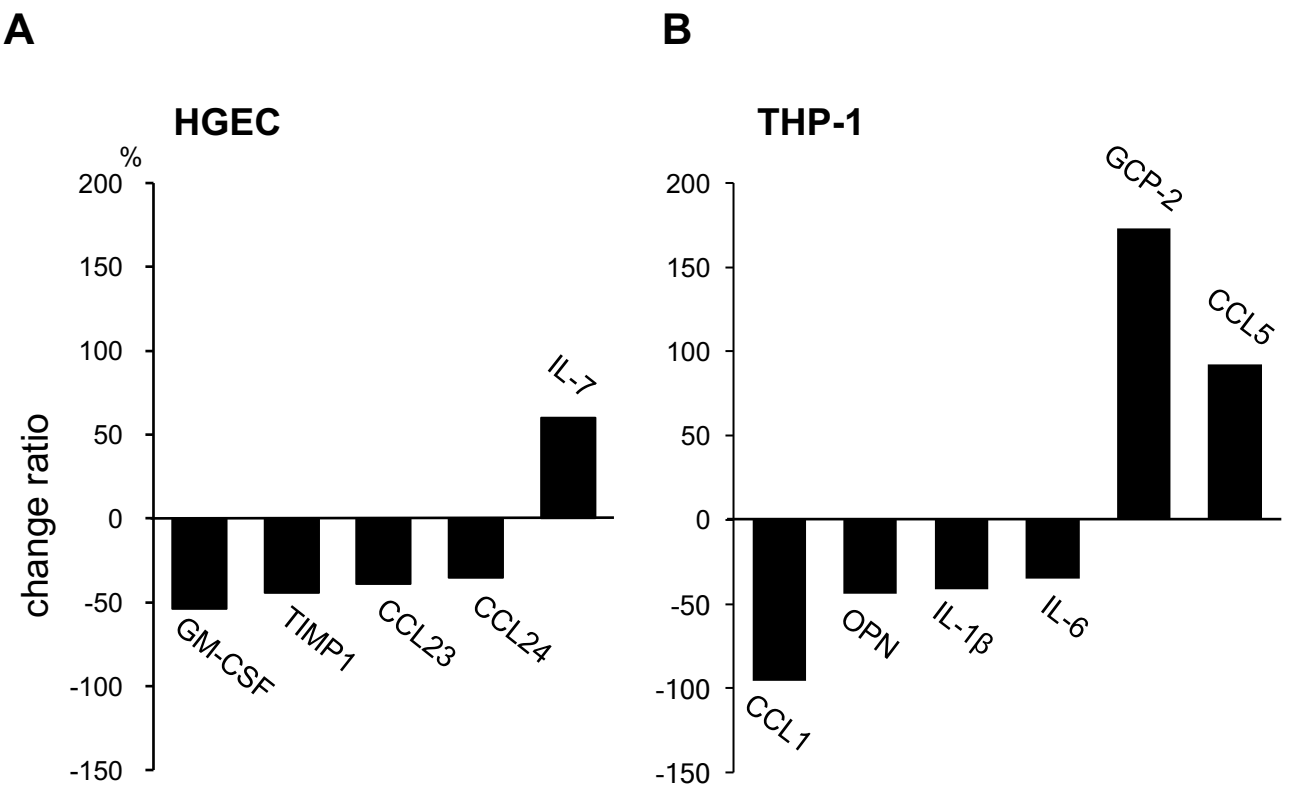

Fig. S1 Cytokine array analysis.

