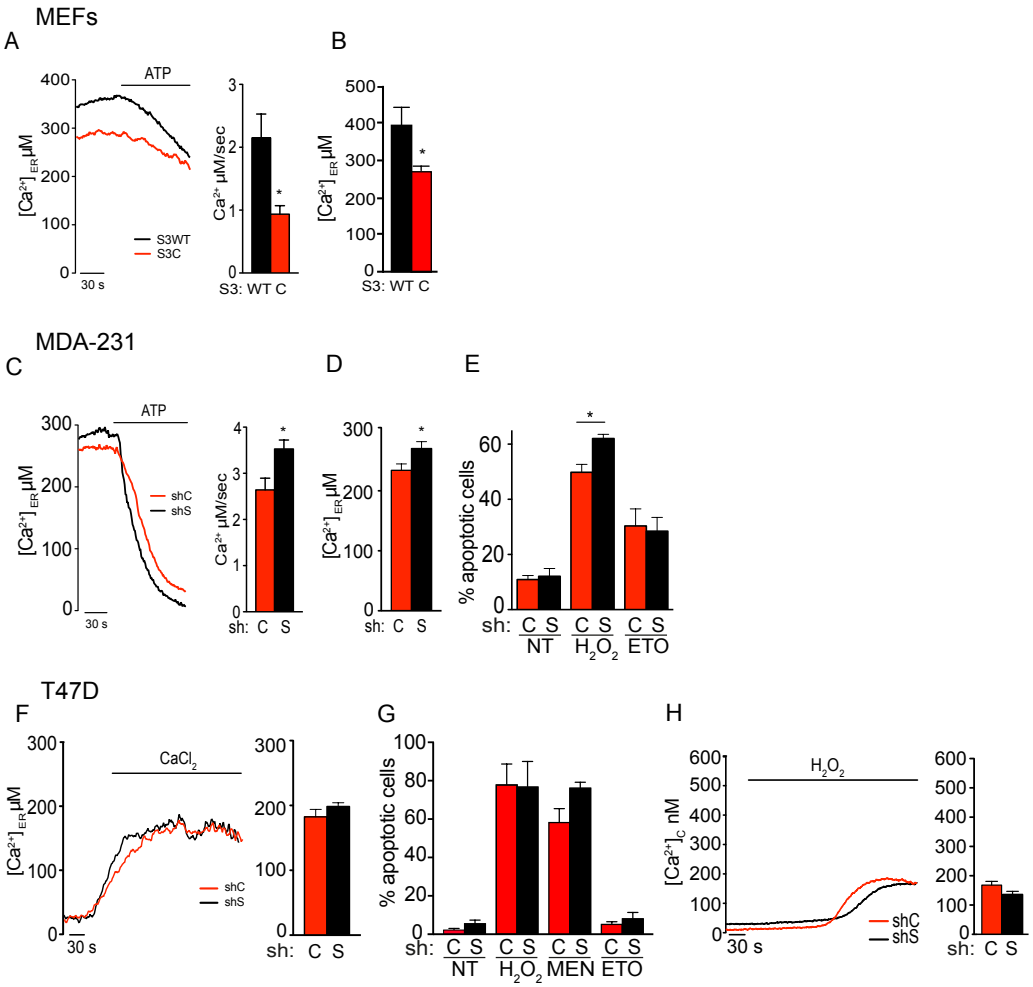
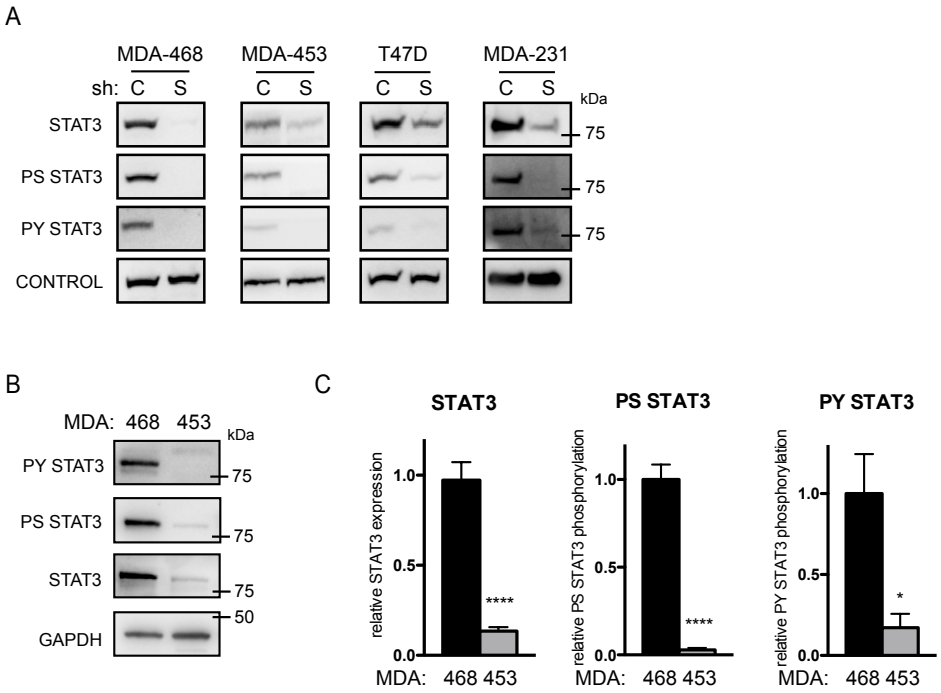
SUPPLEMENTARY INFORMATIONS

