Effects of microbubble size on ultrasound-induced transdermal delivery of high-molecular-weight drugs

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ABSTRACT

Transdermal drug delivery (TDD) can be assisted and enhanced by sonophoresis with ultrasound (US)-aided microbubbles (MBs). The present study determined the different effects of various sizes of MBs that underwent US exposure to enhance the transdermal delivery of high-molecular-weight drugs. The effects of US-mediated MBs of different sizes (1.4, 2.1, and 3.5 μm) and ascorbyl tetraisopalmitate (VC-IP, a cosmetic ingredient for skin lightening) on enhancing skin transdermal delivery were demonstrated both in vitro and in vivo. The results indicated that the US power intensities of a 3 W/cm^2 penetration depth in the US group combined with 3.5-μm MBs and penetrating VC-IP (U+3.5) were 34% and 14% higher than those in the US group combined with 1.4-μm MBs and penetrating VC-IP (U+1.4) and US combined with 2.1-μm MBs and penetrating VC-IP (U+2.1), respectively, for the agarose phantoms; the increases for pigskin were 37% and 19%. The VC-IP concentrations in the U+3.5 group were 23% and 10% higher than those in the U+1.4 and U+2.1 groups, respectively. The whitening effect (luminosity index) of mouse skin in the U+3.5 group significantly increased by 28% after 1 week and 34% after 2 weeks, while it tended to stabilize after 3 weeks (45%), in C57BL/6J mice over a 4-week experimental period.

Specific Aims

With the aim of improving the efficiency VC-IP TDD, the present study evaluated the impact of combining US with MBs of different sizes on the efficiency of TDD for large molecules both in vitro and in vivo.

Materials and Methods

Preparation of albumin MBs of different sizes

1.4-μm, 2.1-μm, and 3.5-μm MBs were generated by sonication in 10 ml of solution mixing the 1.32% albumin with 5%, 20%, and 45% dextrose, respectively, in perfluorocarbon gas in PBS using a sonicator (200 W; Branson Ultrasonics, Danbury, CT, USA) for 2 min.

Measurements of penetration depth in agarose phantoms

Figure 1 shows a schematic diagram of the system setup. The model drug, Evans blue was dissolved in 10 ml of PBS and then stirred for 1 h at 4°C. The round area of each phantom was loaded with Evans blue or MBs. The probe of the sonoporation gene transfection system was positioned 3 mm from the top of the phantom.

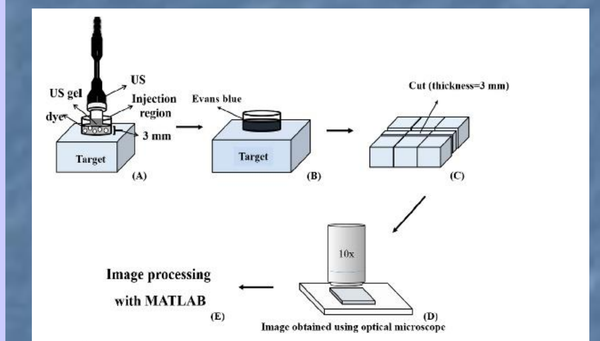


Fig 1. Schematic diagram of the system setup for measuring the penetration depth in agarose phantoms.

Measurements of penetration depth in pigskin

The treatment area on a 2-mm-thick sample of pigskin was sonicated by the 1-MHz US transducer of the sonoporation system successively at power densities of 1 W for 1 min, 2 W for 1 min, and 3 W/cm^2 for 1 min after adding 500 μl of the three different sizes of MBs. The Evans blue solution was then applied to the area on the pigskin and left there for 15 min.

In vitro skin penetration by VC-IP

The dorsal hairs of C57BL/6J mice were shaved with clippers and a full-thickness skin specimen was surgically removed, and cut into square pieces (2 × 2 cm). The in vitro skin penetration was tested using static Franz diffusion cells over an area of 2.14 cm^2.

Animal treatments

The animals were acclimatized for 7 days prior to the experiment. The hair was then removed and the skin color was measured using a Chroma Meter before exposure to UVB irradiation to induce hyperpigmentation, after which the skin color was measured again. The US was applied at 1, 2 or 3 W/cm^2 for 1 min (except when stated otherwise, the power density was 3 W/cm^2), and 3% VC-IP was used in all cases. The change in skin color induced by each of the treatments was assessed at predetermined times using the Chroma Meter.

Histochemistry

Skin tissue samples (8 mm × 8 mm) were cut and stored in a 10% formalin solution. H & E staining was applied, and the samples were analyzed by an expert histologist.

