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Influence of Sucrose Esters on the In Vivo Percutaneous Penetration of Octyl Methoxycinnamate Formulated in Nanocapsules, Nanoemulsion, and Emulsion

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ABSTRACT The influence of sucrose laureate and sucrose oleate on the in vivo percutaneous penetration of octyl methoxycinnamate (OMC) formulated in i) colloidal suspensions (nano-emulsions and nanocapsules), and ii) conventional o/w emulsions was evaluated. The results showed that nano-emulsions formulated with sucrose laureate exhibited the highest penetration in the stratum corneum compared to the other formulations. A two-fold increase in OMC skin deposition was observed with the nano-emulsion containing sucrose laureate when compared to the control. The data obtained suggest that the total amount of OMC detected in the stratum corneum and the penetration depth are strongly dependent upon the formulation's nature, the particle size, and the type of enhancer.

KEYWORDS Nanocapsules, Nano-emulsion, Sucrose esters, Penetration enhancement, Tape-stripping

INTRODUCTION

The importance of the transepidermal pathway to transport a variety of drugs into and across skin has received considerable attention over the last years (Jadoul et al., 1995; Cevc et al., 1996; Ramachandran & Fleisher, 2000; Fonseca et al., 2004; Prausnitz, 2004; Fitzpatrick et al., 2004). It is well known that the rate-limiting step for diffusion through the skin occurs in the intercellular lipid domains of the stratum corneum (the outermost layer of the skin). The polymorphic nature of these lipids, and the interactions among themselves and with proteins, lead to diverse membrane states (e.g., gel-liquid crystal transitions) (Grubauer et al., 1989; Potts & Francouer, 1991). Several approaches have been reported to reversibly remove the skin barrier resistance (Denet et al., 2004; Kalia et al., 2004; Williams & Barry, 2004; Cotte et al., 2004). We recently reported that skin treatment with sucrose esters (SEs) transiently

increased membrane permeability due to the ability of this molecule to extract and "fluidize" the lipid domains. Fluidization is related to an increase in the ratio of gauche/trans conformers along the lipid hydrocarbon chains (Ayala et al., 2003). However, the properties of SEs to promote the penetration of active agents formulated in colloidal formulations remain to be determined. Colloidal preparations have shown to improve drug delivery across membranes by mediating a more direct and prolonged contact of the carrier than would be possible with a single molecule dispersed in a solution. Nanocapsules (NCs) and nano-emulsion (NE), for instance, possess numerous advantages including the possibility of controlled drug release and drug targeting, and the incorporation of a great variety of therapeutic actives. It would be desirable to use colloidal formulations in combination with safe surfactants such as SEs to improve the transport of highly lipophilic substances through the skin.

SEs are nonionic surfactants with a sucrose substituent as the polar head group (Fig. 1). Because they are relatively innocuous and biodegradable surfactants, they are suitable for therapeutic and cosmetic applications (Mitsubishi-Kasei Foods Co., 1987; Thevenin et al., 1996). Their application in dermatology has been accentuated by the ability of some SEs, such as laureate and ricinoleate, to form liquid crystals and microemulsions (Tambini et al., 1993; Lehmann et al., 2001).

This paper focuses on the penetration of octyl methoxycinnamate (OMC) incorporated into NCs, NE, and emulsion (EM) formulations through human skin in vivo, evaluating the enhancement effect of sucrose laureate (SL) and sucrose oleate (SO).

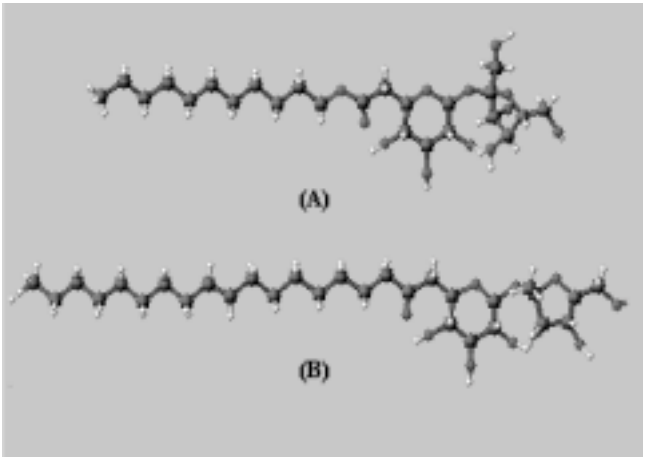

FIGURE 1 **Chemical Structure of Sucrose Esters: (A) Sucrose Laureate and (B) Sucrose Oleate.**

