Figure S1. Autophagic protein catabolism is active in breast cancer cells undergoing antiestrogen treatment as a single agent or in combination with antiprogestin treatment. Evenly seeded MCF-7 cells were treated with E2 (10 nM) alone or in combination with 1.0 μ M 4-OHT, 10 μ M MIF, or 4-OHT + MIF (1.0 μ M + 10.0 μ M, respectively). At the times indicated and as described in Materials and methods the following were determined: (A) an increase in the number of cytosolic autophagosomes and autolysosomes identified by electron microscopy in cells treated with 4-OHT and/or MIF compared to E2 treated cells; (B) an increase in long-lived protein catabolism in cells treated with 4-OHT, MIF, and 4-OHT plus MIF as compared to E2-treated control cells (long-lived protein catabolism is an independent measure of autophagy); (C) total cell counts utilizing a Coulter Counter to confirm the efficacy of E2 in promoting MCF-7 cell growth, and 4-OHT treatment as a single agent or in combination with MIF in blocking cell growth; and (D) an increase in cytosolic autophagosomes/autolysosomes (designated by arrows) induced by 4-OHT plus MIF treatment versus E2 treatment as determined by electron microscopic examination; mitochondria are designated by arrow heads. Comparisons which are statistically significant (P<0.05 or P<0.001) are designated by an * or **, respectively and are between: athe designated treatment compared to the E2 control cells; and ^bthe designated treatment compared to the 4-OHT treatment.

