

Supplemental Figure 2.  $E_2$  effects on ER+ tumor cell seeding of bone. Quantification of tumor cells detected in proximal tibiae of  $E_2$ -supplemented mice (young or mature, as indicated) vs. mice with no  $E_2$  supplementation, inoculated with DiD-labeled ER+ tumor cells. There were no significant differences (n.s.) in DiD+ tumor cell number in young mice with or without  $E_2$ , between  $E_2$  doses (0.05 vs. 0.72 mg) in young mice, or between young and mature mice treated with 0.72 mg  $E_2$ , as tested by one-way ANOVA with Sidak's posttest (n=10-16/group). DiD+ cells were not detected in naïve mice lacking  $E_2$ -supplementation and tumor inoculation (n=10)