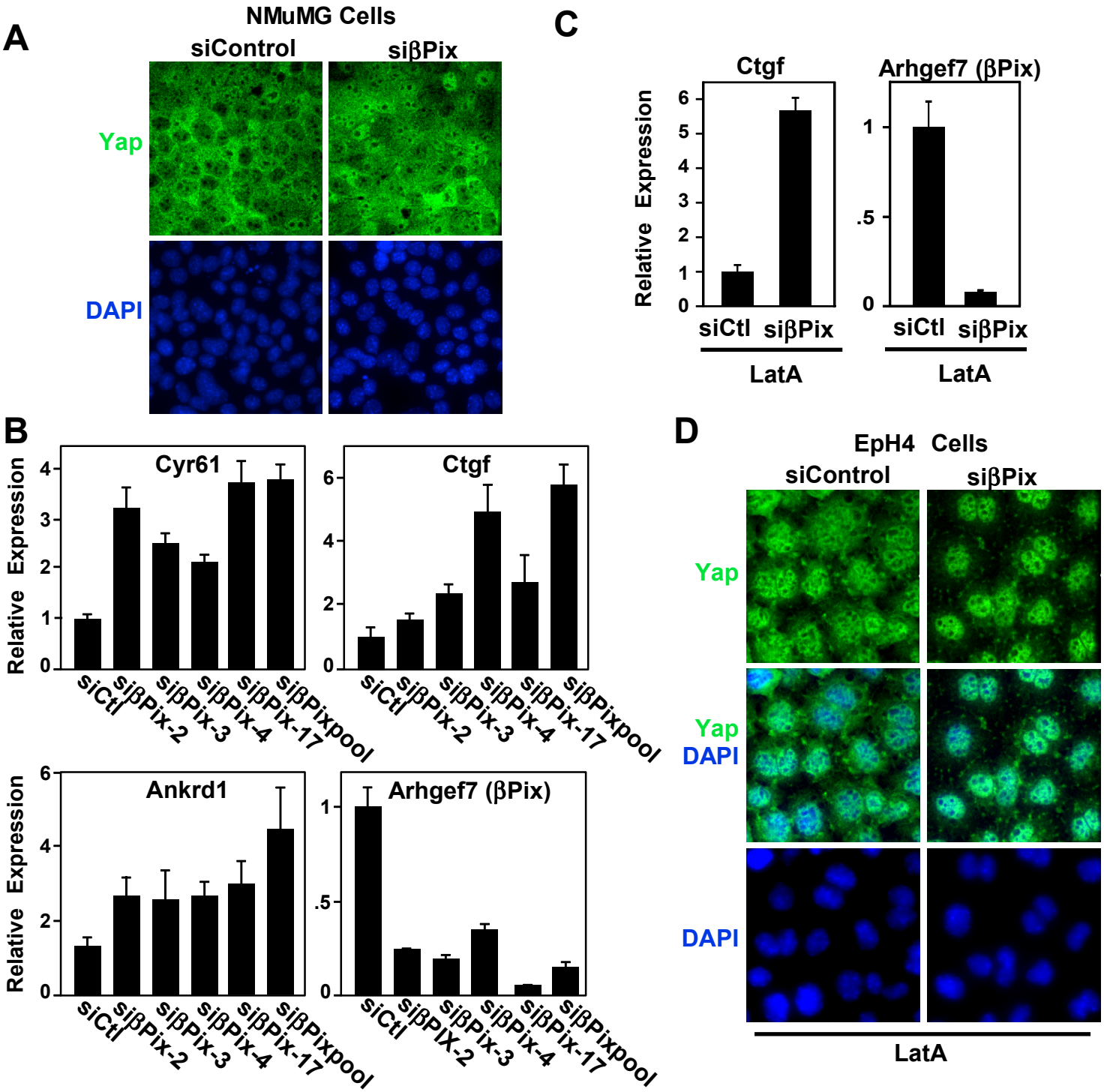
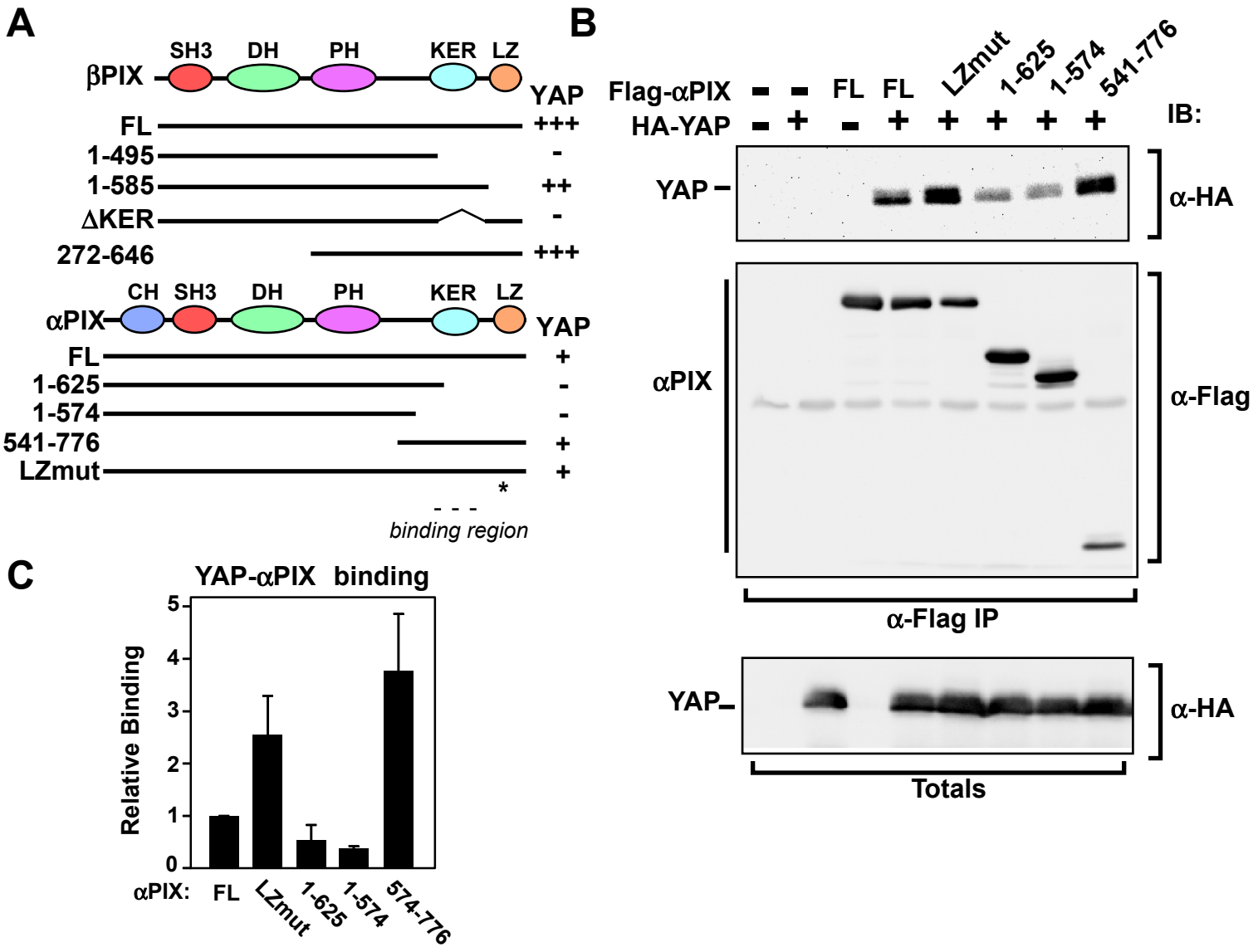
