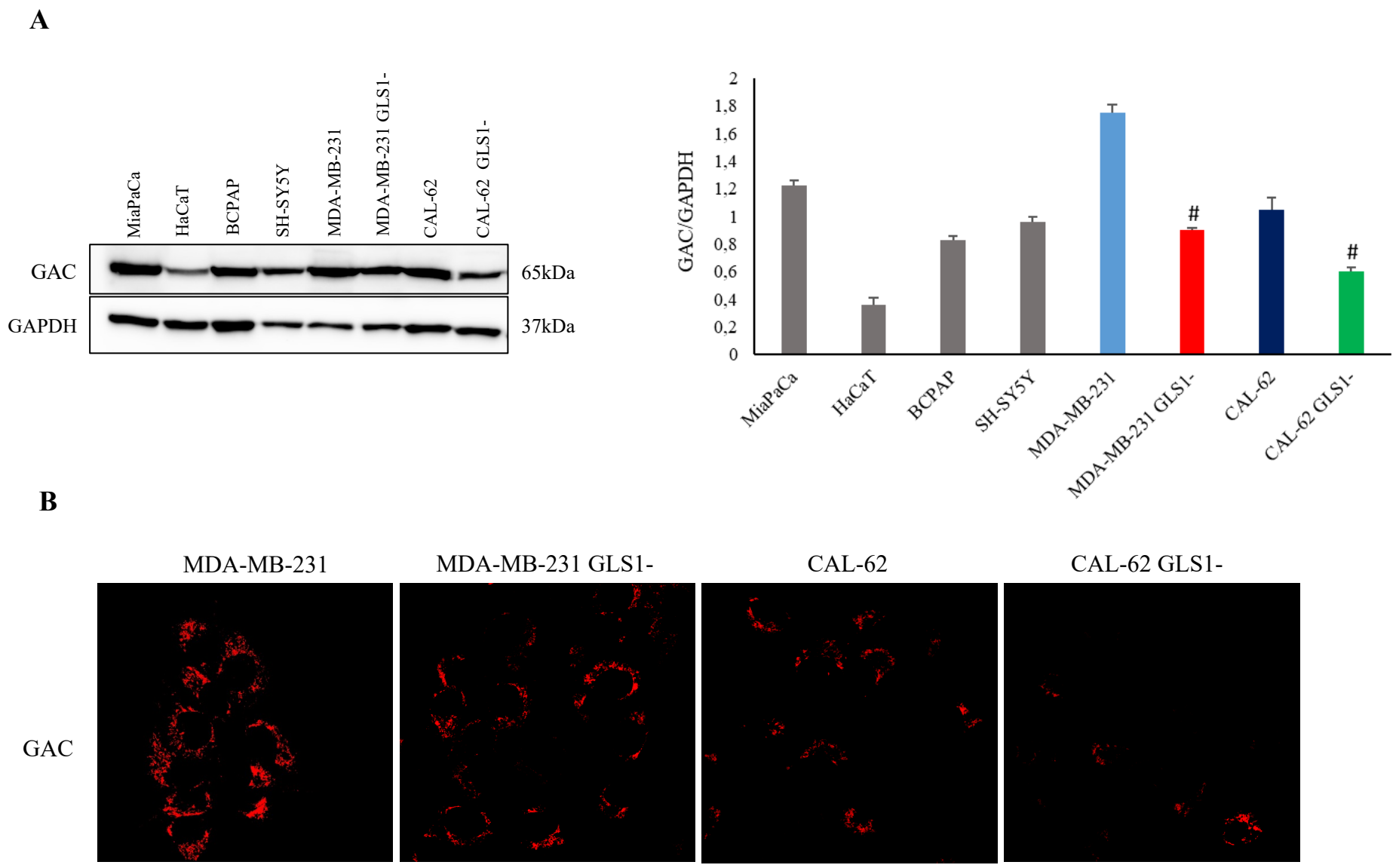

Figure S1. GAC expression in different human cell lines.
(A), Human cancer cell lines (MiaPaCa, BCPAP, SH-SY5Y, MDA-MB-231 and CAL-62) and keratinocytes cell line (HaCaT) were grown as described in Materials and Methods. MDA-MB-231 and CAL-62 cell lines were silenced for GLS1 as described in Materials and Methods. GAC expression was measured by Western blot. GAPDH was used as loading control. GAC expression was normalized with GAPDH and plotted as shown in the graph on the right side. # Significantly decreased compared with wt cells. #, $p < 0.05$.
(B), Confocal immunofluorescence analysis showing GAC expression and localization in MDA-MB-231 and CAL-62 cells and GLS1- clones.