MATERIALS AND METHODS

Materials

Sucrose laureate (SL) (Ryoto Sugar Ester® L-1695) and sucrose oleate (SO) (Ryoto Sugar Ester® O-1570) were kindly provided by Mitsubishi-Kasei Food Corporation (Tokyo, Japan). Octyl methoxycinnamate (OMC) was purchased from Multiquim (Mexico City). Analytical grade 2-butanone was obtained from Fluka, Buchs, Switzerland. Cellulose acetate phthalate (CAP) was purchased from Vita Drug S.A. (Mexico City). The nonionic stabilizing agent was poly(vinyl alcohol) (PVAL; Mowiol® 4–88, mw: 26000, Glomarza, Mexico City). Scotch Book tape 845 was provided by 3M (St. Paul, MN, USA). Aqueous solutions were prepared using deionized water supplied by a Milli-Q water purification system (Millipore Corp., Bedford, MA, USA). All other reagents were of analytical grade and were obtained from Fermont (Monterrey, Mexico).

Preparation of Systems

Nanocapsules (NCs)

Nanocapsules (NCs) were prepared by the emulsification–diffusion technique (Quintanar-Guerrero et al., 1998). Typically, 2-butanone and water were mutually saturated at 25°C for 48 h in order to ensure initial thermodynamic equilibrium of both liquids. Fifty mg of CAP and 100 µl of OMC were dissolved without heat in 5 mL of water-saturated 2-butanone. The resulting organic solution was emulsified with 20 mL of a 5% w/v PVAL 2-butatone-saturated aqueous solution using a mechanical stirrer (Caframo RZR-1, Ontario, Canada; Turbine propeller IKA-1381, Germany) at 2500 rpm for 10 min. Two hundred mL of water were subsequently added to the emulsion to induce diffusion of the solvent into the continuous phase leading to the formation of NCs. The NCs suspensions were then concentrated under reduced pressure at 40°C, to a final volume equivalent to 1% (v/v) OMC. These dispersions were used for skin penetration studies.

Entrapment efficiency (EE) was calculated according to the following equation (Leroux et al., 1995):

$$EE(\%) = \frac{DL(\%)}{\text{Percent of the initial OMC content} \times \text{(1-fraction of residual PVAL)}} \times 100 \quad (1)$$

where DL corresponds to drug loading, and is calculated as follows:

$$DL\ (\%) = \frac{\text{weight of OMC in NCs}}{\text{weight of recovered NCs}} \times 100 \quad (2)$$

Because a certain amount of PVAL is adsorbed on NCs during the manufacturing process, the correction factor (1-fraction of residual PVAL) was introduced into Eq. 1 to avoid underestimation on the EE. The initial OMC content corresponds to the theoretical OMC loading and was calculated from the weight of OMC used in the process divided by the weight of OMC and the weight of the polymer used for NCs preparation. Determinations of EE were made in triplicate.

The process efficiency (PE) was calculated according to the following equation:

$$PE(\%) = \frac{\text{weight of recovered NCs}}{\text{weight of OMC} + \text{weight of CAP} - \text{weight of residual PVAL}} \times 100 \quad (3)$$

The weight of OMC corresponds to the OMC loaded into NCs, and the weight of CAP is the amount of CAP used in NCs preparation. The weight of residual PVAL was determined from the average result of PVAL residual assays. The residual amount of PVAL in NCs was determined by the traditional colorimetric method using iodine in the presence of boric acid (Allémann et al., 1993).

