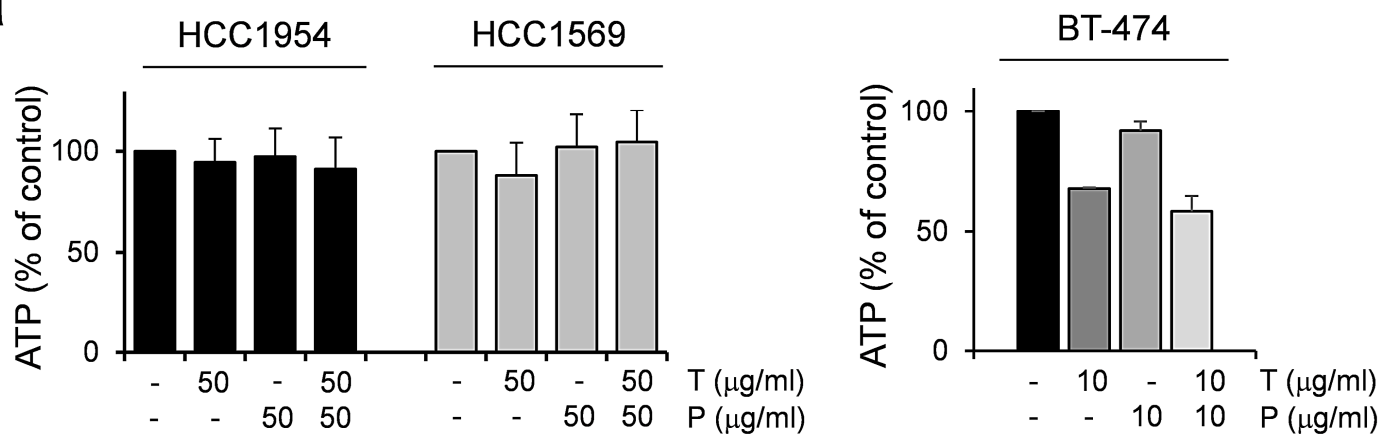
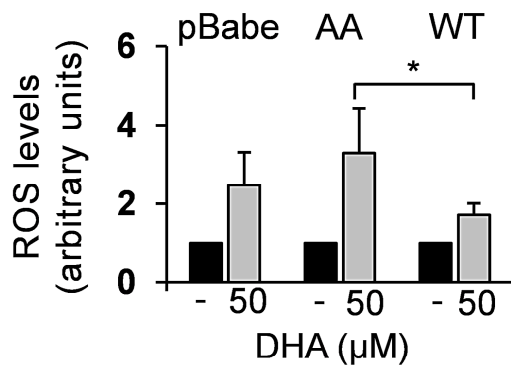
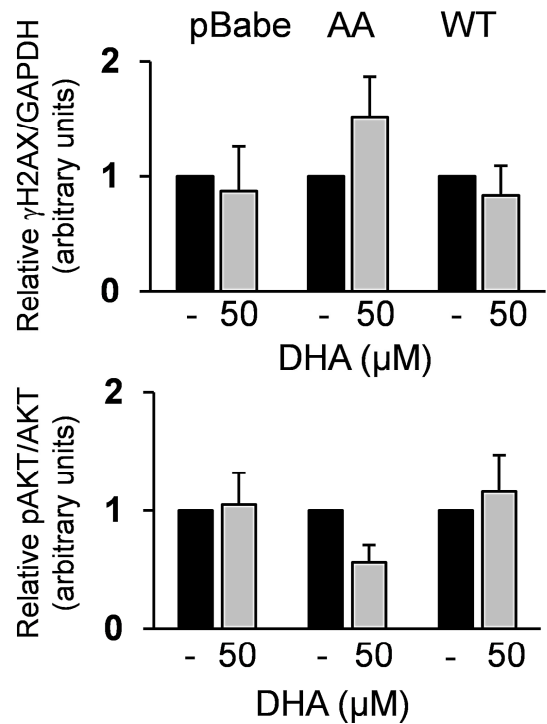
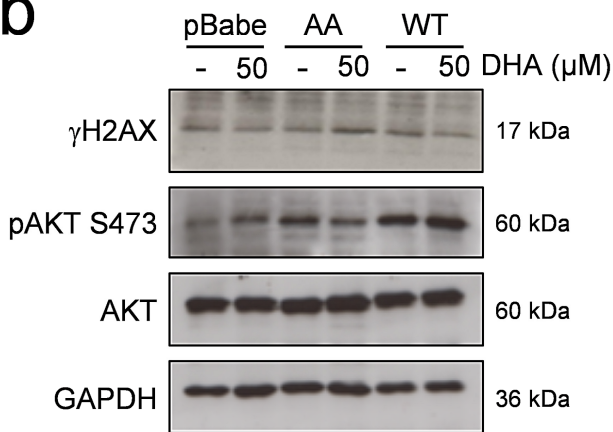