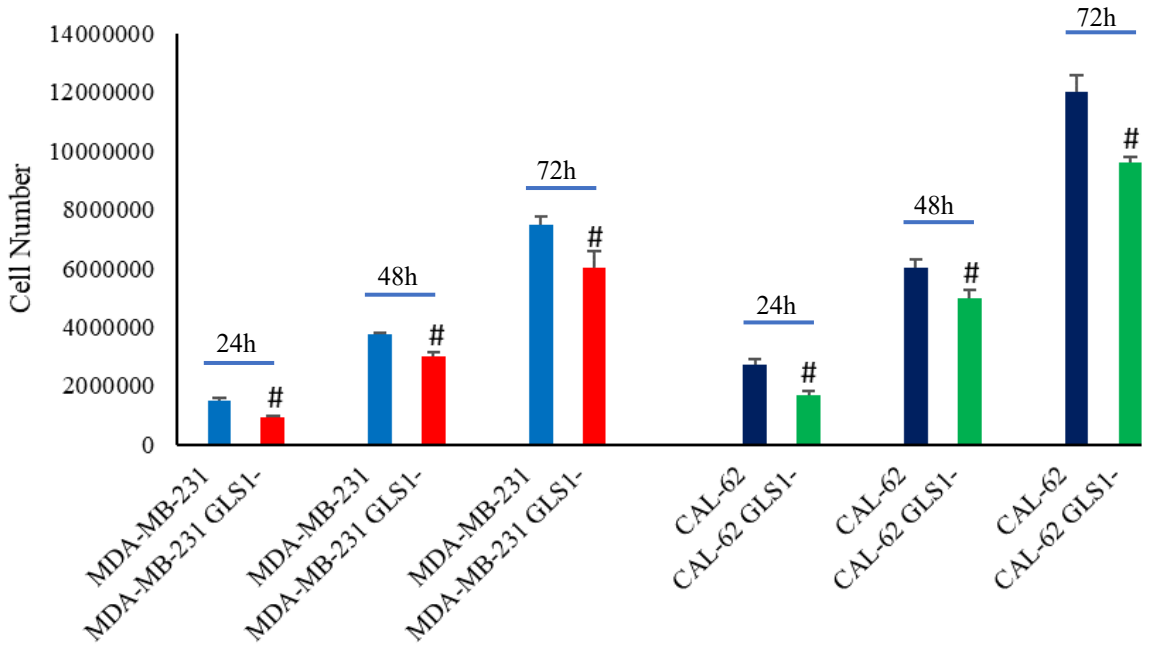

Figure S2. Growth curve of wt and GLS1- MDA-MB-231 and CAL-62 cell lines.
An equal number of wt and GLS1- MDA-MB-231 and CAL-62 cells was plated in 100mm dishes. Growth was assessed by counting living cells after 24, 48 and 72h as described in Materials and Methods. # Significantly decreased compared with the same time of wt cells. #, $p < 0.05$.