Nanoemulsion (NE) and O/W Emulsion (EM)

Nanoemulsion (NE) was prepared by the same technique described earlier using 0.5 mL of OMC without adding the polymer (Chavez et al., 2002). In order to prepare the EM, 0.5 mL of OMC were emulsified with 80 mL of a 5% PVAL aqueous solution. This mixture was agitated for 10 min with a mechanical stirrer at 2500 rpm (Turbine propeller IKA-1381, Germany) and then concentrated to the desired final volume (equivalent to 1% v/v OMC).

For the systems containing the enhancers, 80 mg of either SL or SO, corresponding to a concentration of 1% w/v, were incorporated to each system (NCs, NE, EM) after the formation of the systems.

Particle Size Analysis

The formulations were characterized by their average size using a Coulter N4 light-scattering particle-size analyzer (Coulter, FL, USA). The samples were analyzed in triplicate for all the batches prepared.

Scanning Electron Microscopy (SEM)

Nanocapsules (NCs) morphology was characterized by a fixation procedure with osmium tetroxide. Millipore filter membranes (pore diameter 0.22 μm) were used as the carrier medium. Membrane squares (0.5 × 0.5 cm^2) were dipped into the concentrated NCs suspensions with tweezers, dried, and then exposed to osmium tetroxide vapor. A set of samples of NCs suspensions on the carrier were fixed with unbuffered 1% osmium tetroxide for at least 1 h at 25°C. Finally, the dried samples and the carrier medium were mounted on stubs and coated with gold particles (20 nm) by a Sputter Coater JFC-1100 (JEOL, Japan) and observed under a scanning electron microscope JSM-25SII (JEOL, Japan), operating at 15 KV. The magnification adopted here was 7000×.

In Vivo Penetration Studies

Three healthy volunteers (aged 22–28 years, Caucasians, phototype V) (Martini, 2003), with no history of skin disease, gave their written informed consent. During the study, volunteers were required to maintain the midventral forearm free from the application of any pharmaceutical or cosmetic skin products. The skin sites were gently cleansed with water and dried. At the beginning of the experiment, the subjects were dosed topically with 7 mL of either NCs, NE, or EM formulations containing 1% (v/v) OMC. The systems were applied via perfusion cells (17.34 cm^2). After 1 h of contact, the area was wiped clean with a water-soaked cotton ball and dried gently.

To determine the distribution of OMC across the stratum corneum (SC), 20 sequential tape strippings were made using Scotch Book Tape 845 (3M) (Alberti et al., 2001). Octyl methoxycinnamate (OMC) was

extracted by soaking the tapes in 5 mL of methanol/ethyl acetate (1:1) for 12 h at 25°C. The amount of OMC in each tape was quantified by spectrophotometry at 308 ηm. A solution of methanol/ethyl acetate (1:1) was used as the reference. To anticipate possible interferences of the scotch tape and skin components, the solvent (methanol/ethyl acetate) was previously put in contact with the tapes provided by 20 sequential tape strippings made without formulation. The penetration depth of OMC was calculated assuming a constant tape-stripped area and a density of approximately 1 g/cm^3 for the stripped SC (Kalia et al., 1996). In this way, the mass of the SC removed (determined by weighing the tapes before and after stripping) was converted to the depth of SC sampled (Alberti et al., 2001). The SC thickness removed was normalized with respect to the total SC thickness, thereby allowing direct comparisons between subjects. The total thickness was determined using an original approach described elsewhere (Kalia et al., 1996; Alberti et al., 2001), involving transepidermal water loss (TEWL) measurements, which were performed during the sequential tape-stripping process. Transepidermal water loss (TEWL) was measured with a Courage & Khazaka Tewameter TM 210 (Cologne, Germany) with a resolution of 0.1 g/h.m^2. Measurements were carried out in a ventilated Plexiglas chamber at a temperature of 23 ± 2°C and a relative humidity of 45–55%. Volunteers remained undisturbed and relaxed during the experiment to avoid erratic fluctuations in TEWL. Since the TEWL is inversely proportional to SC thickness, the latter can be deduced from the former. The average and standard deviation of the TEWL values (8/hm^2) were calculated. ANOVA was employed to test the level of significance and Duncan's procedure was used to identify the source of the difference when a significant F value was found.

