

Supplementary Materials

# Extra-Virgin Olive Oil and Its Minor Compounds Influence Apoptosis in Experimental Mammary Tumors and Human Breast Cancer Cell Lines

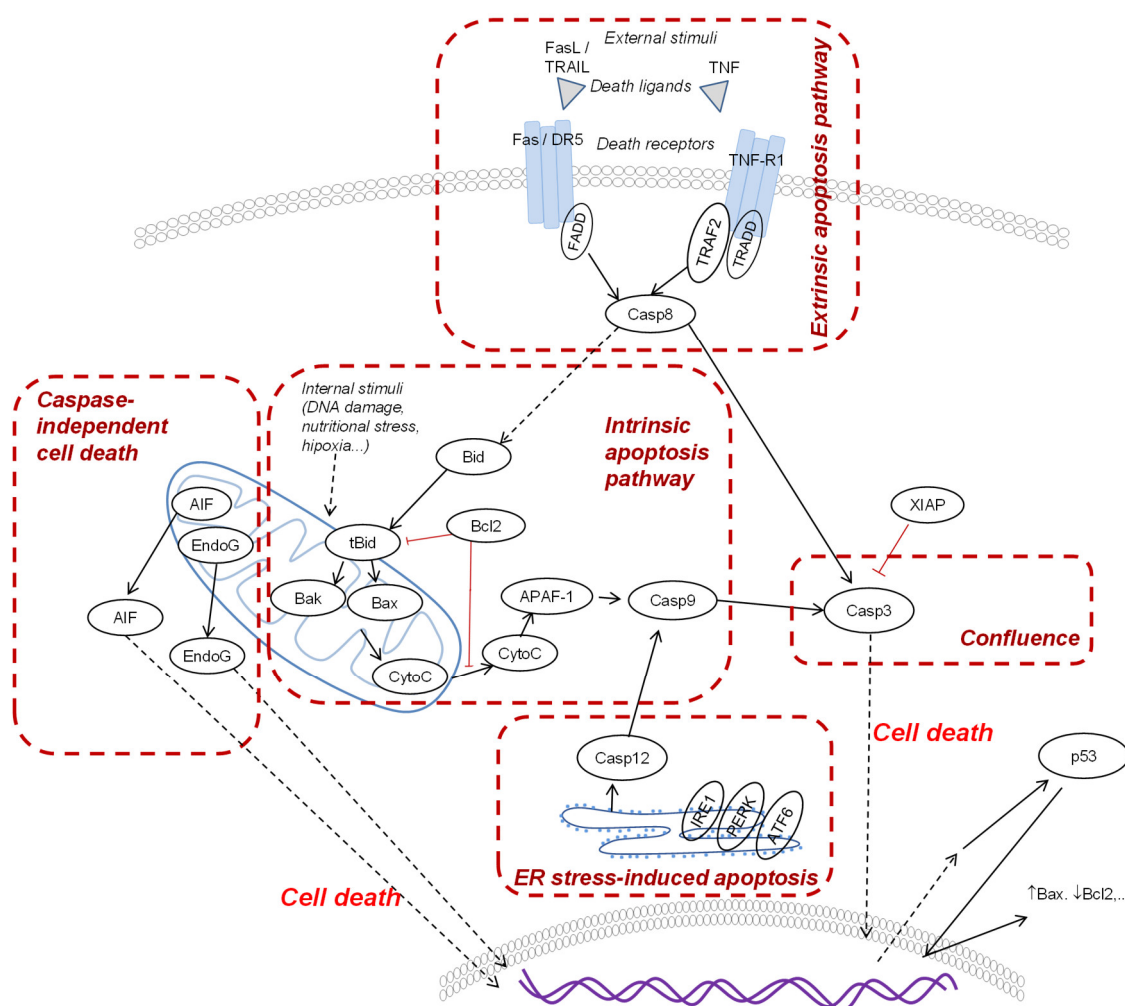
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**Figure S1.** Different cell death signaling pathways: extrinsic apoptosis, intrinsic apoptosis, confluence between extrinsic and intrinsic pathways, ER stress-induced apoptosis, and caspase-independent cell death.