Figure S1.



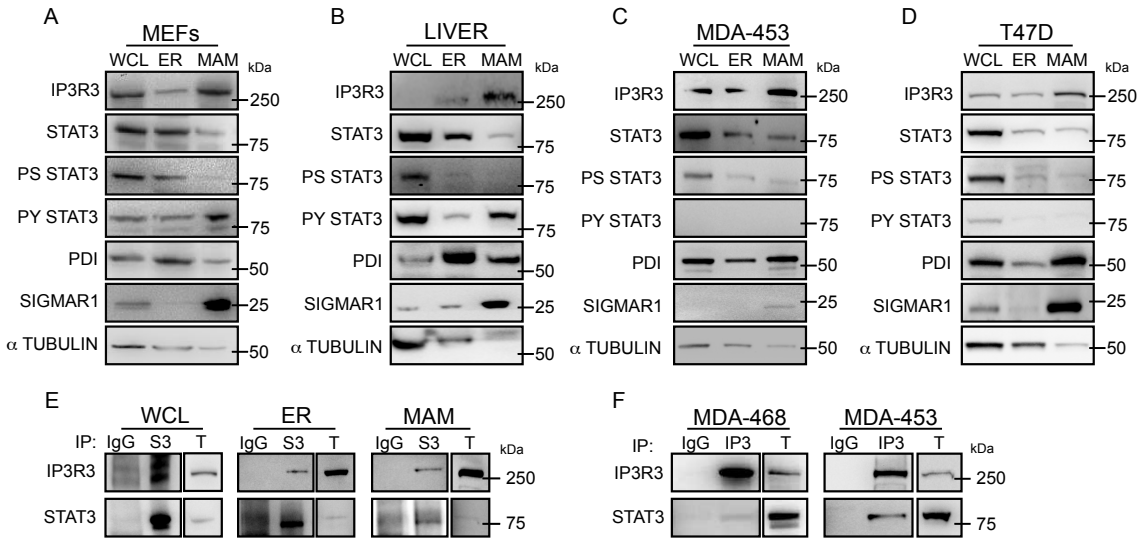
Silencing and apoptosis. (A, B) ER Ca^{2+} release **(A)** and content **(B)** in primary MEF cells of the indicated genotypes (S3WT, STAT3WT/WT, black; S3C, STAT3C/C, red). To induce Ca^{2+} release from the ER, the cells were challenged with ATP. A representative trace is shown. ER calcium release is quantified by the bars (mean± SEM of 10 traces from two independent experiments) and expressed as µM/sec. **(C-E)** MDA-MB-231 cells, silenced or not for STAT3. ER Ca^{2+} release **(C)** and content **(D)** were measured as above; bars are mean± SEM of 27 traces from three independent experiments. **(E)** Apoptotic response in MDA-MB-231 cells, silenced (shS, black) or not (shC, red) for STAT3, upon treatment with H_2O_2 or etoposide (ETO). NT, untreated. Bars represent the percentage of Annexin V positive cells (mean±SEM from at least 3 independent experiments). The asterisks indicate statistically significant differences. *, $p<0,05$. **(F-H)** T47D cells, silenced or not for STAT3. **(F)** Since T47D did not respond with Calcium release to any of the treatments tested, Ca^{2+} content was measured as Ca^{2+}-dependent ER aequorin signals upon $CaCl_2$ administration. shS, shSTAT3; shC, sh control. Bars are mean± SEM of 10 traces from two independent experiments. **(G)** Apoptotic response upon treatment with H_2O_2, menadione (MEN) or etoposide (ETO). NT, untreated. Bars represent the percentage of Annexin V/PI positive cells (mean±SEM from at least 5 independent experiments). **(H)** Cytoplasmic calcium release upon H_2O_2 stimulation in T47D cells. The bars represent mean± SEM of at least 12 measurements from three independent experiments.