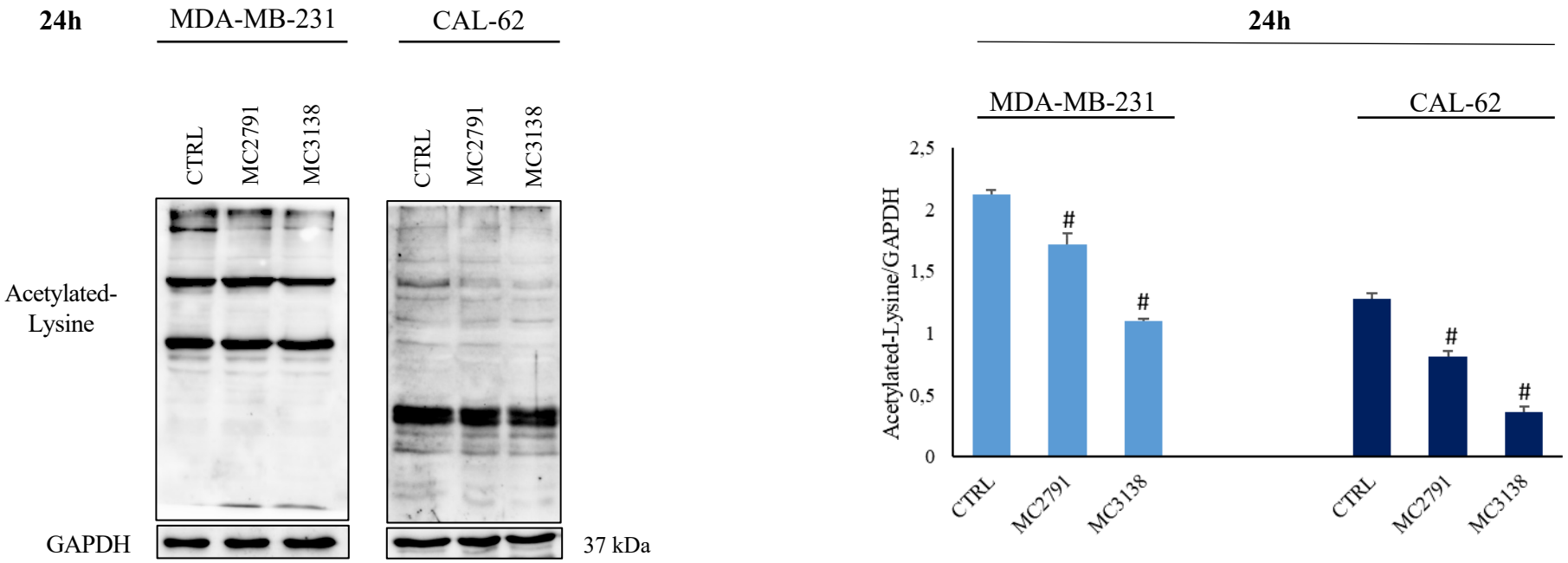

Figure S3. Global lysines acetylation after MC2791 and MC3138 treatment.
MDA-MB-231 and CAL-62 cells were either left untreated or treated with MC2791 (SIRT3 activator) or MC3138 (SIRT5 activator) for 24h. Acetylated lysines were determined as describe in Materials and Methods. GAPDH was used as loading control. Acetylated lysines expression was normalized with GAPDH and plotted as shown in the graph on the right side. # Significantly decreased compared with control cells. #, $p < 0.05$. CTRL, control.