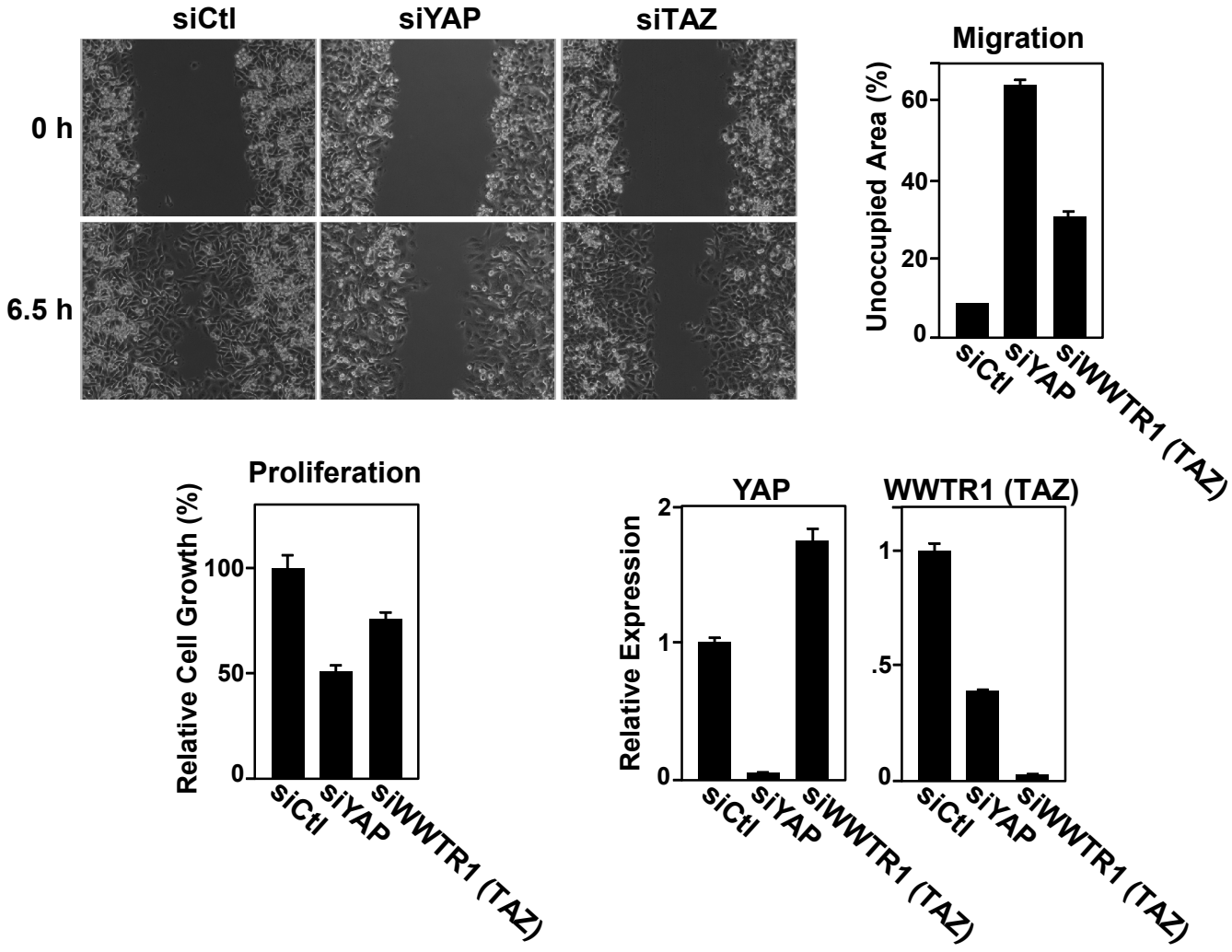


