Low frequency sonophoresis: influence of solid lipid nanoparticles in skin LTRs distribution

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ABSTRACT SUMMARY

Low frequency ultrasound (US) is used to enhance drugs transdermal penetration. The improvement of skin permeability promoted by US is generally related to the formation of heterogeneously distributed localized transport regions (LTRs). Strategies for increasing the number and homogenize the distribution of the LTRs can further improve the skin permeability mediated by US. The aim of this work was to evaluate the influence of solid lipid nanoparticles dispersions (SLN) in LTRs size and distribution. Dermatomed porcine skins were treated by US using SLN or sodium lauril sulfate (SLS) as the coupling medium, until they reached predetermined electrical current threshold of 100 ± 12 µA. LTRs were macroscopically visualized by addition of allura red dye in the coupling medium. When SLS was used a single large LTR was observed, whereas SLNs addition leads to a heterogeneous pattern of several LTRs. The influence of this distribution in model drug permeation is still to be shown.

INTRODUCTION

In recent decades, US has been shown to increase drug delivery through the skin. When pre-treated with US, skin shows two distinct regions, the very permeable ones, known as LTR and the surrounding regions, known as non-LTRs. LTRs are 80 - fold more permeable than non-LTRs. It is known that acoustic cavitation events caused by US are the source of skin perturbation. Because LTRs formation are correlated with the cavitation events, it is believed that the addition of particles in the coupling medium can make a change in the collapse mode of the cavitation bubbles and, therefore, lead to the formation of more evenly distributed LTRs in skin. Based on the above, we assume that the addition of SLN in coupling medium can modify skin LTRs distribution. SLN are nanoparticles composed by solid lipids at corporal and room temperature stabilized with an emulsifying layer in an aqueous dispersion. Both lipids and surfactants that compose these nanoparticles are generally recognized as safe (GRAS). During the US exposure, besides changes the collapse mode of the cavitation bubbles, SLNs components can be incorporated into the skin, changing its permeability and also the routes that a drug, post-administered, has to penetrate.

Therefore, the aim of this first study was to verify the influence of SLN aqueous dispersion used as coupling medium in LTRs distribution. SLN were designed to have components and zeta potential that can positively influence in further drug delivery to the skin. In this way, cationic SLNs containing monoolein, a lipid known for its surface properties and skin permeation enhancer ability, was prepared and characterized before the US studies.

EXPERIMENTAL METHODS

SLN preparation: SLN were prepared by the microemulsion method. Stearic acid, cetyltrimethylammonium bromide (CTAB): monoolein (2:1) and deionized water (40/20/40) were mixture at 90 ± 2°C until the formation of a thermodynamically stable microemulsion. The microemulsion was added to cold water (2-5°C) at the ration of 1:20 (microemulsion:water) under vigorous stirring at 20,000 rpm (IKA-T25 Ultra-turrax, Germany) for 10 minutes to obtain the SLN dispersion. Then, SLN aqueous dispersion was subjected to high pressure homogenization (EmulsiFlex – C3, Germany) at 500 bars for 10 minutes.

US treatment of the skin: dermatomed porcine skin (700 µm thickness) was mounted in a Franz diffusion cell. US was applied to the skin samples using a VCX 500 and a coupler probe (Sonics & Materials, Newtown, CT) at the frequency of 20 kHz, intensity of 7 W/cm², pulse length of 5 s on, 5 s off, and distance between the probe tip and the skin of 3 mm. The US intensity was calibrated using calorimetry. Two coupling medium were used to treat the skin: (i) SLS 1.0 % in PBS and (ii) SLN aqueous dispersion. Allura red at 0.025% was also added to both coupling medium. After each minute of US application, the coupling medium was replaced to minimize thermal effects. Samples were treated until they reached predetermined electrical current threshold of 100 ± 12 µA. Upon completion
of the US treatment, skin samples were rinsed thoroughly with PBS to remove all the formulation excess from the skin surface and 0.025% allura red in PBS was added to the donor compartment of the diffusion cell containing the US pretreated skin. After 1 h, the skins were imaged using a Panasonic Lumix, 16 Mega pixels, and auto macro setting, at a distance of approximately 20 cm above the skin surface.

RESULTS AND DISCUSSION

The SLN developed showed monomodal distribution with mean particle size of 170.0 ± 14.8 nm, polydispersity index (PdI) of 0.27 ± 0.10 and zeta potential of +60.6 ± 0.6 mV. It has been shown that positively charged nanoparticles seems to penetrate better the skin when US is applied and, in addition, monoolein enhance skin permeation, which can favor drugs deeper skin penetration.

The images obtained by atomic force microscopy (AFM) showed that SLN presented spherical form and particle sizes around 150 nm, corroborating with photons correlation spectroscopy results.

Figure 1: A) topographic image , B) phase image

Figure 2 shows skin LTRs distribution after skin US treatment with SLS and with the SLN. It can be seen that treating the skin until the resistivity of ~1 kΩ cm², which is the resistivity reported in a US clinical study showed very different LTRs distribution on skin surface. When SLS, that is considered a “gold standard” in US treatment, was used, a single large LTR was observed. On the other hand, when SLN dispersion was used as the coupling medium, multiple very small LTRs were noticed distributed heterogeneously over the entire skin surface. This pattern is in fact very similar to that obtained with SLS when the skin is treated until smaller resistivity of the 225 275 and 335 µA.

CONCLUSION

Our studies demonstrated that it is possible to obtain cationic SLN containing monoolein with nanometric size and reasonable PdI. The use of SLN as coupling medium changed size and distribution of the LTRs formed when US was applied. Further permeation experiments using a model drug loaded in SLN will be conducted to determine the influence of this change in drug penetration.

REFERENCES


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