

Supplemental Figure 1



Supplemental Figure 2



Supplemental Figure 3

Supplemental Figures

Supplemental Figure 1. Estren- α affects uteri of both C57BL/6 and Swiss Webster mice. Both C57BL/6 (a) and Swiss Webster (b) mice were sham-operated (SHAM) or ovariectomized and treated with implanted slow-release pellets delivering vehicle (OVX) or estren- α (Estren) as described in Methods. Estren- α exerted uterotrophic effects in both strains, increasing uterus weight significantly above the sham level. ⁰p<0.05 and ⁰⁰⁰p<0.001 vs. sham controls.

Supplemental Figure 2. Estren- α modulates ER and AR transcriptional activity.

The abilities of estren- α and PSK3471 to activate ER and AR transcriptional activity were assayed using HeLa cells stably transfected with plasmids containing (**A**) ER α and (**B**) ER β or HeLa cells transiently transfected with an ERE-luciferase reporter and ER α (**C**) or an ARE-luciferase reporter and AR (**D**) as described in Methods. The transfected cells were treated with the test compounds as indicated for 24 hours, then lysed. Luciferase activity was measured as described in Methods. Triplicate determinations of reporter activity were performed in all experiments. Estren- α stimulated transcription from both estrogen- or androgen-responsive promoters whereas PSK3471 did not display significant agonist activity in these assays. Data are presented normalized to the maximal activity of E₂ (**A-C**) or DHT (**D**).

Supplemental Figure 3. The effects of estren- α on bone and seminal vesicles cannot be separated by lowering the dose.

Wild-type male mice were sham-operated or orchidectomized. The orchidectomized mice were treated with implanted slow-release pellets delivering vehicle, DHT or decreasing amount of estren- α as indicated. After the treatment, the bone volume (BV/TV) (**A**), quantified by histomorphometry, and the seminal vesicle wet weight (**B**) were determined. As before, the higher doses of estren- α were as effective as DHT in preventing bone loss and increasing seminal vesicle weight. Both the bone volume and the seminal vesicle wet weight were somewhat decreased (20 and 27% respectively, compared to sham-operated mice) at the lowest dose of estren- α but only the effect on the seminal vesicles reached statistical significance. ⁰p<0.05 and ⁰⁰⁰p<0.001 vs. sham-operated controls.