## **Supplemental Data**



Supplemental Figure 1. Generation of osteoclast-specific cdc42<sup>-/-</sup> mice and cdc42GAP<sup>-/-</sup> radiation chimeras. (A) PCR genotyping of WT and cdc42<sup> $\Delta$ OC/ $\Delta$ OC</sub> BMMs (day 0), pre-osteoclasts (day 3) and mature osteoclasts (day 5). (B) Western blots of cdc42 protein expression in WT and cdc42<sup> $\Delta$ OC/ $\Delta$ OC</sub> BMMs (day 0), pre-osteoclasts (day 3) and mature osteoclasts (day 0), pre-osteoclasts (day 3) and mature osteoclasts (day 5). (B) Western blots of cdc42 protein expression in WT and cdc42<sup> $\Delta$ OC/ $\Delta$ OC</sup> BMMs (day 0), pre-osteoclasts (day 3) and mature osteoclasts (day 5).  $\beta$ -actin serves as loading control. (C) PCR genotyping of BMMs from WT>WT and cdc42GAP<sup>-/-</sup> >WT chimeric mice. (D) Activation of cdc42 and Rac1 in WT and Cdc42GAP<sup>-/-</sup> osteoclasts in response to M-CSF, as determined by pull-down assay.  $\beta$ -actin serves as loading controls.</sup></sup>



Supplemental Fig 2. cdc42<sup> $\Delta$ oc/ $\Delta$ oc</sub> osteoclasts differentiate normally. WT and cdc42<sup> $\Delta$ OC/ $\Delta$ OC</sub> BMMs were exposed to RANKL and M-CSF with time. Expression of the osteoclast differentiation markers c-Src and cathepsin K (CtsK) were determined by immunoblot.  $\beta$ -Actin serves as loading control.</sup></sup>



Supplemental Fig 3. cdc42<sup> $\Delta$ oc/ $\Delta$ oc</sub> osteoclasts exhibit delayed actin ring formation. See legend Fig 6C.</sup>



Supplemental Fig 4. Osteoclasts express aPKCs and Par isoforms. WT BMMs were treated with RANKL and M-CSF for as long as 5 days. Atypical PKC and Par mRNA and protein isoform expression was determined by (A) RT-PCR and (B) immunoblot , respectively. Osteoclast differentiation mRNAs were also measured. GAPDH and  $\beta$ -Actin serve as respective RT-PCR and immunoblot loading controls.



**Supplemental Fig 5. cdc42**<sup> $\Delta$ oc/ $\Delta$ oc</sub> osteoclasts develop filopodia. F-Actin of WT and cdc42<sup> $\Delta$ OC/ $\Delta$ OC</sub> osteoclasts was stained and the cells examined by fluorescent microscopy. Arrows indicate filopodia.</sup></sup>