

Figure S1. DNA fragmentation in EGF- or LL-37-treated OTSCC HSC-3 cells. DNA fragmentation in control, EGF- or LL-37-treated HSC-3 cells was determined after 48 h using the *In situ* Cell Death Detection Kit. The nuclei were stained with DAPI. Scale bar, 50 μm . EGF, epidermal growth factor; OTSCC, oral tongue squamous cell carcinoma; LL-37, antimicrobial peptide leucine-leucine-37.

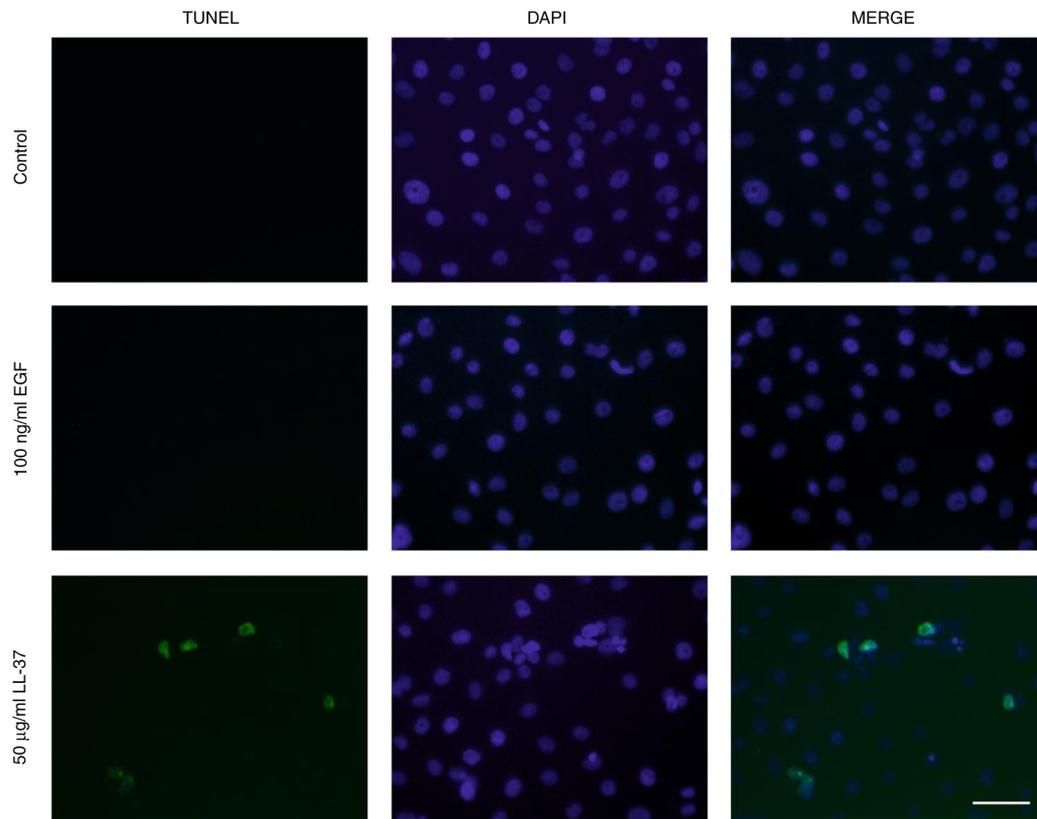


Figure S2. Immunofluorescence staining of EGFR in EGF- or LL-37-treated OTSCC HSC-3 cells. Cells were starved for 24 h and stimulated with the indicated doses of EGF or LL-37 for 1 h. The nuclei were stained with DAPI. Scale bar, 50 μ m. EGFR, epidermal growth factor receptor; EGF, epidermal growth factor; OTSCC, oral tongue squamous cell carcinoma; LL-37, antimicrobial peptide leucine-leucine-37.