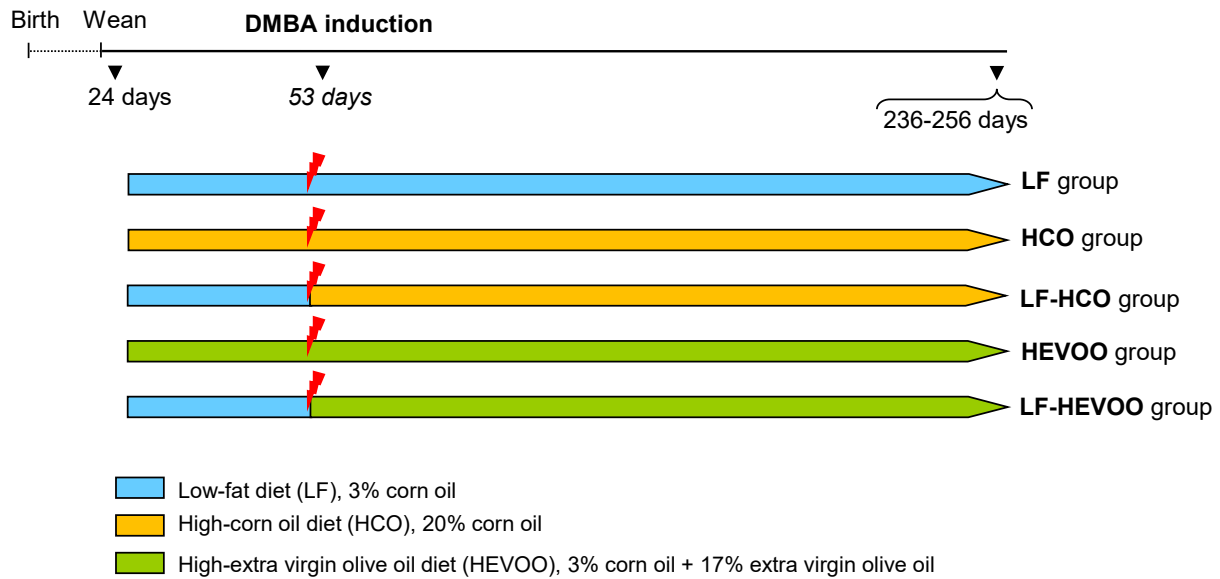
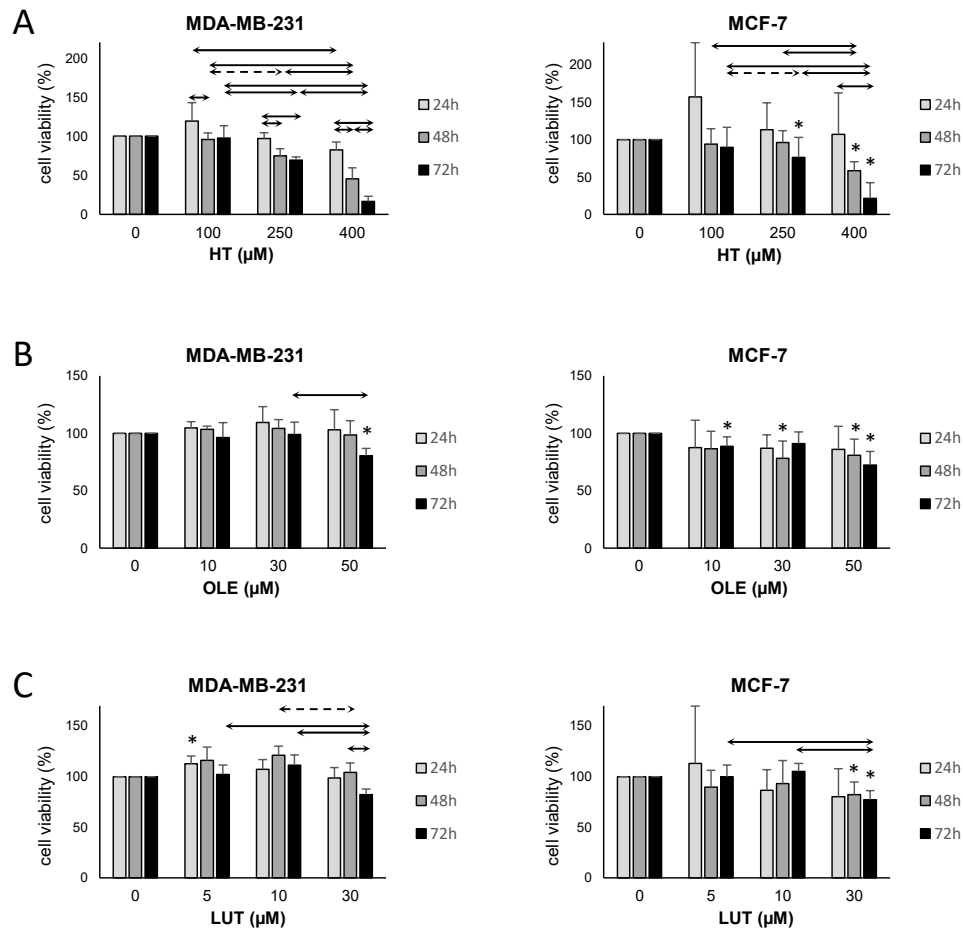