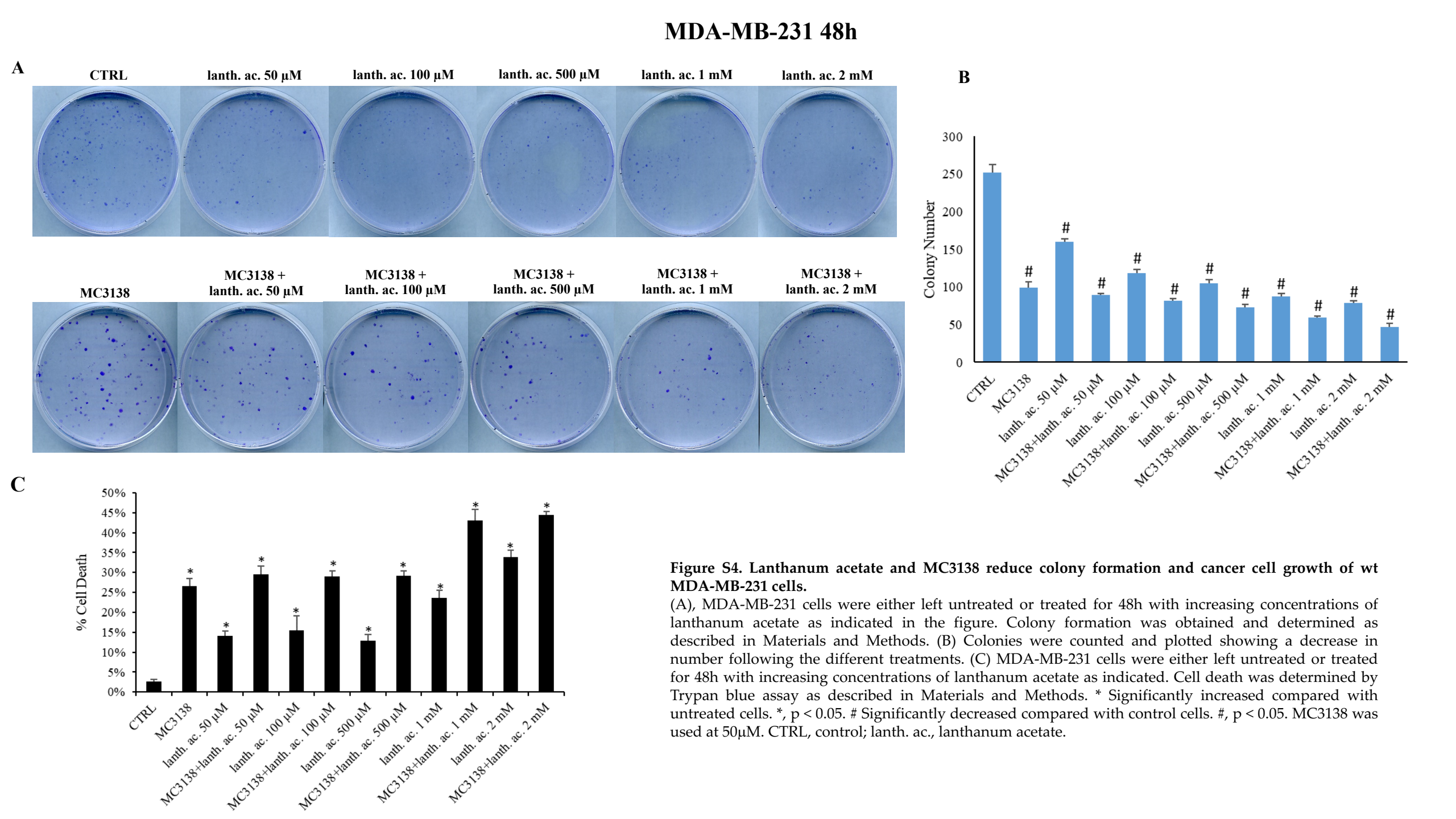

Figure S4. Lanthanum acetate and MC3138 reduce colony formation and cancer cell growth of wt MDA-MB-231 cells.

(A), MDA-MB-231 cells were either left untreated or treated for 48h with increasing concentrations of lanthanum acetate as indicated in the figure. Colony formation was obtained and determined as described in Materials and Methods. (B) Colonies were counted and plotted showing a decrease in number following the different treatments. (C) MDA-MB-231 cells were either left untreated or treated for 48h with increasing concentrations of lanthanum acetate as indicated. Cell death was determined by Trypan blue assay as described in Materials and Methods. * Significantly increased compared with untreated cells. *, $p < 0.05$. # Significantly decreased compared with control cells. #, $p < 0.05$. MC3138 was used at 50µM. CTRL, control; lanth. ac., lanthanum acetate.

