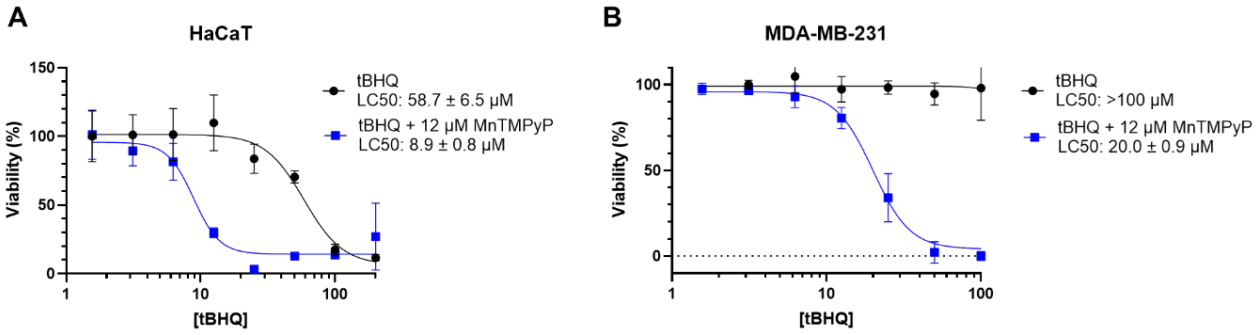

Figure S1. The combination of tBHQ + MnTMPyP is toxic to immortalized keratinocyte line HaCaT (**A**) and cancer line MDA-MB-231 (**B**). Viability (measured by the BCA assay) was normalized to the lowest concentration of tBHQ. $n = 6$ technical replicates.

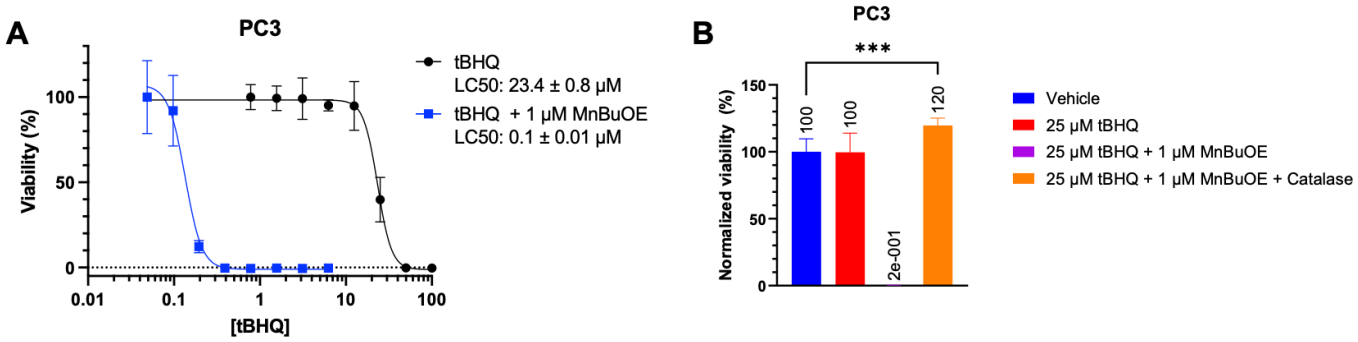

Figure S2. Supporting data for key results in PC3 cells for experiments shown in the main text, with a different cell passage number. (**A**) Data from cells at a different passage number to that in Figure 1C showing the combination of tBHQ + MnBuOE is much more toxic to PC3 cells than tBHQ alone. PC3 cells were treated with increasing concentrations of tBHQ alone or in combination with MnBuOE. Viability was measured using a CellTiter Glo assay and normalized to the signal for the lowest concentration of treatment. $n = 6$ technical replicates. (**B**) Data from cells at a different passage number to that in Figure 2E showing catalase rescues the toxicity caused by the tBHQ-MnBuOE combination. PC3 cells were treated with 25 μM tBHQ alone or in combination with 1 μM MnBuOE. Viability was measured using a CellTiter Glo assay and normalized to the signal for the lowest concentration of treatment. $n = 6$ technical replicates. "***" in the figure indicates a p value of 0.0005.