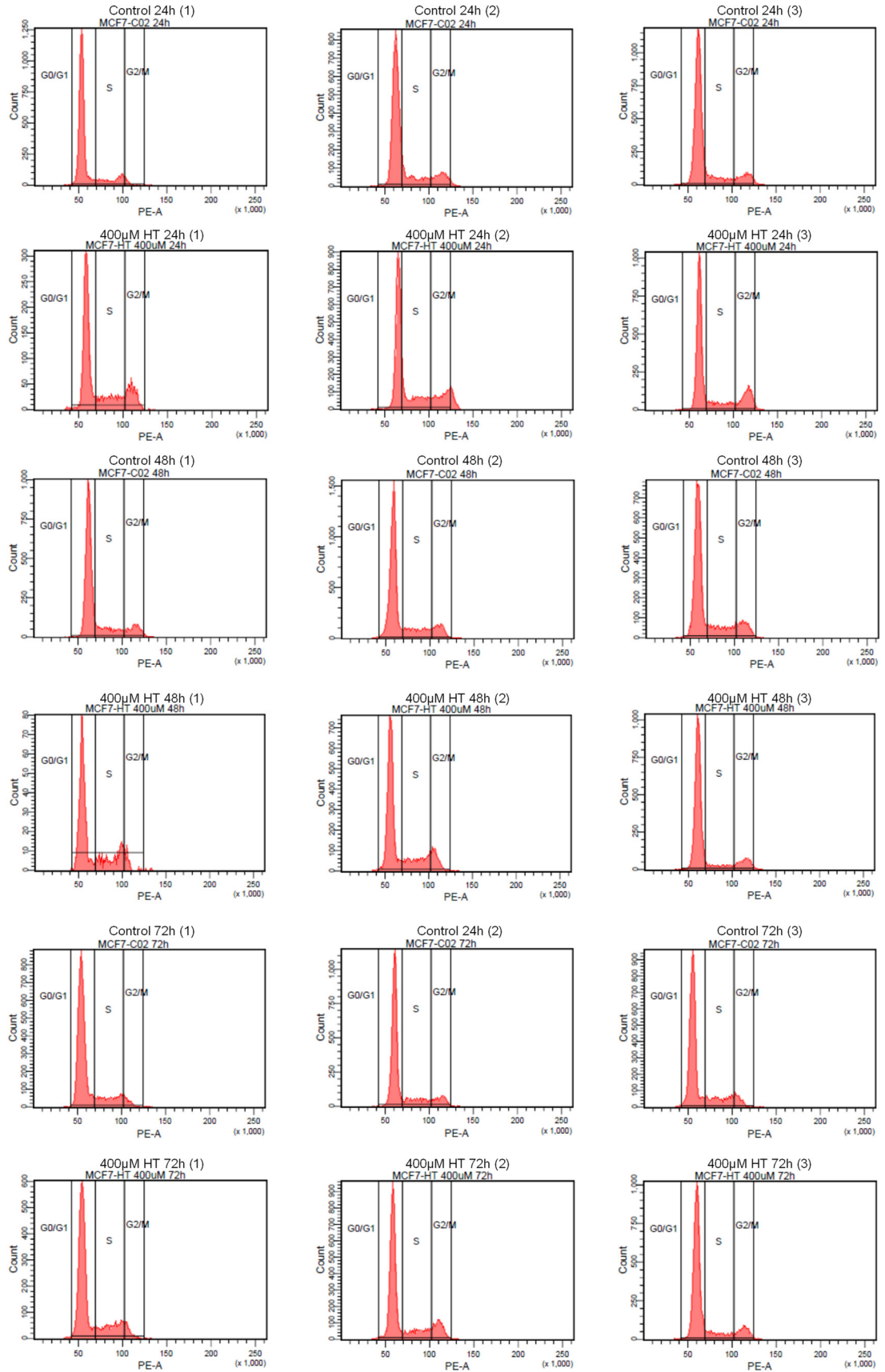


