Supplementary Figure 1. Simultaneous inhibition of JAK2-STAT3 and SMO signaling pathways suppresses the BT474-Ttzmr and TNBC cancer stem cell population. A) Representative flow cytometry scatter plots of CD44+/CD24- BT474-Ttzmr cells. B) Pacritinib-Sonidegib combination treatment suppressed the CD44+/CD24- population. C) Pacritinib-Sonidegib combination treatment reduced TNBC MDA-468 mammosphere formation. D) Representative images of MDA-468 mammospheres. Images were taken under 5X magnification. Scale bar represents 200 μm. Data are presented as mean ± SEM of 3-5 experimental replicates. One-way ANOVA with Tukey’s multiple comparison post hoc test was used to compute p values. *, p < 0.05; ***, p < 0.001.
Supplementary Figure 2. Pacritinib-Sonidegib combination treatment does not disrupt the STAT3-GLI1/tGLI1 interaction. STAT3 similarly interacted with GLI1 and tGLI1 regardless of treatment as shown by immunoprecipitation western blot using MDA-231 cells co-transfected with STAT3 and either Flag-GLI1 or Flag-tGLI1.
| Gene  | Forward sequence (5’-3’) | Reverse Sequence (3’-5’) |
|-------|--------------------------|-------------------------|
| Bcl-2 | TGTTAATATCAGTCTACTTCTCTCTGTG | GCTATAACTGGAGAGTGCTGAA |
| Nanog | CTAAGAGGTGGCAGAAAAACA | CTGGTGTTAGGAAGAGTAAAGG |
| VEGFA | TGTCTAATGCCCTGGAGCCT | TTAACTCAAGCTGCCTCGCC |
| GAPDH | ACTGCCAACGTGCAGTG | GTGTCGCTGTTGAAAGTCAGA |