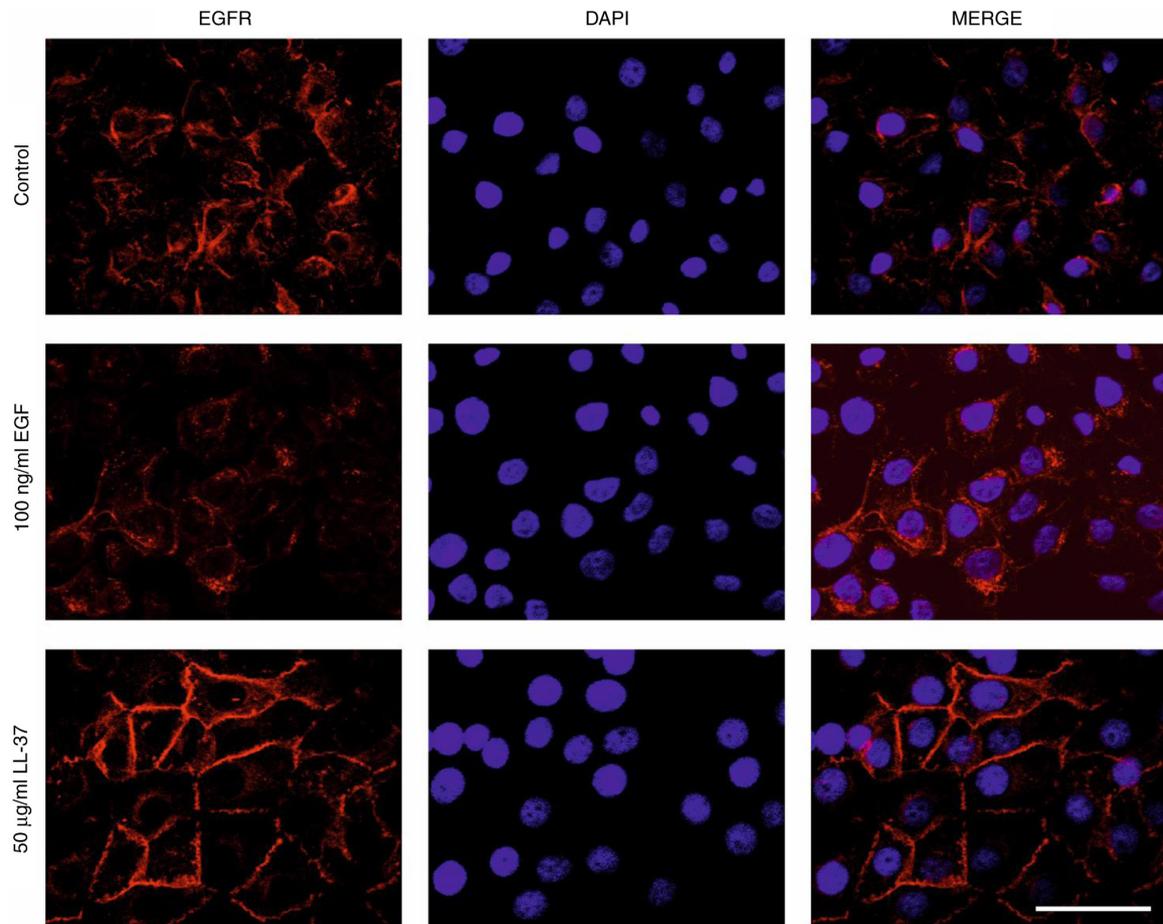


Figure S3. Immunoblot analysis of EMT markers in EGF- or LL-37-treated OTSCC cell lines and IHGK cells. Cells were treated with the indicated doses of EGF or LL-37 for 48 h. Cell homogenates (30 μg of soluble protein) were separated on SDS-PAGE gels. The amount of the epithelial marker E-cadherin (E-cad) and the mesenchymal marker vimentin was analyzed with immunoblots in (A) HSC-3, (C) SCC-25, (E) SAS and (G) IHGK cells. In A, C, E and G, representative immunoblots are shown. β -actin was used as a control to normalize the quantities of proteins in (B) HSC-3, (D) SCC-25, (F) SAS and (H) IHGK cells using Fiji software. The results represent average \pm SD of two independent experiments, separated three times on SDS-PAGE gels. P-values were calculated with the independent-samples Kruskal-Wallis test and significance was adjusted by the Bonferroni correction for multiple tests. * $P < 0.05$, compared with the control. EMT, epithelial-mesenchymal transition; EGF, epidermal growth factor; OTSCC, oral tongue squamous cell carcinoma; LL-37, antimicrobial peptide leucine-leucine-37.

