molecules

MDPI

SUPPLEMENTARY MATERIALS

Article

Coptis chinensis Franch directly inhibits proteolytic activation of kallikrein 5 and cathelicidin associated with rosacea in epidermal keratinocytes.

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Supplementary materials

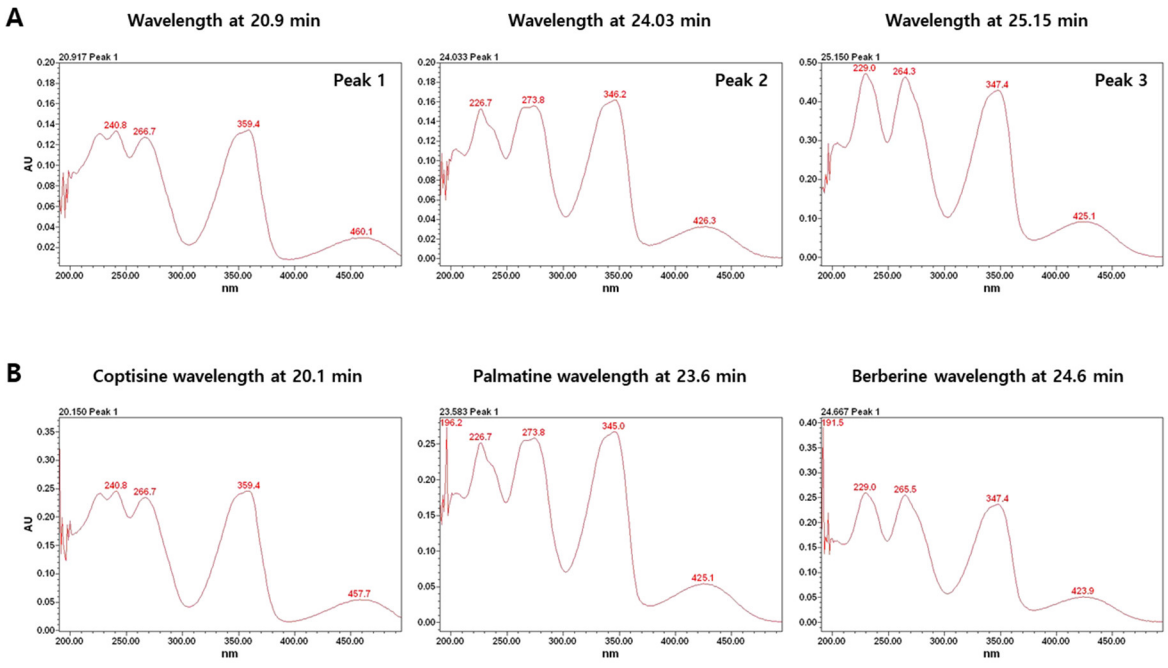

Figure S1. Peak identification of CCE (A) by comparing the retention times and UV spectra for individual standards (B).

