippings, keratin films and bovine hoof plates – effect of formulation ingredients

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Introduction:
Differential scanning calorimetry (DSC) measurements are employed to analyse the interaction between formulations and human stratum corneum (SC). If the SC is heated up to 120 °C, four endothermic transition shifts are detected (T1 – T4) with T2 und T3 appearing reliable in each thermogram (Barry, 1987). In this contribution, we analogously tried to investigate the influence of excipients and formulations on nail clippings and nail plate models. As nail plate models, keratin films (KF) manufactured according to Lusiana et al. (2011) and bovine hoof plates were utilised. The formulations included in this assay were composed of poloxamer 407 (P407), propylene glycol (PG), isopropyl alcohol (IPA), medium chain triglycerides (MCT), double distilled water and the antifungal active ingredient ciclopirox olamine.

Methods:
The P407-based formulations were manufactured according to Täuber and Müller-Goymann (2015). DSC measurements were carried out to evaluate the interaction between excipients/formulations and nail clippings as well as artificial nail plate models. Prior to the experiments, nails, KF and bovine hoof plates were hydrated in a desiccator filled with saturated sodium chloride solution for at least 48 h at room temperature. Following, insertion either in the excipient or in the P407-based formulation (n = 3 – 5) for 0.5 – 3 h at 32 °C was carried out. Subsequent DSC measurements were executed from 30 – 250 °C with a heating rate of 20 K/min by using a DSC 1 MultiSTAR HSS7 System (Mettler Toledo GmbH, Gießen, Germany).

Results:
When heating up nails and nail plate models to 250 °C, three endothermic transition shifts (T1 – T3) were detected. However, only T2 appeared reliable in each thermogram and was hence used for evaluation. T2 of the nail plate models was detected at approximately 140 °C, whereas T2 of the nail clippings was at 167 °C. Incubating the nail clippings for 0.5 h in the excipients led to a slight increase of T2 with exception of water. Regarding the nail plate models, only MCT evoked a considerable transition shift. Prolonging incubation time up to 3 h evoked an increase of T2 for all membranes when incubating them in PG and IPA,

respectively, despite high standard deviation (SD) up to 29 °C in the case of IPA. Incubation of nails and nail plate models in the P407-based formulations for 0.5 h did not evoke significant transition shifts of T2.

Conclusion:
Influence of excipients on nails and nail plate models could be measured with DSC. Both nail plate models indicated similar transition shifts, but differed from human nail clippings.

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