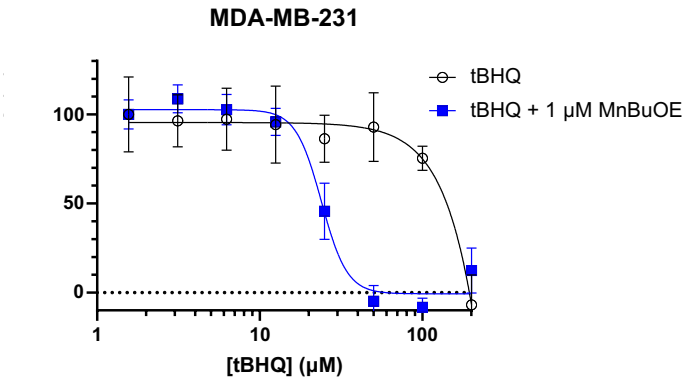

Figure S3. The combination of tBHQ + MnBuOE is toxic to cancer line MDA-MB-231. Viability (measured by the BCA assay) was normalized to the lowest concentration of tBHQ. $n = 6$ technical replicates.

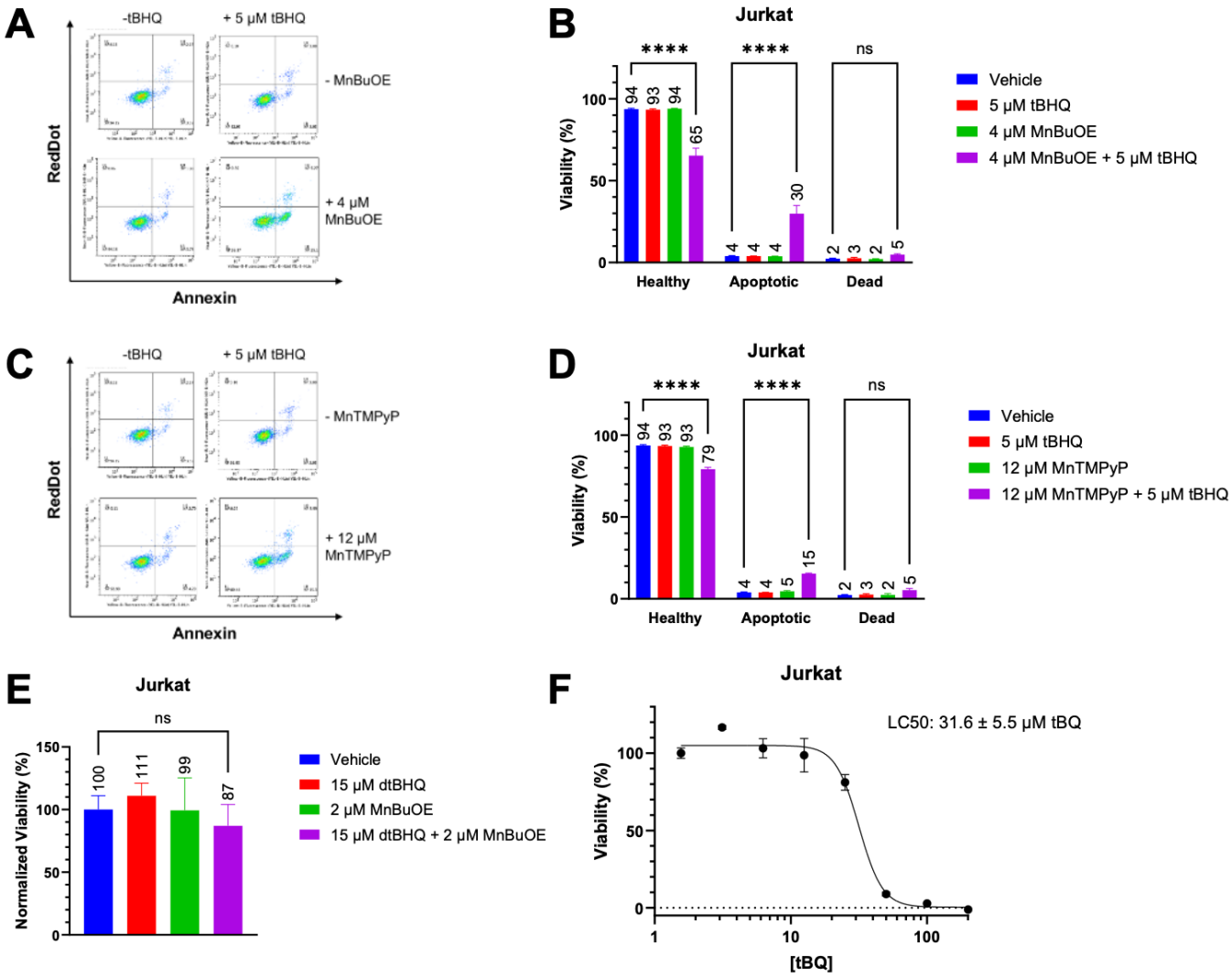

Figure S4. Supporting data for key results in Jurkat cells for experiments shown in the main text, with a different cell passage number. (**A, B**) The Annexin V/RedDot assay was used to measure apoptotic and necrotic cell death in Jurkat cells exposed to 5 µM tBHQ and 4 µM MnBuOE, singly or in combination. $n = 4$ (technical replicates). (**C, D**) Using the same assay as for *A* and *B*, apoptosis or necrosis was measured in Jurkat cells exposed to 5 µM tBHQ and 12 µM MnTMPyP, singly or in combination. $n = 4$ (technical replicates). (**E**) The CTF assay was used to measure the viability of Jurkat cells treated with 15 µM dtBHQ and 2 µM MnBuOE, alone or in combination, for 