Figure S2.



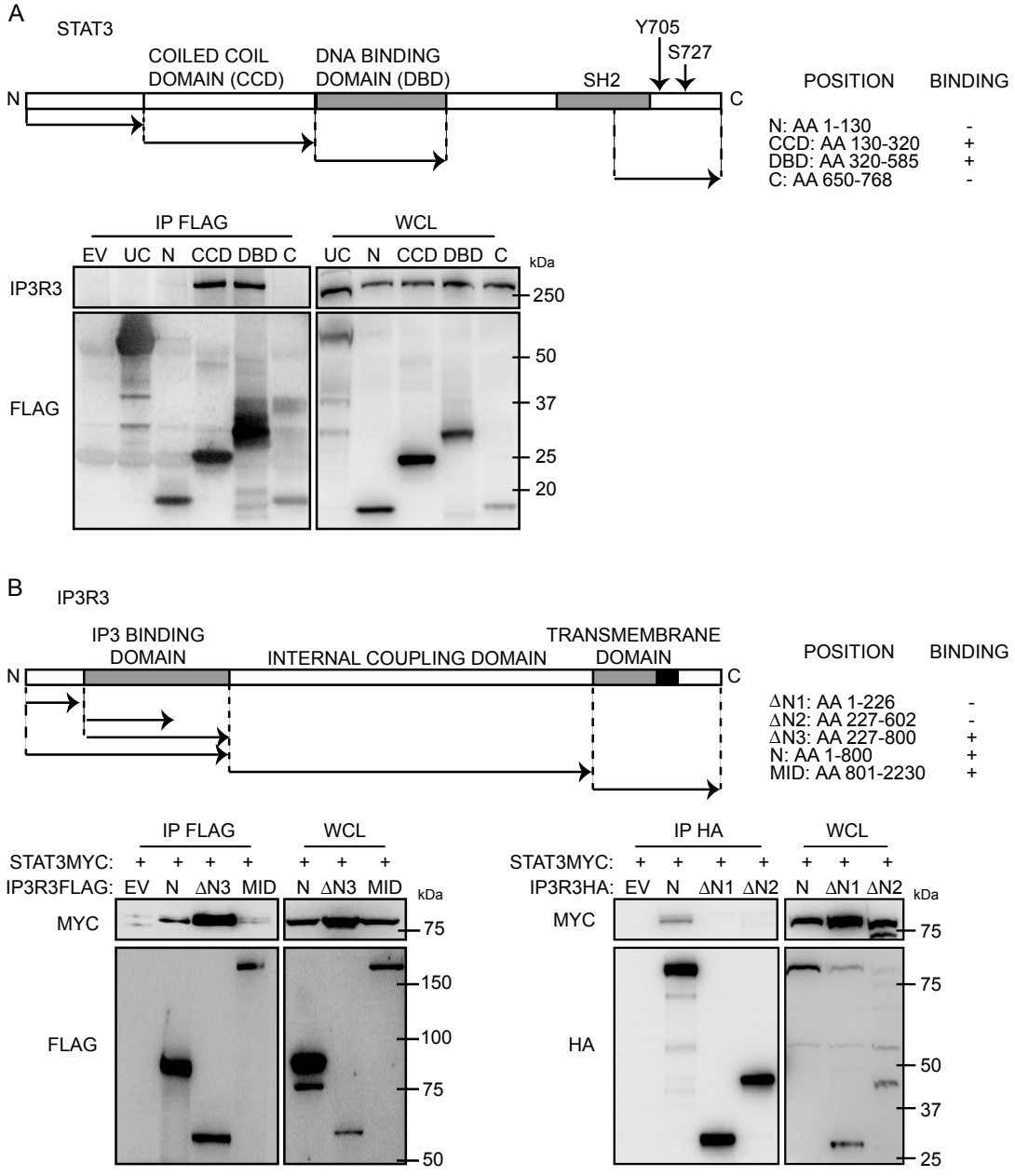
Relative levels of STAT3 and its phosphorylation in breast cancer cells.
(A) STAT3 silencing. The indicated cell lines were transduced with lentiviral vectors expressing either control (C) or STAT3 (S) shRNAs, and silencing was assessed by immunoblotting. **(B)** MDA-MB-468 and MDA-MB-453 whole cell lysates were analysed by Western blot with the indicated antibodies. **(C)** Quantification of 4 independent Western blots as in B. Bars represent mean±SEM of expression/phosphorylation levels of 4 independent experiments. The asterisks indicate statistically significant differences. *, $p<0,05$; ****, $p<0,001$.

