Supplemental Fig. S1. Tamm-Horsfall protein-1 (THP-1) activation and effects of irisin on lipopolysaccharide (LPS)-induced RAW264.7.
(A) Phorbol 12-myristate 13-acetate (PMA)-differentiated THP-1 macrophages were polarized into inflammatory M1 phenotype with LPS 10 ng/mL. Real-time polymerase chain reaction (RT-PCR) analysis of relative mRNA expression of tumor necrosis factor alpha (TNF-α), interleukin 6 (IL-6), IL-1β normalized to glyceraldehyde-3-phosphate dehydrogenase (GAPDH) in THP-1 cells after 12 hours treatment. (B-F) RAW264.7 macrophages were treated with LPS 100 ng/mL and irisin 0 to 50 nM for 6 or 24 hours. (B) Nitrite concentration in the supernatant of RAW264.7 cells after 24 hours treatment. (C, D) IL-6 (C) and TNF-α (D) concentration in the supernatant of RAW264.7 cells after 6 hours treatment. (E, F) RT-PCR analysis of relative mRNA expression of TNF-α and IL-6 normalized to GAPDH in RAW264.7 cells after 6 hours treatment. (F) Xanthine oxidase inhibition rate of superoxide dismutase in the supernatant of RAW264.7 cells after 24 hours treatment. All data are presented as mean±standard error of the mean (n=2). ELISA, enzyme-linked immunosorbent assay; SOD, superoxide dismutase.