Supplementary Figure 1. Loss of βPix promotes Yap activity.

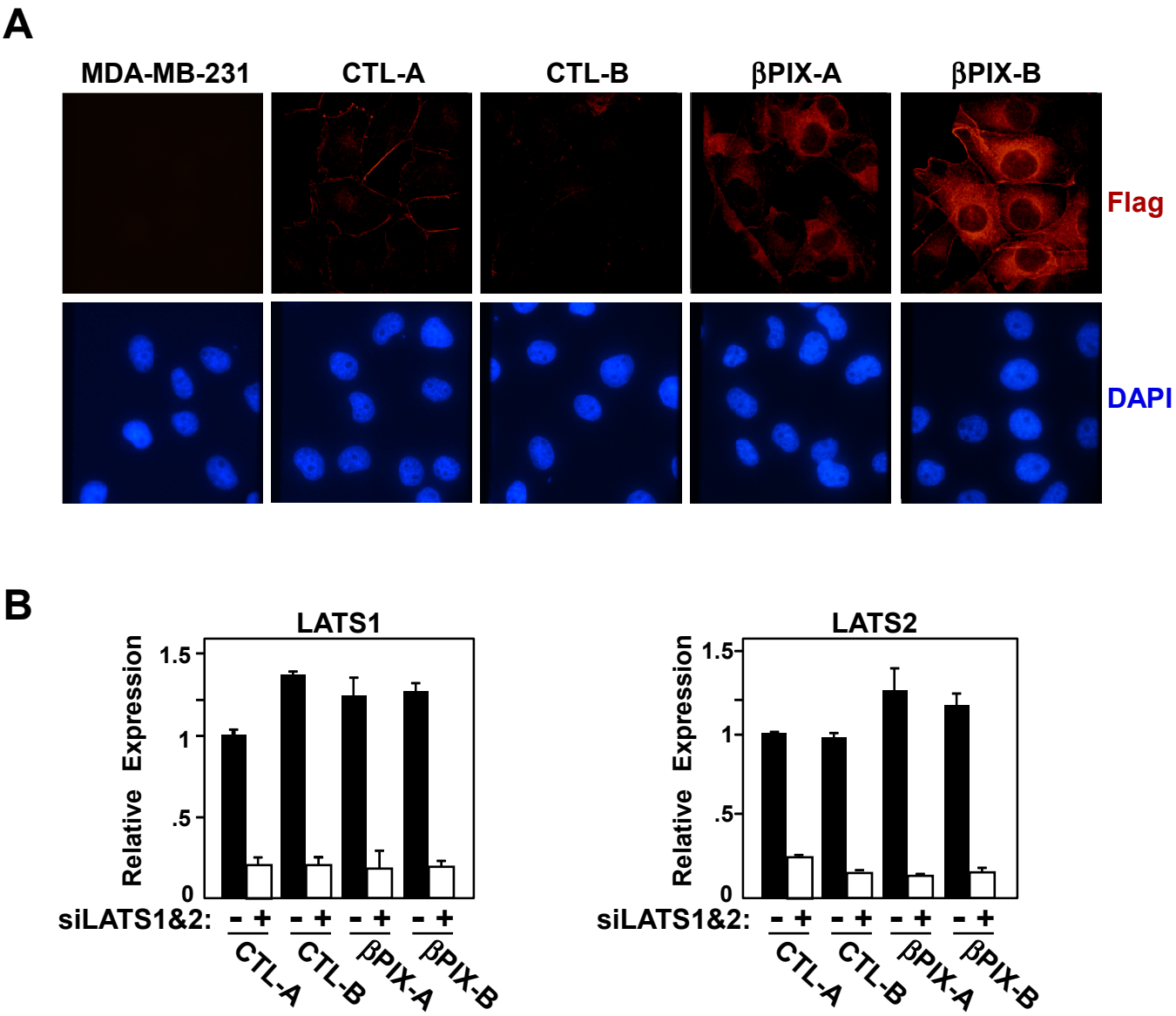
(A) βPix regulates Yap localization in NMuMG cells at high cell density. NMuMG cells were transfected with control siRNA or siRNA targeting βPix and were plated at high cell density. After 48 h, Yap localization was visualized by immunofluorescence confocal microscopy. **(B)** Deconvolution of βPix siRNAs. NMuMG cells were transfected with single siβPix oligonucleotides that comprised the pool. The expression of Yap/Taz target genes and βPix knockdown efficiency was determined by qPCR. Data from 2 independent experiments is plotted as average fold over no siRNA ± the range. **(C,D)** βPix regulates Yap/Taz activity in response to actin disruption. EpH4 cells transfected with siCTL or siβPix were treated with LatA. **(C)** Relative expression of Ctgf and the knockdown efficiency of βPix was determined by qPCR. **(D)** Yap localization was visualized by immunofluorescence confocal microscopy.