HGECs and THP-1 were cultured for 48 hours with TNF-α (10 ng/ml) and anti-HMGB1 antibody (10 μg/ml) or only with TNF-α (control cells). Cytokine array analysis was performed using the Human Cytokine Antibody Array G5 (Raybiotech, Norcross, CA) according to the manufacturer's instructions. The antibody array allows for the detection of eighty kinds of cytokines, chemokines, growth factors, proteases, soluble receptors, and other proteins from the conditioned media. The signal intensity was measured at 532 nm using a GenePix4000B (Molecular Devices, Sunnyvale, CA). Five proteins in HGECs and seven proteins in THP-1 were selected. These were expressed by more than 1.5-fold or less than 0.65-fold in anti-HMGB1 antibody-treated cells compared to in control cells, in accordance with the manufacturer's instructions; The fold change rate (%) is shown on the y-axis (+, upregulation; –, downregulation). Abbreviations: tissue inhibitor of metalloproteinase (TIMP), C-C motif ligand (CCL), Osteopontin (OPN), granulocyte chemotactic protein (GCP).

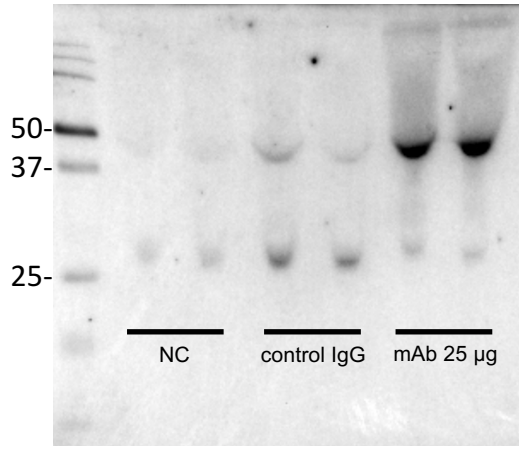

Fig. S2 Anti-HMGB1 antibody detection in mice blood serum.
Western blot analysis of anti-HMGB1 antibody level in mice blood serum. 24 hours after IP injection of the antibody, we collected blood from each mouse and extracted blood serum. Rat IgG antibody (Ambion) was used for primary antibody. NC: negative control (without antibody administration), control IgG: 10 µg/mice, mAb 25 µg: anti-HMGB1 antibody 25 µg/mice. N=2.