24. Viability was normalized to the untreated control. n=5 (technical replicates). (**F**) Cell death from treatment with tBQ was measured in Jurkat cells by the CTF assay. Viability was normalized to the signal for the lowest concentration of tBQ. $n = 6$ (technical replicates). "****" in the figure indicates a p value <0.0001, and "ns" indicates a p value that was not significant.

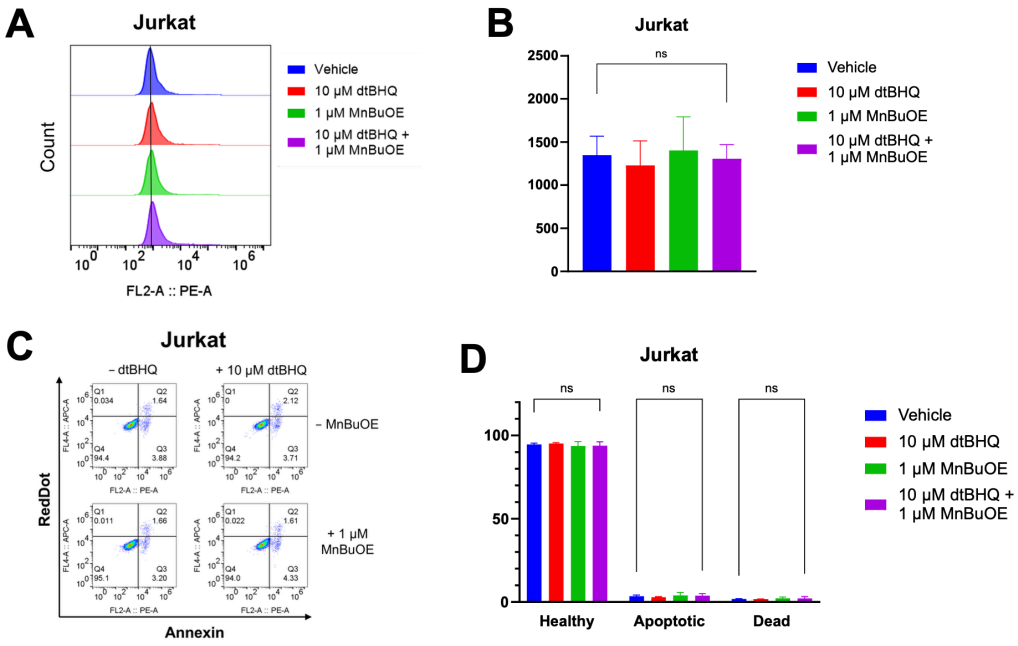

Figure S5. Use of dtBHQ in Jurkat cells to probe the mechanism of cell death suggests that oxidative stress is not from tBHQ redox cycling. (**A, B**) MitoSOX fluorescence was measured in Jurkat cells. Cells were treated with dtBHQ and MnBuOE alone or in combination. $n = 2$ biological replicates with a total of 4-6 technical replicates. (**C, D**) In the same treatments as for *A* and *B*, apoptosis or necrosis in Jurkat cells was measured by the Annexin V/RedDot assay. $n = 2$ biological replicates with a total of 6 technical replicates.