Supplementary Figure 2. Mapping the Interaction of YAP and αPIX and determining LATS localization. **(A)** A schematic depicting the different αPIX and βPIX cDNA constructs used for mapping interactions is shown. Positive or negative interactions with YAP are indicated on the right. **(B)** Interaction mapping between HA-YAP and Flag-αPIX WT and mutant constructs. Cells were co-transfected with wild-type or mutant constructs of Flag-αPIX along with HA-YAP and cell lysates were subject to anti-Flag IP. The presence of YAP was determined by anti-HA immunoblotting. **(C)** Quantitation of the interaction mapping of YAP to αPIX from replicate experiments is plotted. **(D)** Flag-LATS1 is cytoplasmically-localized in NMuMg cells. Localization of Flag-LATS1 in transiently-transfected NMuMG cells was determined byconfocal microscopy.



Supplementary Figure 3. YAP and TAZ promote cell proliferation and migration in MDA-MB-231 cells. Cells were transfected with control siRNA or siRNA targeting YAP, or TAZ (WWTR1) and the effect on cell proliferation or migration was determined by a SRB or wound healing assay, respectively. Knockdown efficiency was confirmed by qPCR. A representative experiment is shown.



Supplementary Figure 4. Analysis of MDA-MB-231 stable cell clones.

Expression of Flag-βPIX in MDA-MB-231 cells.

(A) MDA-MB-231 cell clones stably transfected with empty vector (CTL-A, CTL-B) or Flag-βPIX (βPIX-A, βPIX-B) were fixed and βPIX expression and localization was confirmed by immunofluroresence confocal microscopy **(B)** Analysis of LATS1 and LATS2 knockdown efficiency associated with Figure 7. MDA-MB-231 parentals, control (CTL) or βPIX expressing stable clones were transfected with control siRNA or siRNAs target_ing both LATS1 and LATS2. Knockdown efficiency of LATS1 and LATS2 was confirmed by qPCR.