



Additional File 3: Overexpression of a MEK1 dominant negative mutant protein (MEKDN) induces ROS-dependent apoptosis in MCF-7 cells undergoing hormonal treatments in the presence or absence of IGF-1. (A-B) ROS levels were determined for MCF-7 cells infected with Ad-CMV alone or Ad-CMV-MEK1DN. (C) The percent of mitochondrial membrane depolarization was determined for MCF-7 cells infected with Ad-CMV or Ad-CMV-MEK1DN. (A-C) Cells were infected with either Ad-CMV alone or Ad-CMV-MEK1DN for 24 h, after which cells were treated with E2, E2 + 4-OHT, E2 + MIF, or E2 + 4-OHT + MIF in presence of IGF-1 (20ng/mL). The cells were harvested for ROS determination or mitochondrial membrane depolarization levels as described in materials and methods at 6 h (A) and 24 h (B) and 72 h (C). The Values are expressed as mean \pm S.D (n=3). Statistical significance was determined by One-Way ANOVA using sigma plot 11 for windows. Significant differences are indicated between treatment groups as follows: ^a E2 + IGF-1 (Ad-CMV- versus Ad-CMV-MEK1DN-infected); ^b E2 + 4-OHT + IGF-1 (Ad-CMV- versus Ad-CMV-MEK1DN-infected); ^c E2 + MIF + IGF-1 (Ad-CMV- versus Ad-CMV-MEK1DN-infected); ^d E2 + 4-OHT + MIF + IGF-1 (Ad-CMV- versus Ad-CMV-MEK1DN-infected). The symbol * represent statistical significance at P<0.001.