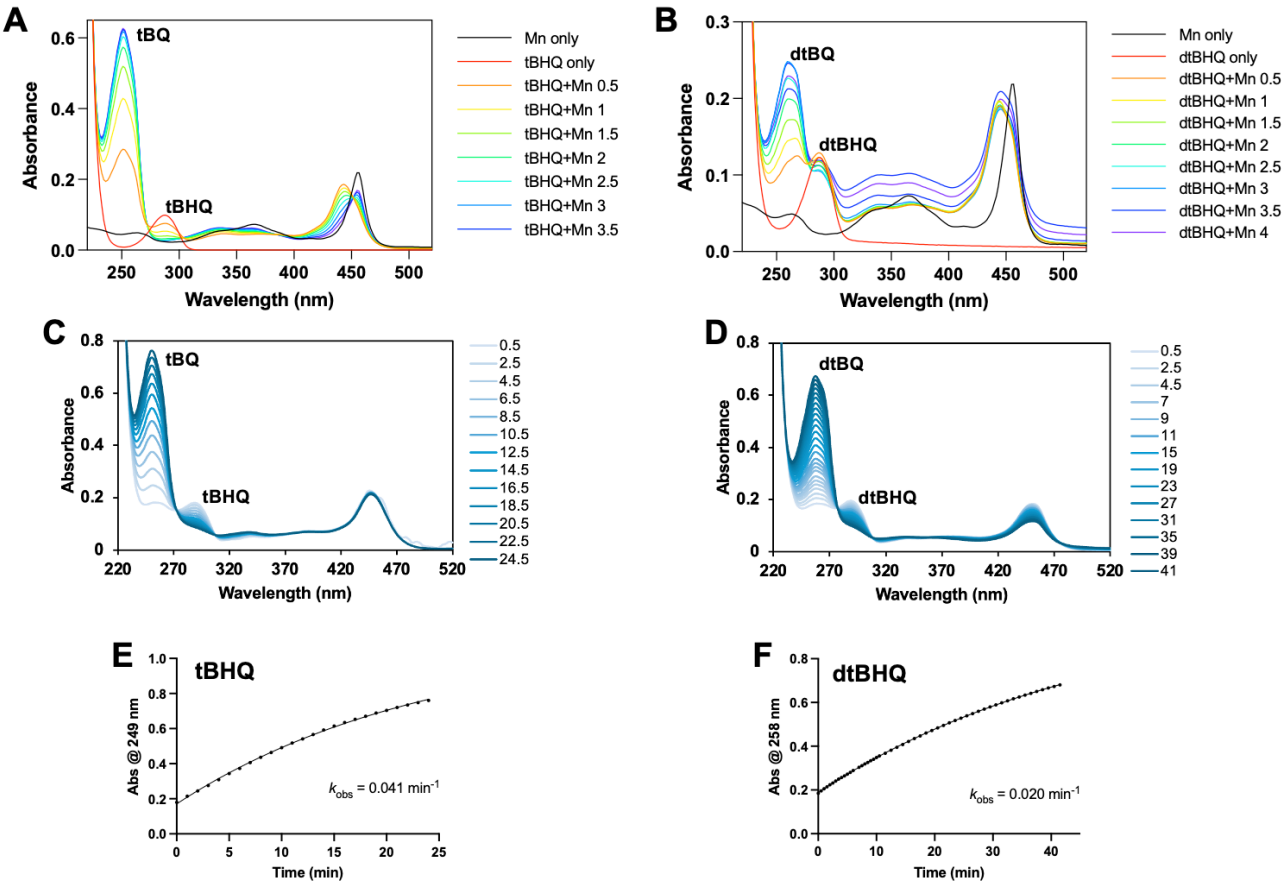

Figure S6. Catalysis of (d)tBHQ oxidation by MnBuOE. (**A**) UV-Vis spectroscopy was used to monitor the oxidation of tBHQ to tBQ, catalyzed by MnBuOE, in aqueous buffer at pH 7.4, at 21°C (room temperature). The trace labels show the time elapsed since reaction initiation, in min. (**B**) As for *A*, for dtBHQ. The increase in absorbance at 500 nm at later timepoints indicates the formation of insoluble dtBQ at 21°C. (**C, D**) As for *A* and *B*, but in 50% ethanol to maintain solubility of the product dtBQ. The trace labels show the time elapsed since reaction initiation, in min. (E, F) The absorbance from *C* and *D* at the indicated wavelengths, plotted over time. The data were fit to a single-phase decay to obtain k_{obs}. "Mn" in the legend is the abbreviation for "MnBuOE".

Supplementary Methods

HaCaT Cell Culture

The HaCaT cell line (Cell Lines Service, 300493, Sioux Falls, SD, USA) was grown in complete media consisting of DMEM supplemented with L-glutamine, 10% FBS, and 15 mM HEPES (4-(2-hydroxyethyl)-1-piper-azineethanesulfonic acid) pH 7.2. Cells were passaged when they had reached 60-80% confluency.