Results

Evaluations of the penetration depth in pigskin

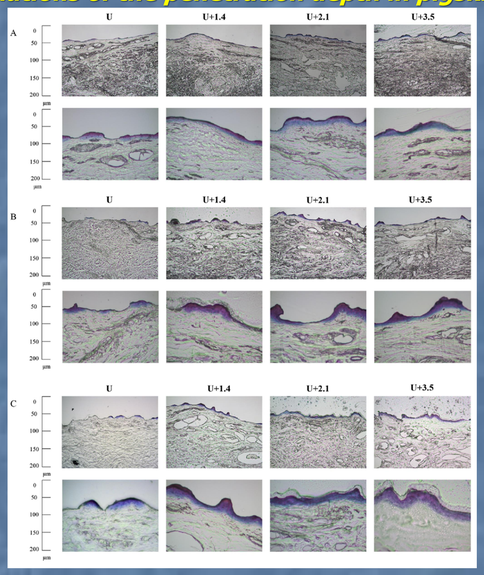


Figure 2: Representative histological sections of pigskin in groups U, U+1.4, U+2.1, and U+3.5 after US treatment at 1 W/cm2 (A), 2 W/cm2 (B), and 3 W/cm2 (C).

In vitro skin penetration by VC-IP

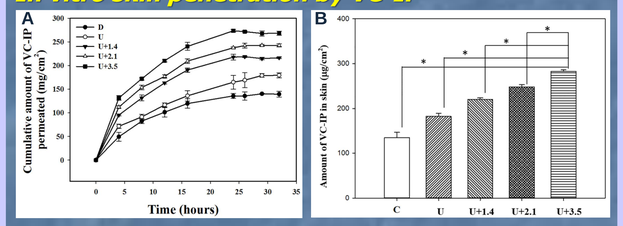


Figure 3: (A) VC-IP concentrations in gps C, U+1.4, U+2.1, and U+3.5 for percutaneous penetration over 32 hours as analyzed using the UV/visual spectrophotometer. (B) The amount of VC-IP deposited in the skin appeared to be higher in gp U than in gp C, and significantly so in gps U, U+1.4, U+2.1, and U+3.5.

Animal treatments

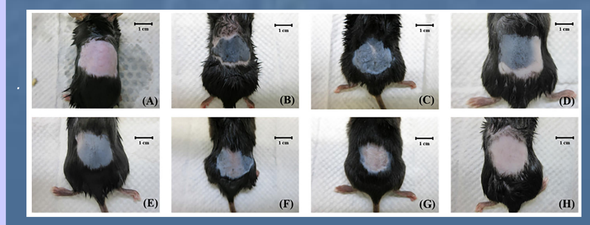


Figure 4: Photographs of mouse skin in a completely untreated animal (A), after UVB irradiation (B), and in groups C (C), D (D), U (E), U+1.4, (F), U+2.1 (G), and U+3.5 (H) at week 4.

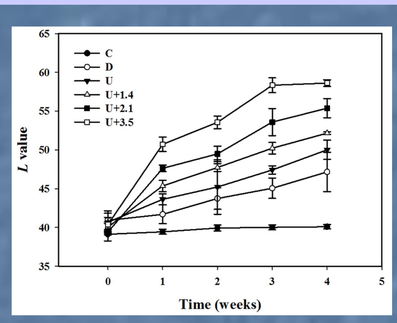


Figure 5: Skin-whitening effects of VC-IP on UV-induced hyper-pigmentation in groups C, D, U, U+1.4, U+2.1, and U+3.5 over 4 weeks.

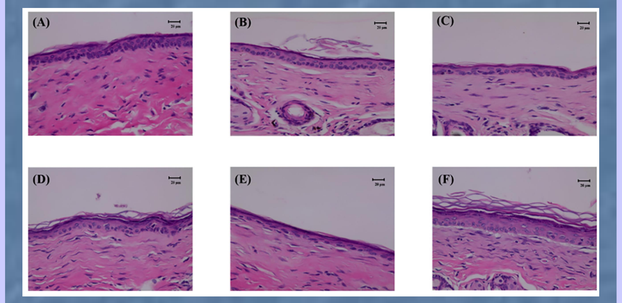


Figure 6: Histology images for groups C, D, U, U+1.4, U+2.1, and U+3.5 at 4 weeks.

Conclusion

In conclusion, to the best of our knowledge, this is the first study to demonstrate that US combined with MBs of different sizes can produce different degrees of skin permeability to enhance the delivery of high-molecular-weight drugs like VC-IP for skin brightness without damaging the skin.

Acknowledgements

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