Figure S3.



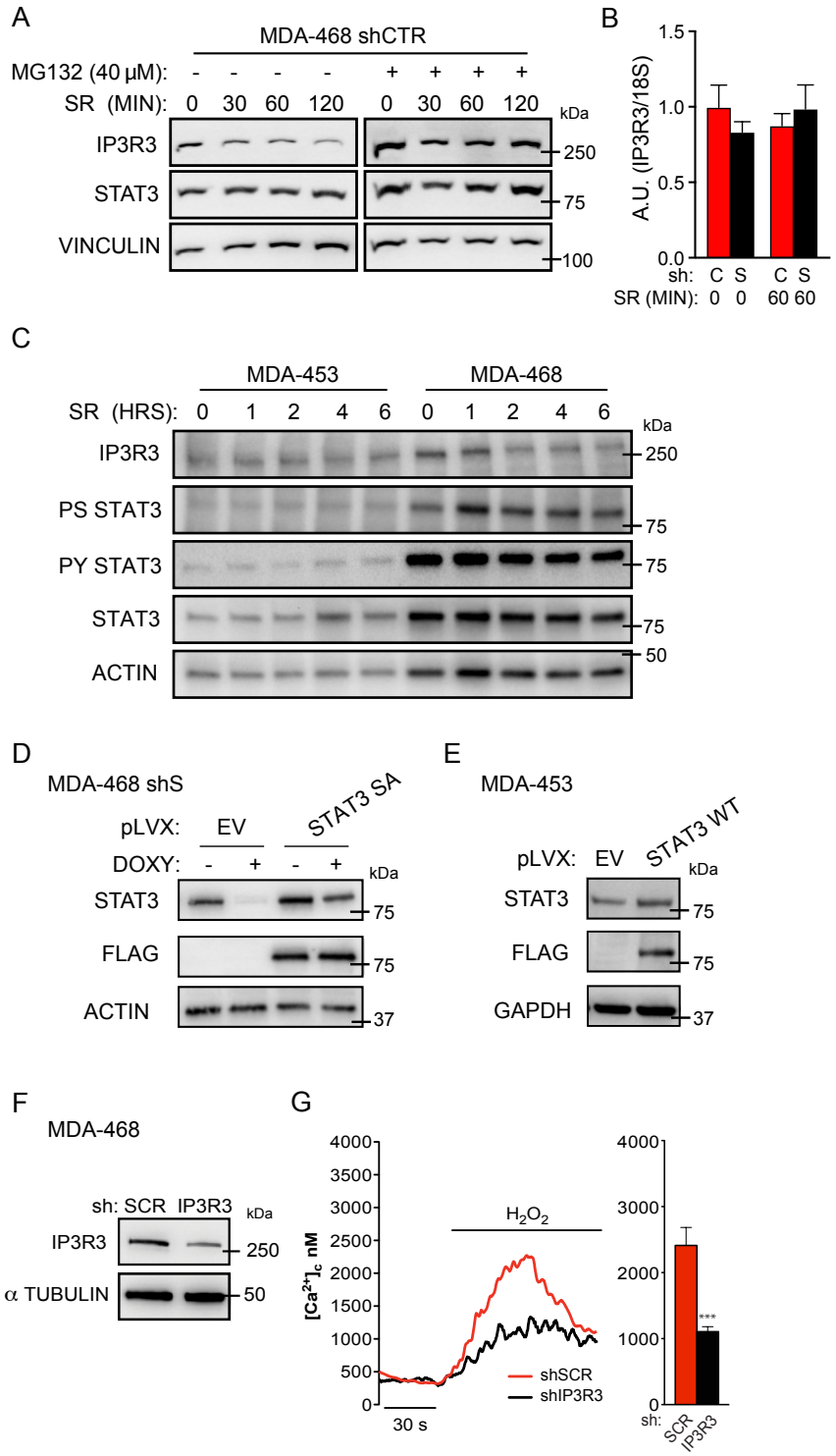
STAT3 localization and interaction with IP3R3. **(A-D)** Western blot analysis upon cell fractionation of MEF cells **(A)**, liver **(B)**, MDA-MB-453 cells **(C)** and T47D cells **(D)**. Representative of at least 2 independent experiments. **(E)** STAT3:IP3R3 co-immunoprecipitation from whole cell lysates (WCL), ER or MAM fractions of MDA-MB-453 cells, using anti-STAT3 antibodies. Representative of at least 2 independent experiments. **(F)** IP3R3:STAT3 co-immunoprecipitation from whole cell lysates of the indicated cells, using anti-IP3R3 antibodies. Representative of at least 2 independent experiments.