RESULTS AND DISCUSSION

All of the formulations were characterized by the average particle size. As shown in Table 1, the size of the systems follows the next order: EM >> NCs > NE. The formation of NCs was confirmed by measuring density using differential centrifugation (Olvera-Martínez et al., 2005), and the structure was visualized by a direct SEM technique without freeze-fracture.

Figure 2 shows the characteristic spherical shape of these systems using osmium tetraoxide as fixative agent. Samples presented the same spherical morphology, whose size is consistent with the measurements previously made.

TABLE 1 **Average Particle Size of OMC Loaded CAP Nanocapsules Prepared by the Emulsification–Diffusion Method (*n* = 3)**

OMC systems	Particle size ± S.D. (nm)	Aggregates[a]
Control EM	2707.9 ± 163.6	Non
1% SO-EM	2644.0 ± 198.1	Non
1% SL EM	2889.1 ± 51.3	Non
Control NE	161.7 ± 23.8	Non
1% SO-NE	127.8 ± 12.9	Non
1% SL-NE	124.4 ± 14.9	Non
Control NCs	458.3 ± 25.8	Non
1% SO-NCs	417.3 ± 37.7	Non
1% SL-NCs	362.8 ± 40.2	Non

[a]Flakes of polymer on the surface after preparation.

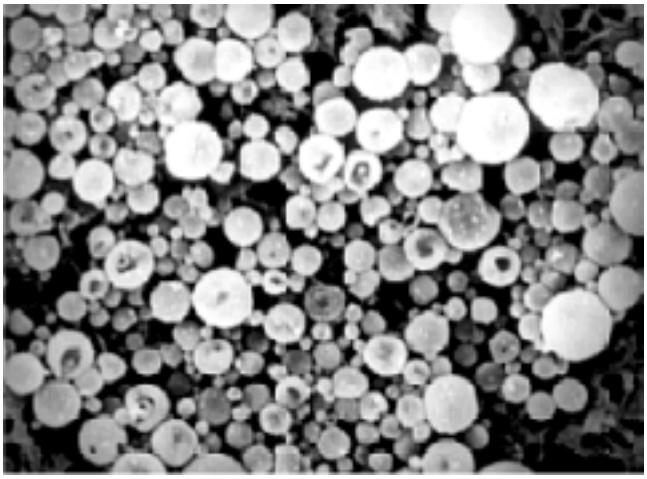

FIGURE 2 **SEM Images of OMC-NCs Stained with Osmium Tetraoxide (7 000×).**

The systems' efficacy was evaluated as a function of their ability to improve OMC penetration through the SC. Two parameters were determined: i) the total amount penetrated, and ii) the penetration depth.

Figure 3 shows the total amount of OMC determined in the tapes representing the quantity penetrating through the SC. As it can be seen, OMC penetration was immensely affected by both formulations and the presence of SEs. These findings indicated that in the absence of enhancers, NE and EM deliver OMC into skin layers much more efficiently than NCs (F = 27.1095; $F_{0.05/2,2}$ = 3.5546). This is reasonable since both systems contain OMC in a liquid phase. Moreover, NE globules can be easily

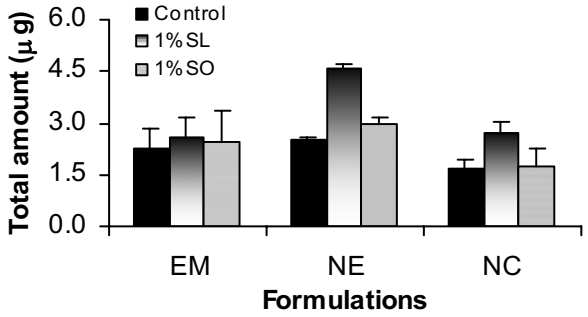

FIGURE 3 **Total Penetrated Amount of OMC Delivery from Emulsion (EM), Nanoemulsion (NE), and Nanocapsules (NCs) Per Unit Area of SC (*n* = 3).**