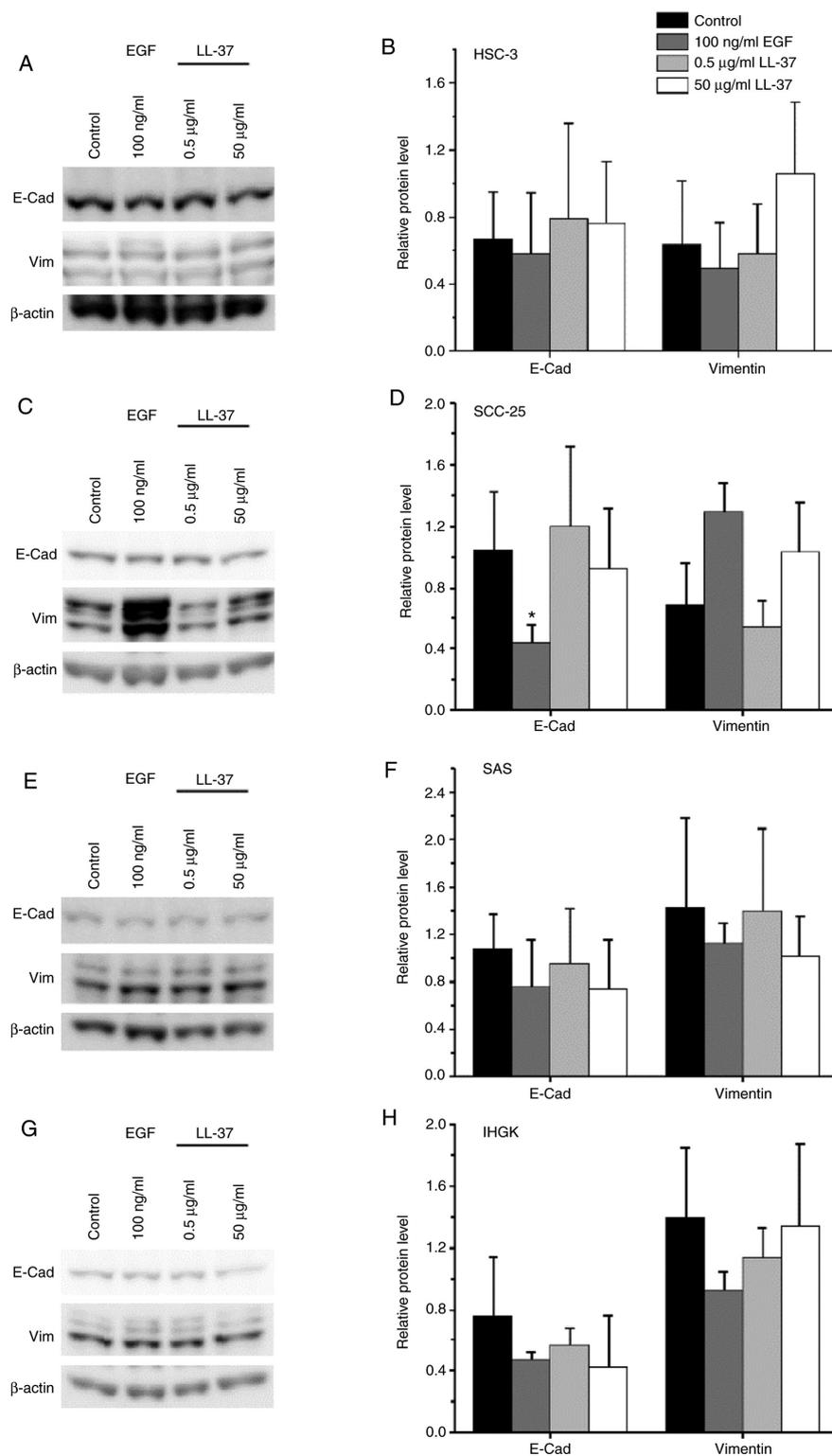


Figure S4. Zymography of EGF- or LL-37-treated OTSCC cell lines and IHGK cells. Cells were treated with the indicated doses of EGF or LL-37 for 24 h. In zymography, conditioned concentrated media of (A) HSC-3, (C) SCC-25, (E) SAS and (G) IHGK cells were separated on SDS-PAGE casted in the presence of fluorescently labelled gelatin. (A) Purified gelatinase standards are shown on the left: pro-MMP2 (72 kDa), active MMP2 (62 kDa), pro-MMP9 (92 kDa) and active MMP9 (82 kDa). In A, C, E and G, a representative zymogram gel is shown. The amount of pro-MMP2 and MMP9, and active MMP2 in (B) HSC-3, (D) SCC-25, (F) SAS and (H) IHGK zymogram gels was quantified with Fiji software, and intensities were normalized to the cellular soluble protein concentration. The results represent the average \pm SD of two separate sample sets each analyzed three times. P-values were calculated with the independent-samples Kruskal-Wallis test and significance was adjusted by the Bonferroni correction for multiple tests. * $P < 0.05$, compared with the control. MMP, matrix metalloproteinase; EGF, epidermal growth factor; OTSCC, oral tongue squamous cell carcinoma; LL-37, antimicrobial peptide leucine-leucine-37.