Figure S2. Experimental design.

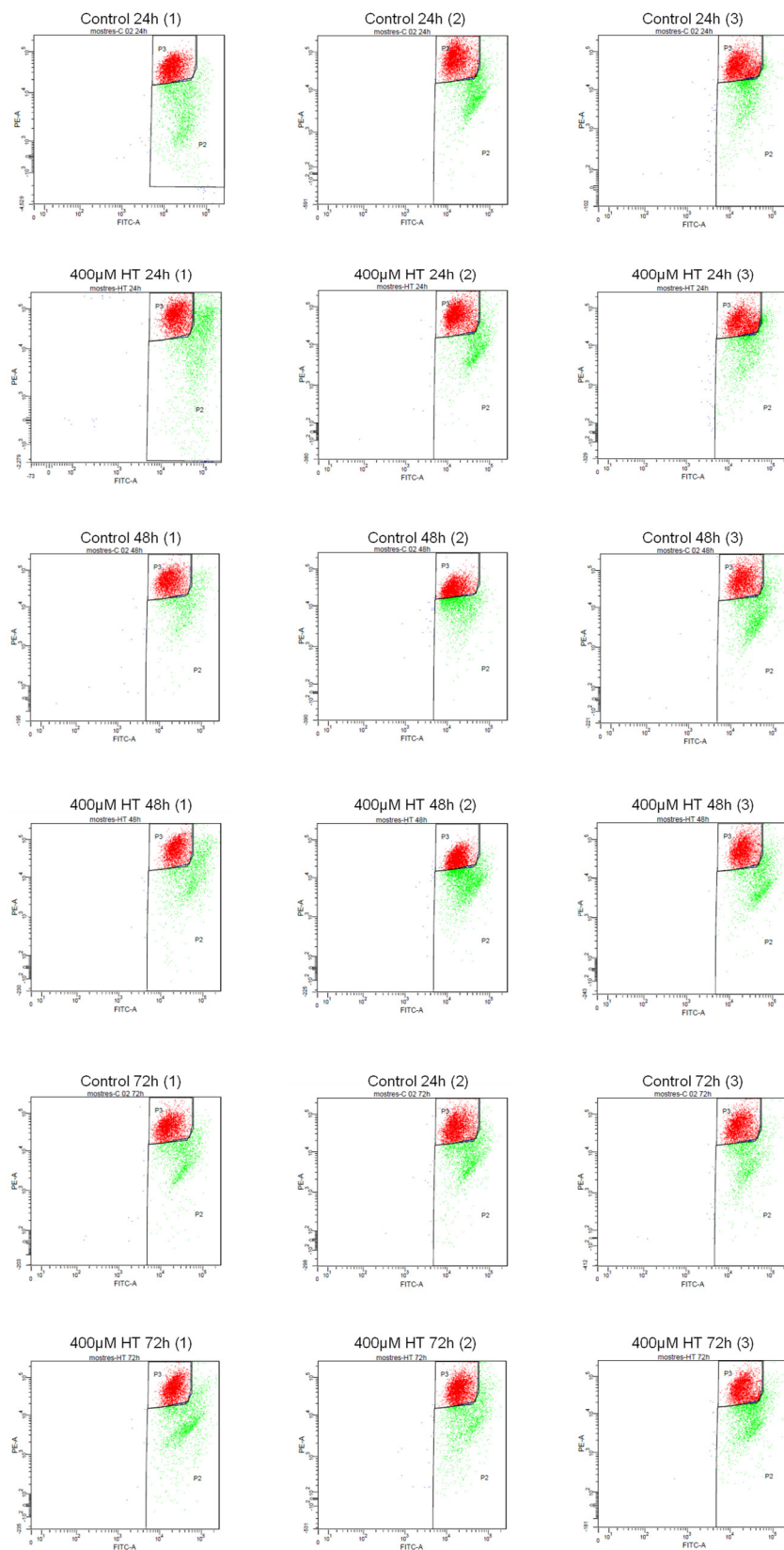


**Figure S3.** Effect of treatment with the EVOO minor compounds HT, OLE and LUT on cell viability in MDA-MB-231 and MCF-7 cell lines. Cell viability analysis using the MTT assay of MDA-MB-231 and MCF-7 cell lines treated with different doses of EVOO minor compounds for 24h, 48h or 72h. (A) Hydroxytyrosol (HT). (B) Oleuropein (OLE). (C) Luteolin (LUT). Mean + standard deviation of three independent experiments. \*:  $p < 0.05$  compared to control 0.1% DMSO-treated cells. Solid lines connecting bars indicate statistically significant differences ( $p < 0.05$ ) between doses or time of exposure, dashed lines indicate differences close to significance ( $p < 0.1$ ).

A



B



**Figure S4.** Original plots of the in vitro analysis of cell cycle and apoptosis by flow cytometry. (A) DNA content of cells stained with propidium iodide (G0/G1 and G2/M phase peaks are separated by the S-phase). (B) Dot plots of gating of JC1 aggregates (live cells, red fluorescence) and JC1 monomer (low mitochondrial membrane potential, apoptotic cells, green fluorescence) populations.