MDA-MB-231 GLS1- 48h

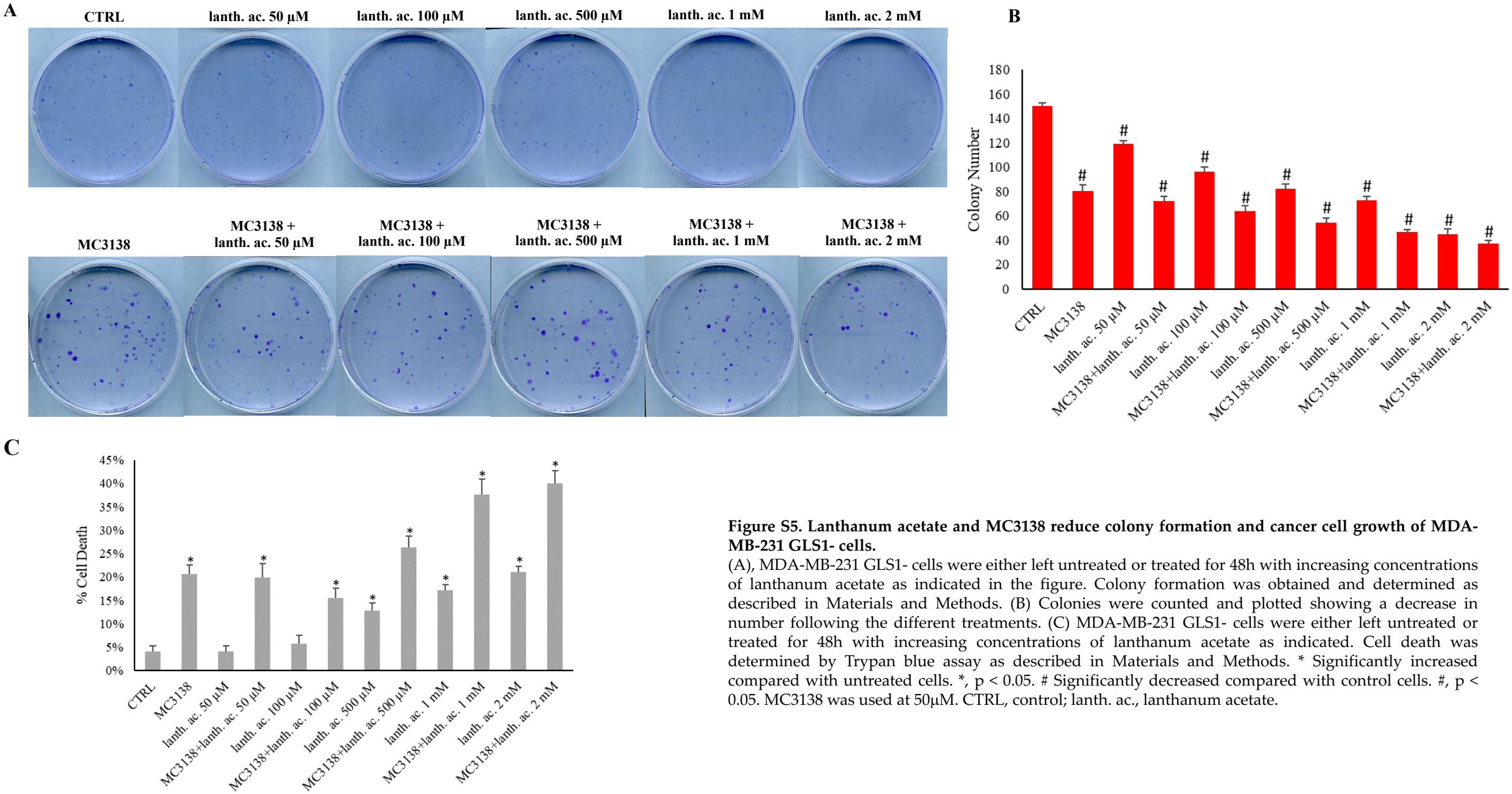

Figure S5. Lanthanum acetate and MC3138 reduce colony formation and cancer cell growth of MDA-MB-231 GLS1- cells.

(A), MDA-MB-231 GLS1- cells were either left untreated or treated for 48h with increasing concentrations of lanthanum acetate as indicated in the figure. Colony formation was obtained and determined as described in Materials and Methods. (B) Colonies were counted and plotted showing a decrease in number following the different treatments. (C) MDA-MB-231 GLS1- cells were either left untreated or treated for 48h with increasing concentrations of lanthanum acetate as indicated. Cell death was determined by Trypan blue assay as described in Materials and Methods. * Significantly increased compared with untreated cells. *, $p < 0.05$. # Significantly decreased compared with control cells. #, $p < 0.05$. MC3138 was used at 50µM. CTRL, control; lanth. ac., lanthanum acetate.

