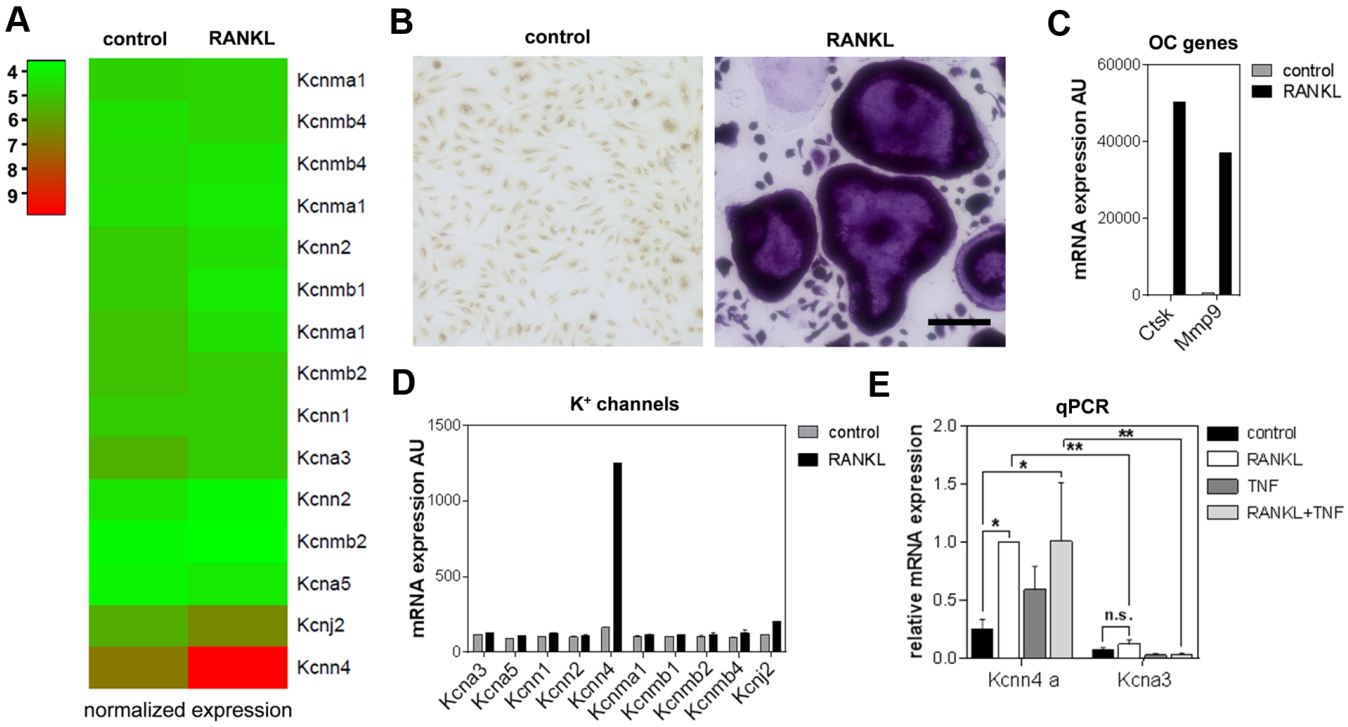
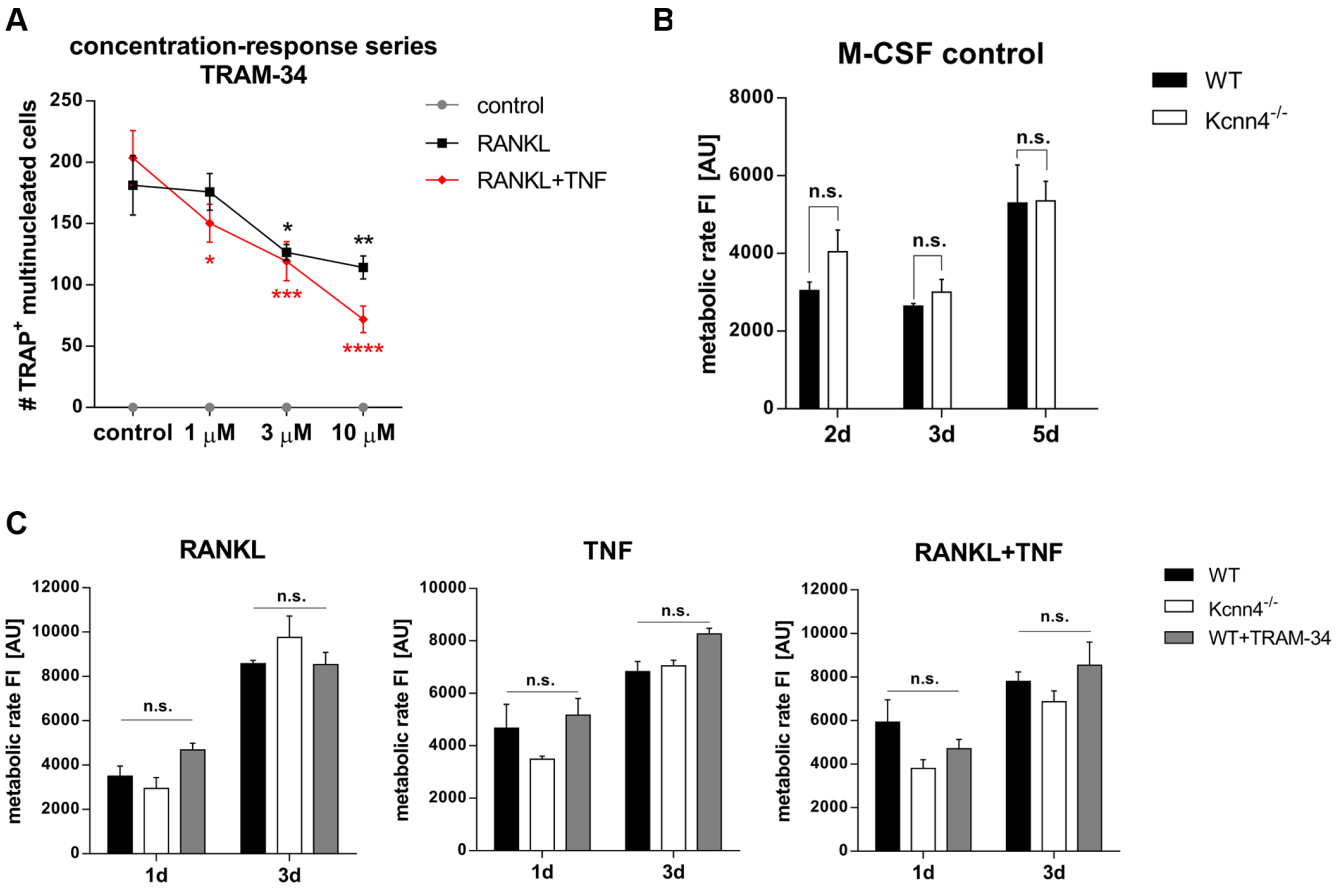
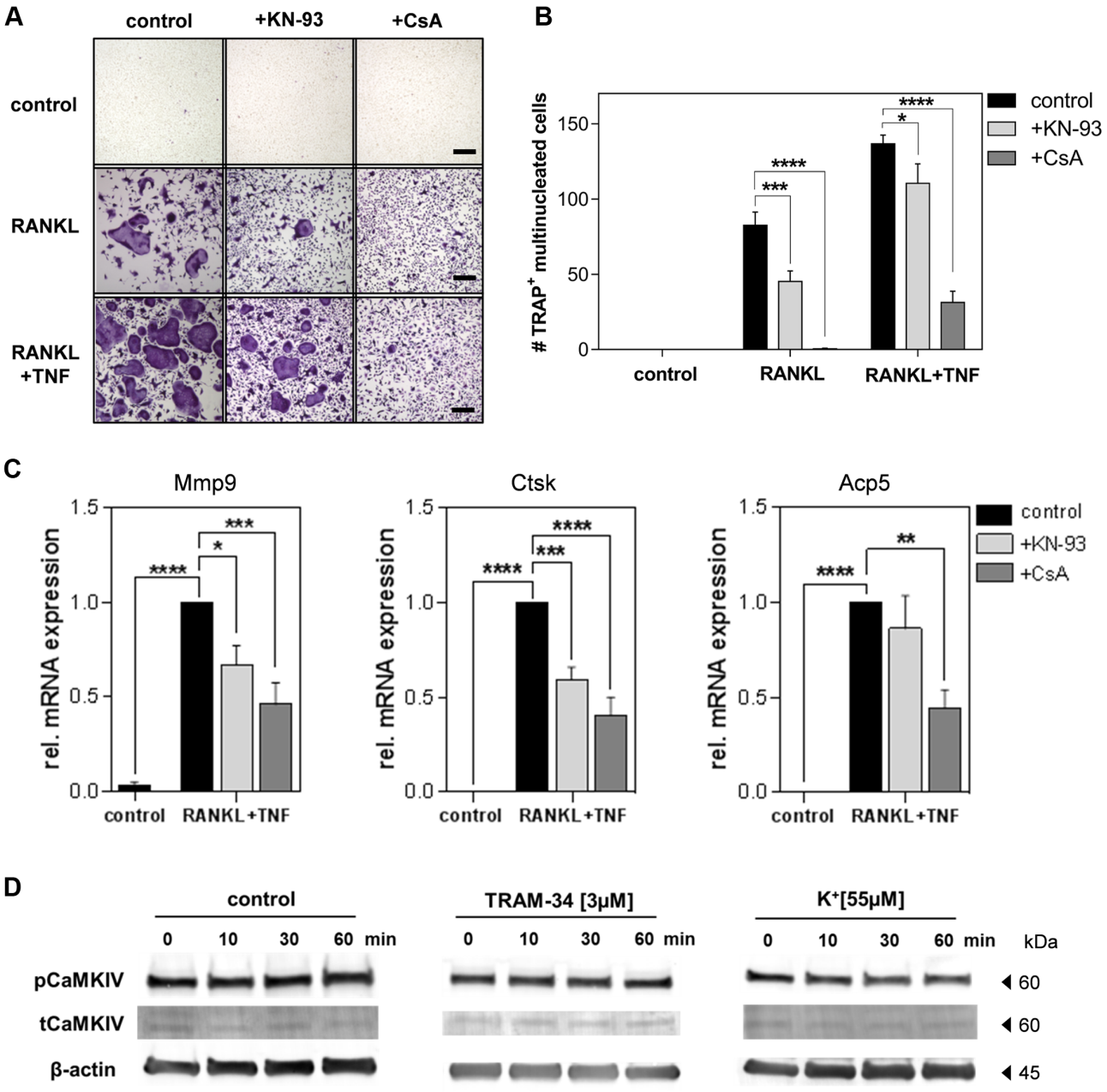


Supplemental figure 1: *Kcnn4* is up-regulated during osteoclastogenesis. A) Hierarchical cluster analysis of RNA microarray data obtained from mouse BMMs before (control) and after treatment with RANKL (RANKL) displaying the differential gene expression patterns of a selected panel of K^+ channels. B) Osteoclast formation was confirmed by cytochemical staining of TRAP (purple) in multinucleated giant cells (scale bar = 100µM) and C) expression of osteoclast-specific genes (OC genes) *Ctsk* and *Mmp9* in arbitrary units (AU). D) Expression of K^+ channels obtained from microarray raw data in arbitrary units (AU). E) qPCR analysis comparing KCa3.1 (*Kcnn4 a*) and Kv1.3 (*Kcna3*) mRNA expression in BMMs cultivated in respective conditions for three days; one-way ANOVA followed by post-hoc multiple comparison test was performed (*N=3)*; **=P<0.05, **=P<0.01.*