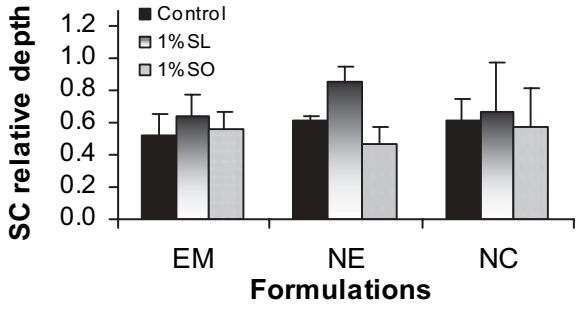

FIGURE 4 **Effect of Sucrose Laureate (SL) and Sucrose Oleate (SO) on the SC Penetration Depth of OMC (*n* = 3), Emulsion (EM), Nanoemulsion (NE), and Nanocapsules (NCs).**

distributed into the skin surface due to their particle size and deformability.

When 1% SL was added to NCs and NE suspensions, the OMC amount penetrated was appreciably increased (about 2-fold greater than the control). Analysis of variance and Duncan's procedure revealed significant differences between the amount penetrated with NE containing 1% SL and that penetrated with the control NE, NE containing 1% SO, NC, and EM formulations. NCs containing SL apparently increased the quantity of drug penetrated into the SC. Nevertheless, no significant differences were found when comparisons were made with control NCs and NCs containing SO. It is important to mention that although NCs are efficient as drug delivery systems, the nature of the active agent, type of polymer, as well as the administration route, are factors to be taken into account. On the other hand, treatment of the skin with formulations containing 1% SO did not modify the OMC content compared to the control systems. Although NEs containing SO showed a slight increase in the OMC amount penetrated, the effect was not as marked as with those containing SL.

The total penetration depth of OMC for the skin treated with NE, NCs, and EM is depicted in Fig. 4. As shown, the penetration depth did not differ among the control formulations. Apparently, the particle size did not affect OMC penetration depth.

A similar trend was observed with the subsequent addition of SL and SO to OMC systems. Analysis of variance did not reveal any difference between the type of formulation (F = 0.1110; $F_{0.05/2,2}$ = 3.5546) and the enhancer (F = 1.4711; $F_{0.05/2,2}$ = 3.5546).

It is well known that the penetration enhancement of drugs through the skin is related with the alteration of the properties of the lipoidal intercellular pathway and the polar pathway of SC. In this sense, the interaction of the formulation's components with SC intercellular lipids is of crucial importance for the effectiveness of a penetration enhancement action. Enhanced permeation of lipophilic molecules through SC is frequently associated with transitions (especially gel to liquid crystalline transition) involving the hydrocarbon chains of the SC lipid components. Previous to the transition, the acyl chains of the lipids are all primarily planar trans conformers. Immediately after transition occurs, substantial numbers of gauche conformers are introduced into the fatty acid chains, resulting in an increased molecular motion (fluidization) (Suhonen et al., 1999).

Many penetration enhancers are capable of inserting themselves between the hydrophobic tails of the bilayer, thus disturbing their packing, increasing their fluidity, and subsequently, leading to an easier diffusion of lipid penetrants. In the case of SL, the potential exists for their long hydrocarbon chain to be inserted between the lipophilic tails allowing the sucrose ring to interact with the polar lipid head groups. As we know, the interaction with these groups can modify hydrogen bonds and ionic forces, disturbing the hydration spheres of the lipids, thus resulting in alteration of the head group domain (Barry, 1987). Biophysical evidence on the extent of the effect of this enhancer has been provided through measurements of attenuated total reflectance Fourier transform infrared (ATR-FTIR) spectroscopy (Ayala et al., 2003). This study demonstrated that treatment of the skin with SL significantly increased the extent of penetration of a model penetrant relative to the control. The C-H_2

asymmetric and symmetric stretching bands of the lipid methylene groups of the skin were characterized by decreased absorbances and frequency shifts to higher number wavenumbers. It has been shown that shifts in these frequencies reflect increased lipid fluidity and increase in the proportion of gauche conformers along the lipid hydrocarbon chains (Casal & Mantsch, 1984; Knutson et al., 1987).