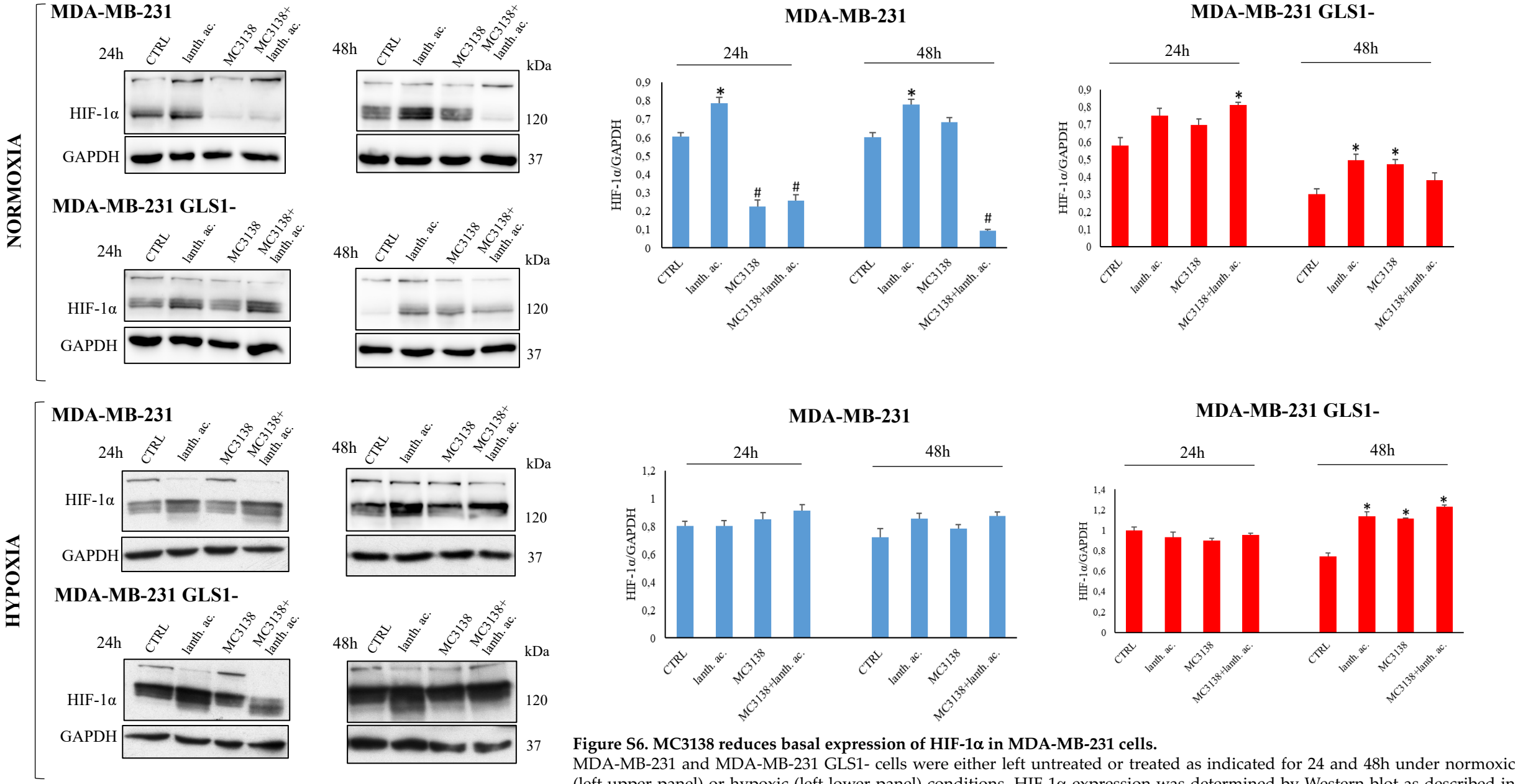

Figure S6. MC3138 reduces basal expression of HIF-1α in MDA-MB-231 cells.
MDA-MB-231 and MDA-MB-231 GLS1- cells were either left untreated or treated as indicated for 24 and 48h under normoxic (left upper panel) or hypoxic (left lower panel) conditions. HIF-1α expression was determined by Western blot as described in Materials and Methods. GAPDH was used as loading control. HIF-1α expression was normalized with GAPDH and plotted as shown in the graph on the upper right side for normoxia and in the lower right side for hypoxia. * Significantly increased compared with untreated cells. *, $p < 0.05$. # Significantly decreased compared with control cells. #, $p < 0.05$. CTRL, control; lanth. ac., lanthanum acetate.

