

Fig. S2 Breast cancer-secreted EVs did not regulate osteoclast differentiation. Mouse bone marrow cells were cultured in 40 ng/ml M-CSF for 3 days before EV treatment and further induction of osteoclast differentiation with 40 ng/ml M-CSF and 100 ng/ml RANKL for up to 7 days. **a** Representative TRAP staining images of EV-treated osteoclasts after 7 days of differentiation. **b** Quantitative analysis of TRAP staining in (**a**). Mature osteoclasts were identified as multinucleated TRAP⁺ cells. **c** Relative RNA level of osteoclast differentiation marker genes *Trap* and *Ctsk* normalized to *Rpl19* in primary pre-osteoclast cells treated with indicated EVs and induced for osteoclast differentiation for 5 days.