Figure S4.



Analysis of STAT3-IP3R3 interaction. (A) Co-immunoprecipitation of flagged STAT3 proteins with endogenous IP3R3. The map describes the STAT3-FLAG constructs used. N, amino-terminal; CCD, coiled coiled domain; DBD, DNA binding domain; C, carboxy-terminal domain. HEK293 cells were transiently transfected with the indicated constructs, whole cell lysate was immunoprecipitated with, anti-FLAG antibodies and probed with anti-IP3R3 or FLAG antibodies. UC, unrelated flagged control; EV, empty vector; WCL, whole cell lysate. **(B)** Co-immunoprecipitation of flagged or HA-tagged IP3R3 proteins with STAT3-MYC. The map describes the IP3R3 constructs used. Flagged constructs: N and N3, MID; HA-tagged constructs: N, N1, N2, aa 227-602. The indicated constructs were transiently co-transfected with full length STAT-MYC, followed by anti-FLAG (left panel) or anti-HA (right panel) immunoprecipitation and Western blot with the indicated antibodies. EV, empty vector; WCL, whole cell lysate.

Figure S5.



IP3R3 measurements. (A) MDA-MB-468 shCTR cells were serum starved and pre-treated for 2 hours with MG132 prior to serum restimulation (SR) for the indicated times, plus or minus MG132. Whole cell lysates were analysed by Western blot. Representative of four independent experiments. **(B)** IP3R3 RNA levels as measured by Taqman RT-PCR in MDA-MB-468 cells silenced (shS) or not (shC) for STAT3 upon starvation and serum refeeding (SR). Bars are mean ± SEM from 3 independent experiments. **(C)** MDA-MB-453 and MDA-MB-468 cells were starved for 72 hours prior to serum restimulation (SR) for the indicated times. Whole cell lysates were analysed by Western blot. Representative of two independent

experiments. **(D)** MDA-MB-468 cells carrying the Doxycyclin-inducible shRNA against STAT3 were transduced with a lentiviral vector carrying an shRNA-resistant form of flagged SA STAT3, whose expression is shown by Western blot, plus or minus Doxycyclin treatment. **(E)** MDA-MB-453 cells stably overexpressing flagged WT STAT3 as analyzed by Western blot. **(F-G)** MDA-MB-468 cells, silenced (shIP3R3) or not (shSCR) for IP3R3. **(F)** Western blot. **(G)** Cytoplasmic Ca^{2+} release measured upon H_2O_2 treatment. Bars are mean± SEM of 12 measurements from 3 independent experiments.