**a**

Supplementary Figure S1. Effect of trastuzumab and pertuzumab on cell growth of HER2+ BC cell lines.

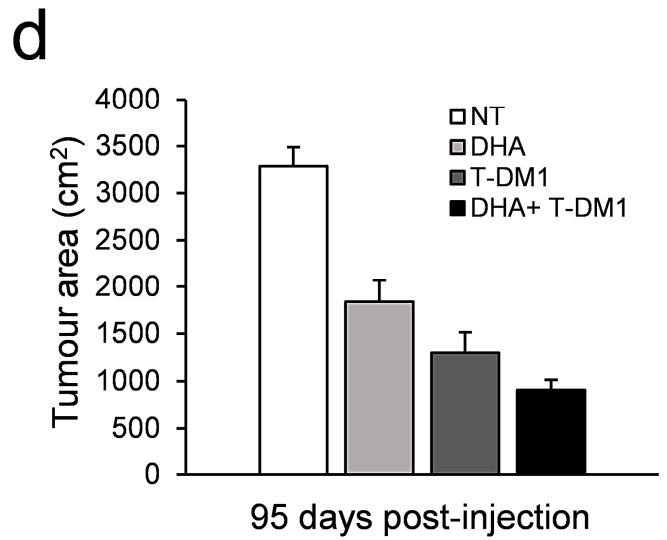
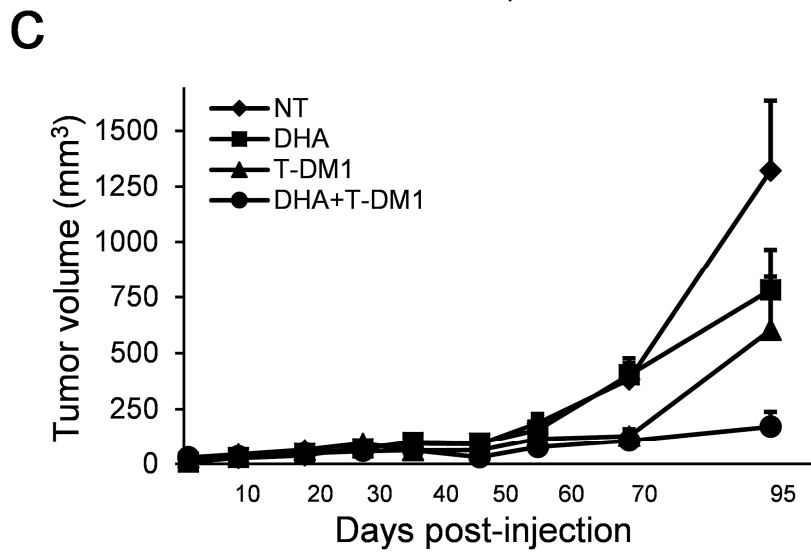
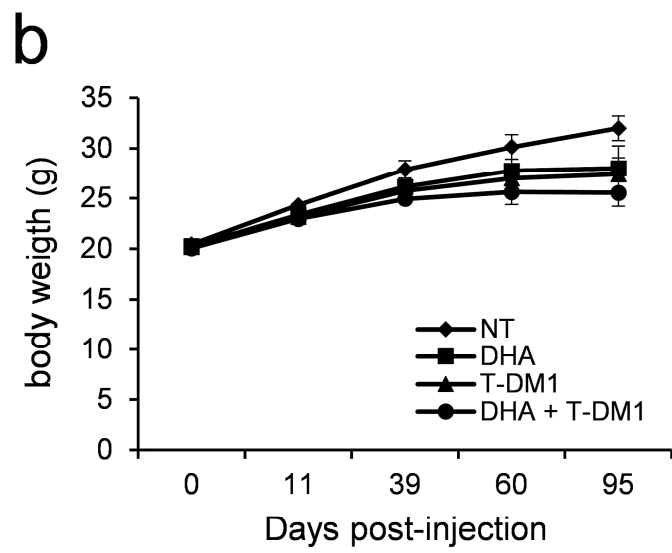
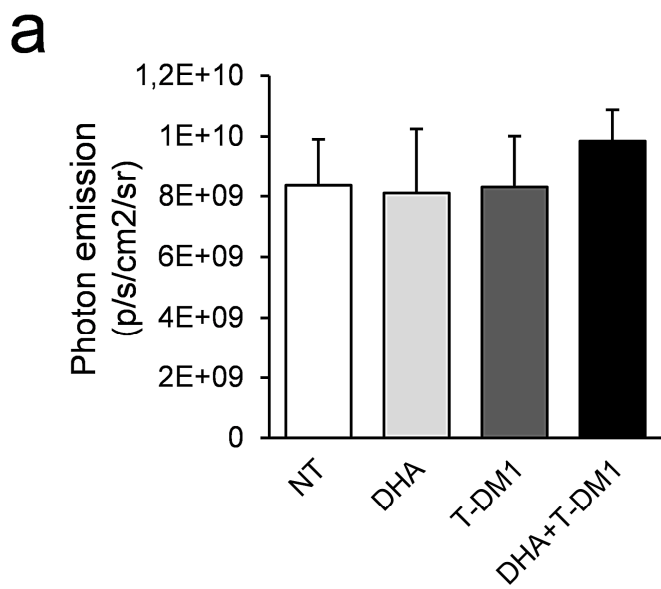
a) Cells were treated with trastuzumab and pertuzumab at the indicated concentration for 6 days. At the end of incubation time, the number of viable cells was determined using ATP-assay. Data are expressed as the percentage of viable cells relative to controls. Values represent the mean  $\pm$  SD,  $n = 3$ .