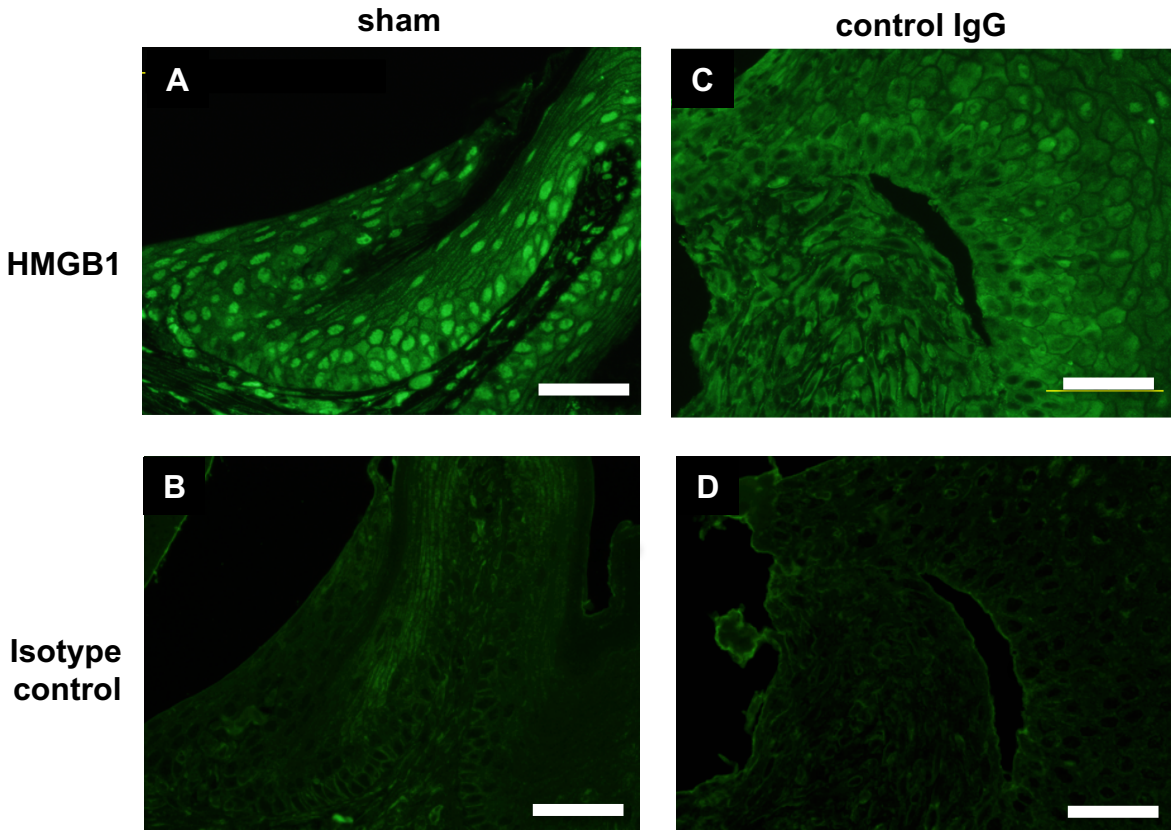

Fig. S3 Negative control for HMGB1 immunofluorescence.

Immunofluorescent analysis of HMGB1 in gingival epithelium of sham-treated mice (A, B) and periodontitis mice (control IgG administration) (C, D) at day 7. Negative control sections (B, D) were incubated with isotype control IgG antibody in place of primary antibody to HMGB1. The fluorescence images were captured under the same condition. No signal was observed with the control antibody. Scale bar: 20.0 μm.

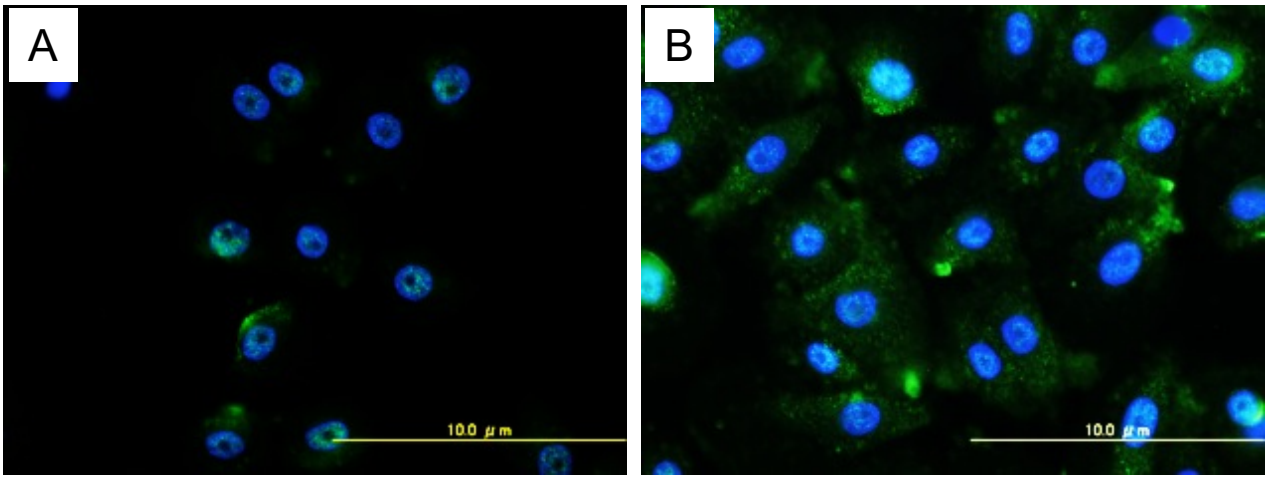

Fig. S4 Intracellular HMGB1 localization in HGEC after TNF-α stimulation.

Immunofluorescent analysis of HMGB1 in HGEC (A) before and (B) after 6 h with 10 ng/ml TNF-α stimulation. Scale bar: 10.0 μm. We represented the typical images from three times expreiment.