The data presented here demonstrated that the inclusion of SL in colloidal suspensions, predominantly NEs, increase the transport of OMC into the SC. It is interesting to note that enhancement is not the result of only one factor such as size, the system's nature, or the kind of enhancer, but of a combination of them. In this sense, SL-NE allowed OMC to penetrate to a great extent. This may be attributed to NE size, the interaction of SL with intercellular lipids, and the deformability of the globules.

In the case of NCs, the interaction of SE within the lipid domain of SC was not enough. It is possible that the polymeric matrix of NCs decreases the release of OMC molecules. Furthermore, the rigidity of the particles is a factor to be taken into account.

CONCLUSIONS

It can be concluded that SEs promote the delivery of OMC across skin predominantly from NE suspensions. Of the two SEs evaluated, SL was significantly more effective than its oleate analogue. The results of this study suggest that the enhanced permeability of OMC from NEs is associated with alterations in the lipoidal pathway of SC generated by the saturated hydrocarbon chain of SL. The interaction of SL with the membrane in the case of NCs does not appear to alter the passage of OMC probably due to their rigid polymeric matrix.

Sucrose laureate (SL) seems particularly promising to enhance the penetration of drugs formulated in NE systems; however, this new approach has to be verified using different active agents, and evaluating the stability and mechanism of action of the colloidal systems.

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REFERENCES

Alberti, I., Kalia, Y. N., Naik, A., Bonny, J. D., & Guy, R. H. (2001). In vivo assessment of enhanced topical delivery of terbinafine to human stratum corneum. *J. Control. Rel.*, *71*(3), 319–327.

Allémann, E., Doelker, E., & Gurny, R. (1993). Drug loaded poly(lactic acid) nanoparticles produced by a reversible salting-out process, purification of an injectable dosage form. *Eur. J. Pharm. Biopharm.*, *39*(1), 13–18.

Ayala, B. H., Quintanar, G. D., Kalia, Y. N., Naik, A., Cornejo-Bravo., J., & Ganem-Quintanar, A. (2003). Effects of sucrose oleate and sucrose laureate on in vivo human stratum corneum permeability. *J. Pharm. Res.*, *20*(8), 1267–1273.

Barry, B. W. (1987). Mode of action of penetration enhancers in human skin. *J. Control. Rel.*, *6*(1), 85–97.

Casal, H. L., & Mantsch, H. H. (1984). Polymorphic phase behavior of phospholipid membranes studied by infrared spectroscopy. *Biochim. Biophys. Acta.*, *779*(84), 381–401.

Cevc, G., Blume, G., Schätzlein, A., Gebauer, D., & Paul, A. (1996). The skin: a pathway for systemic treatment with patches and lipid-based agent carriers. *Adv. Drug Deliver. Rev.*, *18*(3), 349–378.

Cotte, M., Dumas, P., Besnard, M., Tchoreloff, P., & Walter, P. (2004). Synchrotron FT-IR microscopic study of chemical enhancers in transdermal drug delivery: example of fatty acids. *J. Control. Rel.*, *97*(2), 269–281.

Chávez, F., Olvera, B. I., Ganem-Rondero, A., & Quintanar-Guerrero, D. (2002). Liberación de sustancias lipofílicas a partir de nanocápsulas poliméricas. *Rev Soc Quím Méx*, *46*(4), 349–356.

Denet, A-R., Vanbever, R., & Préat, V. (2004). Skin electroporation for transdermal and topical delivery. *Adv. Drug Deliver. Rev.*, *56*(5), 659–674.

Fitzpatrick, D., Corish, J., & Hayes, B. (2004). Modelling skin permeability in risk assessment—the future. *Chemosphere*, *55*(10), 1309–1314.