Supplementary Figure S1

**a****b**

Supplementary Figure S2. DHA induces an increase of ROS levels in MCF10A-pBabe, MCF10A-AATCTP and MCF10A-wTCTP cells.

- Cells were treated with DHA for 4 days. ROS production was measured at the end of incubation time. Data are expressed as ROS levels relative to controls.  $n = 3$ . Significant differences between treated and control cells, at any time of treatment, are indicated,  $* = p < 0.05$ .
- Western blot analysis of the indicated proteins in cell lysate of cells after exposition to DHA for 4 days at the indicated concentrations. GAPDH was used as loading control. For densitometric analysis the intensity of each band was normalized to the respective GAPDH. Quantification analysis was performed by using ImageJ software.



Supplementary Figure S3. In vivo efficacy of DHA with T-DM1 in HCC1954 xenografts.

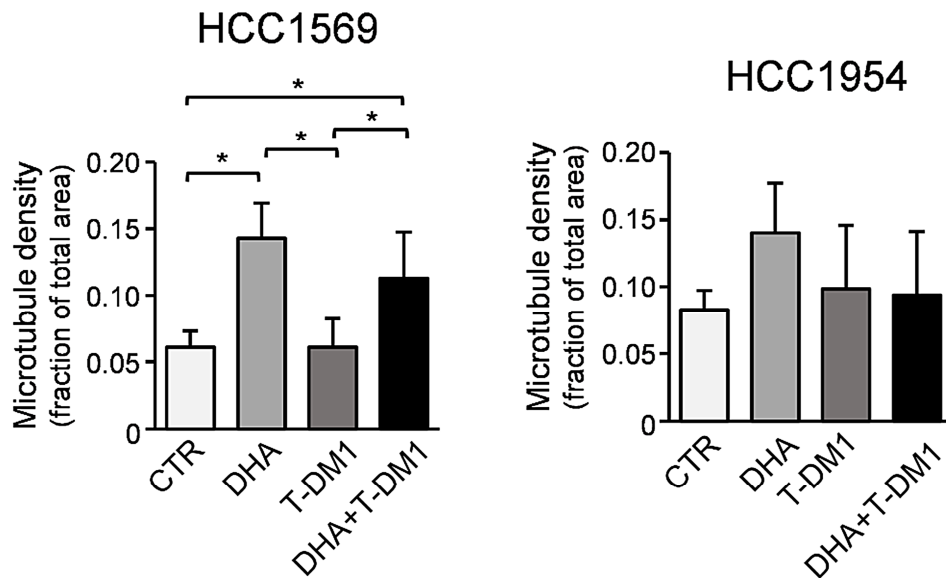
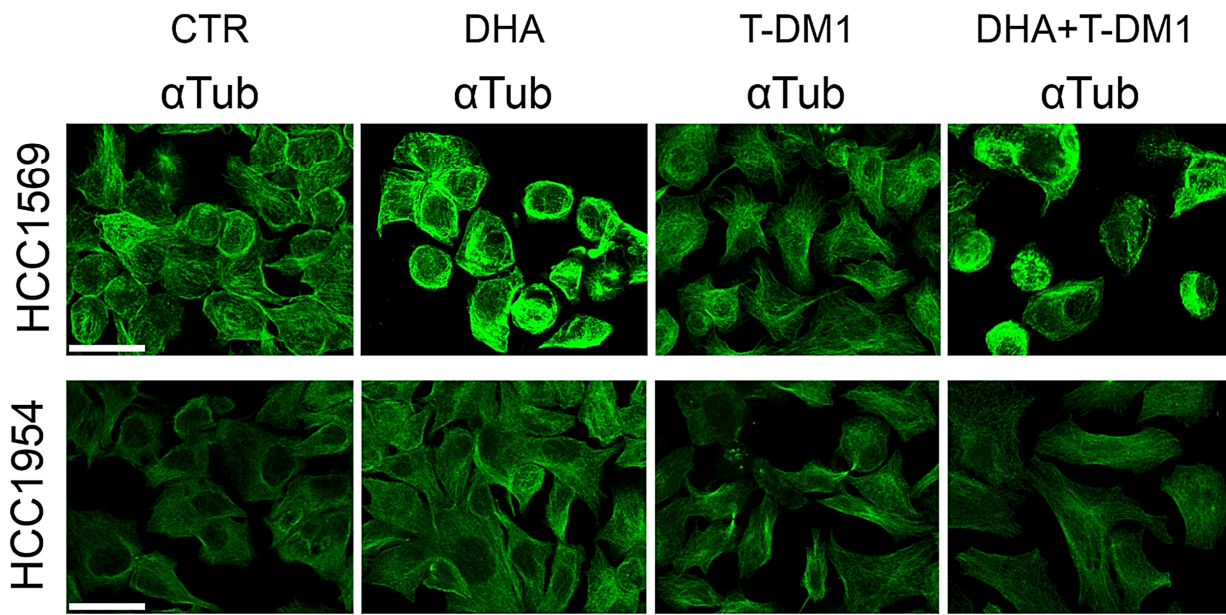
a) Tumor bioluminescent emission after 22 days after tumour cell inoculation. Photon emission is measured as photons/sec/cm<sup>2</sup>/steradian. Mean ± SEM

b) Weight of CB17SCID mice carrying HCC1954 xenografts treated with T-DM1 and DHA, individually or in combination as described in legend 4d. Mean ± SEM of 5 animals is reported.

c) Tumour growth in HCC1954 xenografts treated with T-DM1 and DHA, individually or in combination as described in legend 4d. Mean ± SEM of 5 animals is reported

d) Tumour area (cm<sup>2</sup>) has been measured by automatically drawing the regions of interest upon BLI analysis using the Living Image® software (PerkinElmer). Mean ± SEM of 5 animals is reported

a



Effects of two-drug combination on microtubule density in HCC1569 and HCC1954 cells.

a) Cells were pre-treated with DHA for 24 h and then treated with T-DM1 for 3 days. Immunofluorescence detection of  $\alpha$ Tubulin (green), bar = 25  $\mu$ m (upper panel). Quantitative analysis was performed as described in Materials and Method (lower panel).

Supplementary Figure S4