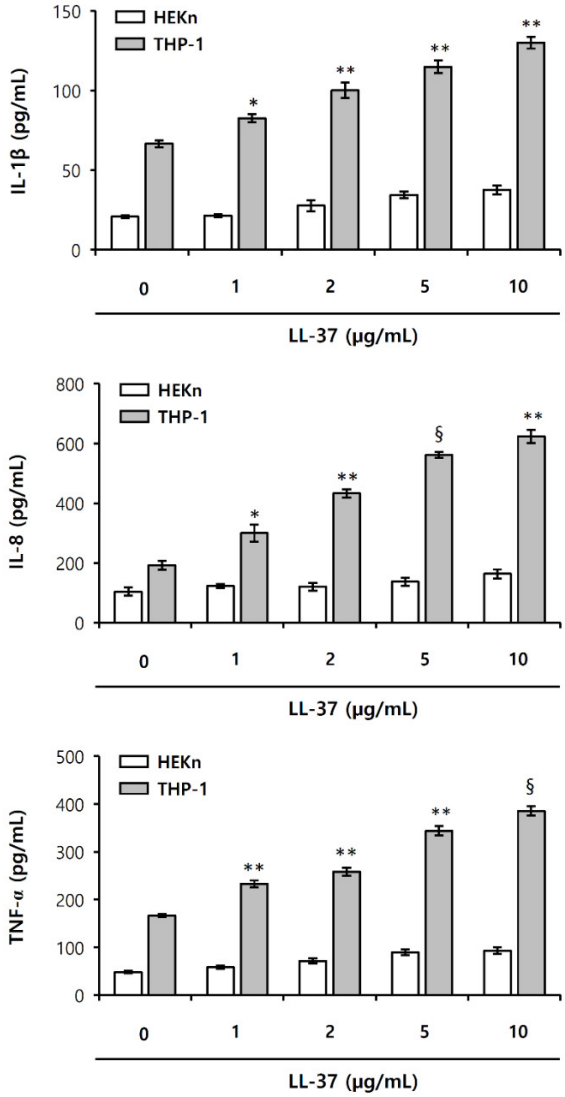

Figure S2. LL-37 induces pro-inflammatory cytokines in THP-1. THP-1 cells were seeded in 24-well plates at 2 × 10^5 cells/well, and differentiated into macrophages using 100 nM PMA for 24 h. The culture media was replaced with fresh DMEM containing 1% FBS and 1% penicillin/streptomycin, and then treated with 1, 2, 5, or 10 μg/mL LL-37 for 48 h. THP-1 culture media was collected, and secreted pro-inflammatory cytokines (IL-1β, IL-8, and TNF-α) were determined by ELISA. HEKn (passage 3) at 90% confluence was treated with 1, 2, 5, or 10 μg/mL LL-37 for 72 h. HEKn culture media was collected, and secreted pro-inflammatory cytokines (IL-1β, IL-8, and TNF-α) were determined by ELISA. *$P < 0.05$ *vs*. LL-37-unteated control; **$P < 0.01$ *vs*. LL-37-unteated control; §$P < 0.001$ *vs*. LL-37-unteated control.

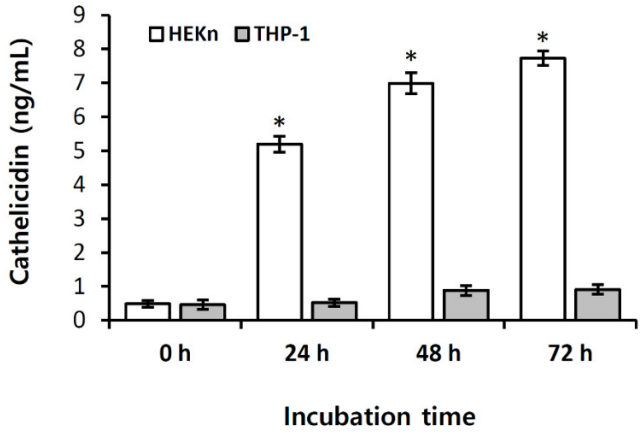

Figure S3. VD_3 induces cathelicidin expression in HEKn, but not THP-1. HEKn (passage 3) at 90% confluence was treated with 200 nM VD_3 for 0, 24, 48, or 72 h. HEKn culture media was collected, and secreted cathelicidins were determined by ELISA. THP-1 cells were seeded in 24-well plates at 2×10^5 cells/well, and differentiated into macrophages using 100 nM PMA for 24 h. The culture media was replaced with fresh DMEM containing 1 % FBS and 1% penicillin/streptomycin. Subsequently treated with 200 nM VD_3 for 0, 24, 48, or 72 h. THP-1 culture media was collected, and secreted cathelicidins were determined by ELISA. *$P < 0.001$ *vs*. 0 h.

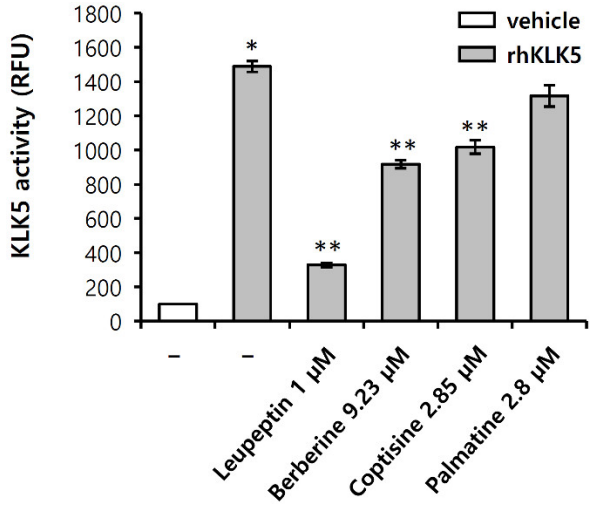

Figure S4. The major constituents of CCE inhibits KLK5 protease activity. KLK5 activity was measure in relative fluorescence units (RFU), using fluorogenic peptide substrate sensitive to KLK5. *$P < 0.001$ *vs*. vehicle-treated control; **$P < 0.01$ *vs*. rhKLK5-treated control; DMSO vehicle control (vehicle).

Full-length Western Blot image of Figure 4