Supplemental figure 2: Concentration-response, cytotoxicity and viability studies for TRAM-34 and *Kcnn4* knockout on macrophages and osteoclasts. A) Concentration-response curves for BMMs treated with indicated amounts of TRAM-34 prior to stimulation with RANKL or RANKL+TNF for three days; graph indicates formation of multinucleated cells (>3 nuclei); B) Cell viability/proliferation assays obtained from Alamar blue staining of BMMs isolated from WT and *Kcnn4*$^{-/-}$ mice, two, three and five days in culture medium; C) Alamar blue assays of WT control-treated, *Kcnn4*$^{-/-}$, and BMMs pretreated with 10 μM TRAM-34 in the culture medium; shown are time points one day and three days after RANKL, TNF or RANKL+TNF stimulation (*N=3)*, two-way ANOVA (*P<0.0001* for both factors) with multiple comparison test was performed, *=*P<0.05, **=P<0.01, ***=P<0.001, ****=P<0.0001*.



Supplemental figure 3: Inhibition of CaMKIV or CaN drastically decreases osteoclast formation and transcription of osteoclast-specific genes in the presence of TNF. A) Representative images of cytochemical TRAP staining of murine WT control BMMs (control), BMMs treated with 1 µM KN-93 (+KN-93) (to inhibit CaMKIV), or 100 nM cyclosporine A (+CsA) (to inhibit CaN) cultured in indicated conditions (*N=3,* scale bar = 100 µM). B) Numbers of multinucleated $TRAP^+$ cells (purple) (>3 nuclei) obtained from A) (*N=3)*; C) qPCR analysis showing normalized mRNA expression of osteoclast-specific genes *(N=4)*; shown are

means ± SEM, one-way ANOVA followed by multiple comparison test was performed, **=P<0.05*, ***=P<0.01, ***=P<0.001, ****=P<0.0001*. D) Western blot studies of BMMs stimulated with 100 ng/mL RANKL for indicated times showing phosphorylated (pCaMKIV) and total CaMKIV (tCaMKIV) versus β-actin; left: control stimulated, middle: pre-treated with 3 μM TRAM-34, right: in culture medium with increased K^+ concentration; blots are representative for two independent experiments.