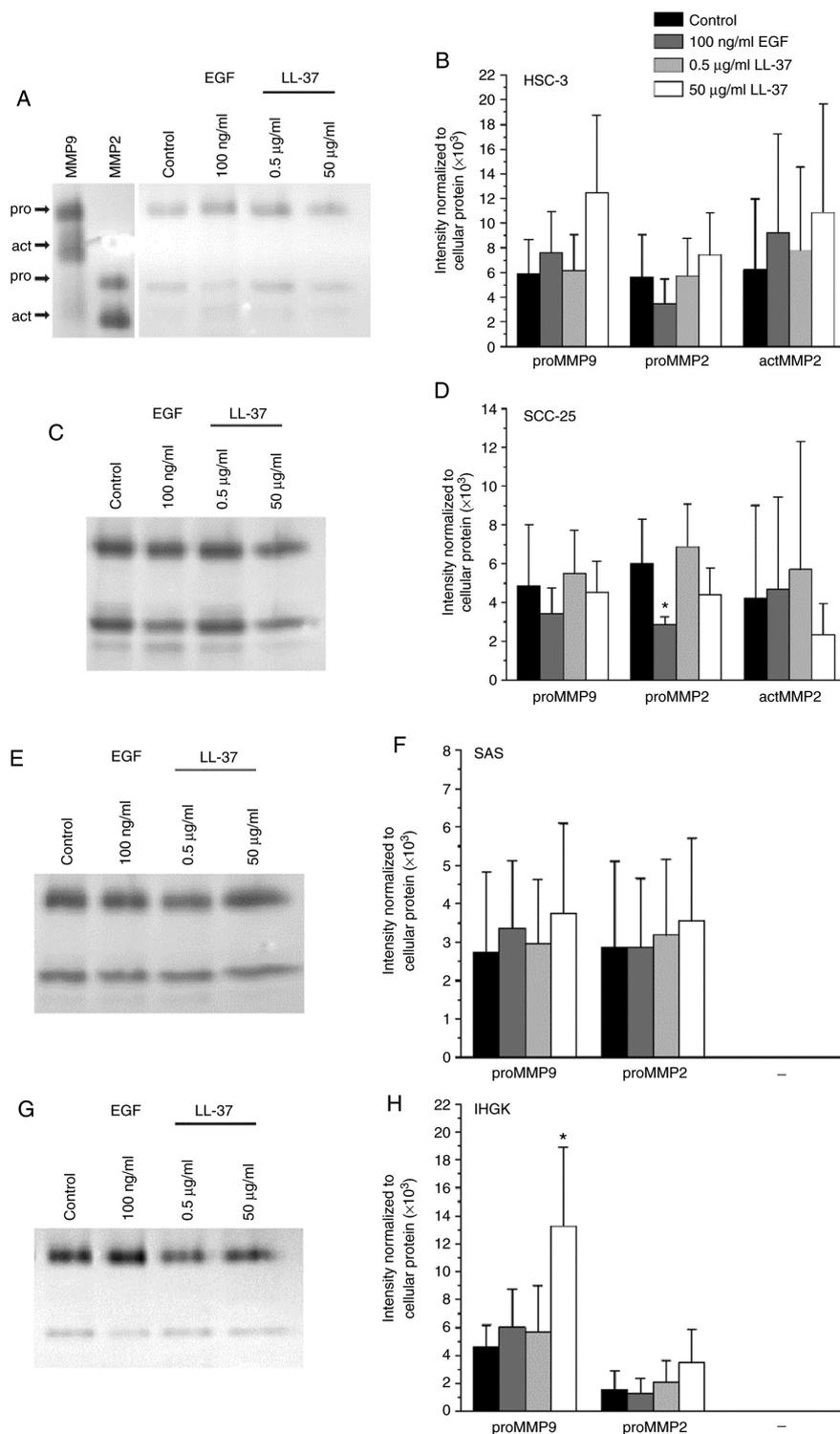


Figure S5. Kaplan-Meier survival curves. Kaplan-Meier cumulative curves for (A) overall, (B) disease-specific and (C) disease-free survival of patients with OTSCC according to expression of hCAP18/LL-37. No significant difference was present in survival times between the high and low hCAP18/LL-37 expression groups ($P > 0.05$). hCAP18/LL-37, human host defense cationic antimicrobial peptide-18/antimicrobial peptide leucine-leucine-37; OTSCC, oral tongue squamous cell carcinoma.