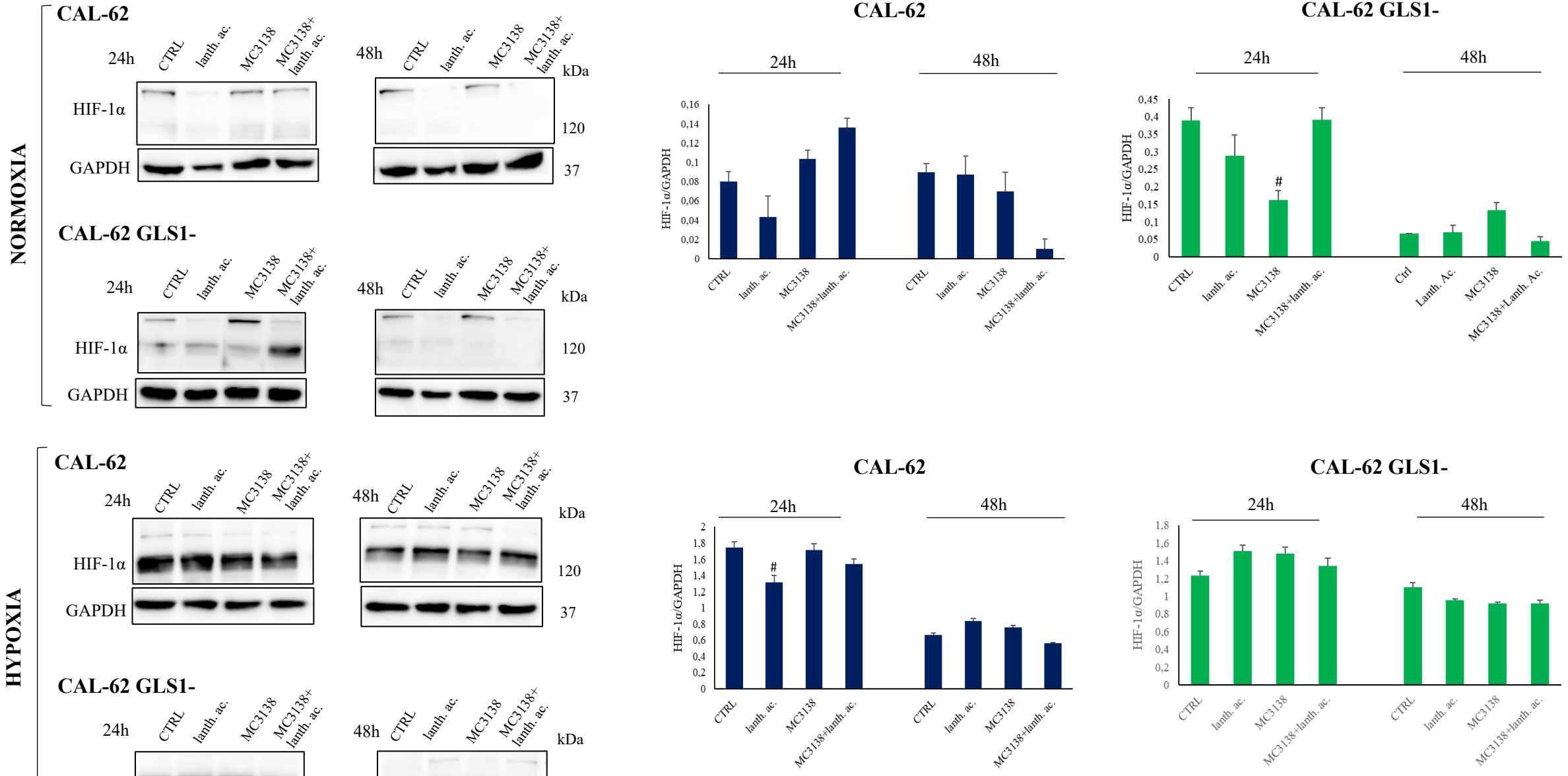

Figure S7. HIF-1α expression in CAL-62 cells.
CAL-62 and CAL-62 GLS1- cells were either left untreated or treated as indicated for 24 and 48h under normoxic (left upper panel) or hypoxic (left lower panel) conditions. HIF-1α expression was determined by Western blot as described in Materials and Methods. GAPDH was used as loading control. HIF-1α expression was normalized with GAPDH and plotted as shown in the graph on the upper right side for normoxia and in the lower right side for hypoxia. * Significantly increased compared with untreated cells. *, $p < 0.05$. CTRL, control; lanth. ac., lanthanum acetate.