Fonseca, R., Lange, N., Guy, R., & Lopes Badra Bentley, M. V. (2004). Photodynamic therapy of skin cancer: controlled drug delivery of 5-ALA and its esters. *Adv. Drug Deliver. Rev.*, *56*(1), 77–94.

Grubauer, G., Feingold, K. R., Haris, R. M., & Elias, P. M. (1989). Lipid content and lipid type as determinants of the epidermal permeability barrier. *J. Lipid Res.*, *30*(1), 89–96.

Jadoul, A., Hanchard, C., Thysman, S., & Préat, V. (1995). Quantification and localization of fentanyl and trh delivered by iontophoresis in the skin. *Int. J. Pharm.*, *120*(2), 221–228.

Kalia, Y. N., Naik, A., Garrison, J., & Guy, R. H. (2004). Iontophoretic drug delivery. *Adv. Drug Deliver. Rev.*, *56*(5), 619–658.

Kalia, Y. N., Pirot, F., & Guy, R. H. (1996). Homogeneous transport in a heterogeneous membrane: water diffusion across human stratum corneum in vivo. *Biophys. J.*, *71*(5), 2692–2700.

Knutson, K., Krill, S. L., Lambert, W. J., & Higuchi, W. I. (1987). Physicochemical aspects of transdermal permeation. *J. Control. Rel.*, *6*(1), 59–74.

Lehmann, L., Keipert, S., & Gloor, M. (2001). Effects of microemulsions on the stratum corneum and hydrocortisone penetration. *Eur. J. Pharm. Biopharm.*, *52*(2), 129–136.

Leroux, J-Ch., Allémann, E., Doelker, E., & Gurny, R. (1995). New approach for the preparation of nanoparticles by an emulsification-diffusion method. *Eur. J. Pharm. Biopharm.*, *41*(1), 14–18.

Martini, M-C. (2003). In *Introduction à la dermopharmacie et à la cosmétologie;*, Martini, M-C., Ed.; Paris: Médicales Internationales and Editions TEC&DOC, 118–121.

Mitsubishi-Kasei Foods Co. Japan, Ryoto Sugar Ester Technical Information, 1987.

Olvera-Martínez, B. I., Cázares-Delgadillo, J., Villalobos-García, R., Ganem-Quintanar, A., & Quintanar-Guerrero, D. (2005). Preparation of polymeric nanocapsules containing octyl methoxycinnamate by the emulsification-diffusion technique: Penetration across the stratum corneum. *J. Pharm. Sci.*, *94*(7), 1552–1559.

Potts, R. O., & Francouer, M. L. (1991). The influence of stratum corneum morphology on water permeability. *J. Invest. Dermatol.*, *96*(4), 495–499.

Prausnitz, M. R. (2004). Microneedles for transdermal drug delivery. *Adv. Drug Deliver. Rev.*, *56*(5), 581–587.

Quintanar-Guerrero, D., Allémann, E., Doelker, E., & Fessi, H. (1998). Preparation and characterization or nanocapsules from preformed polymers by a new process based on emulsification-diffusion technique. *Pharm. Res.*, *15*(7), 1056–1062.

Ramachandran, Ch., & Fleisher, D. (2000). Transdermal delivery of drugs for the treatment of bone diseases. *Adv. Drug Deliver. Rev.*, *42*(3), 197–223.

Suhonen, T. M., Bowstra, J. A., & Urtti, A. (1999). Chemical enhancement of percutaneous absorption in relation to stratum corneum structural alterations. *J. Control. Rel.*, *59*(2), 149–161.

Tambini, D., Bobin, M. F., & Martín, M. C. (1993). Etude physicochimique des sucroesters. Essais de valorisation. *Pharm. Belg.*, *48*, 19–26.

Thevenin, M. A., Grossiord, J. L., & Poelman, M. C. (1996). Sucrose esters/cosurfactant microemulsion systems for transdermal delivery: assessment of biocontinuous structures. *Int. J. Pharm.*, *137*(2), 177–186.

Williams, A. C., & Barry, B. W. (2004). Penetration enhancers. *Adv. Drug Deliver. Rev.*, *56*(5), 603–618.