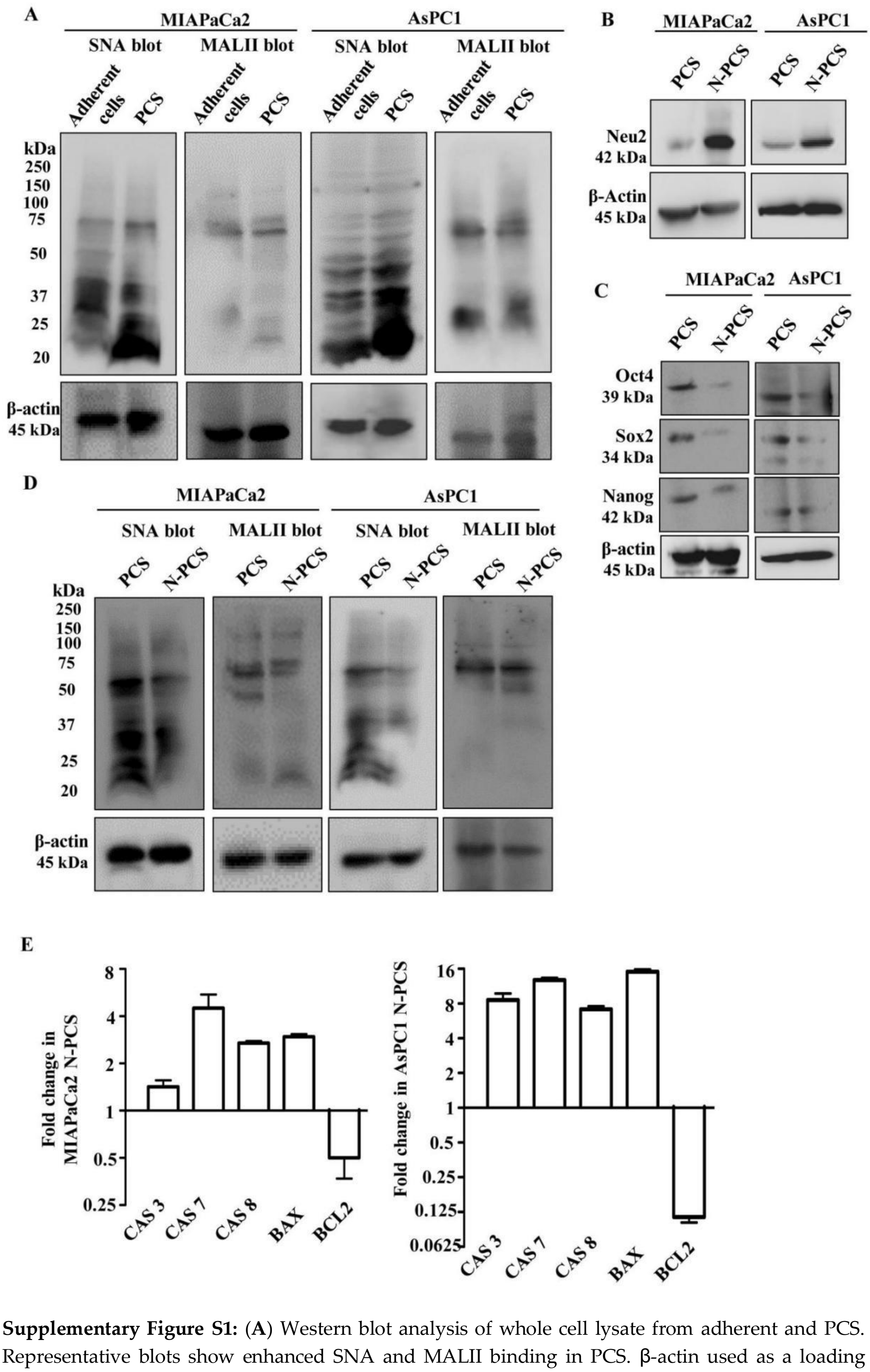
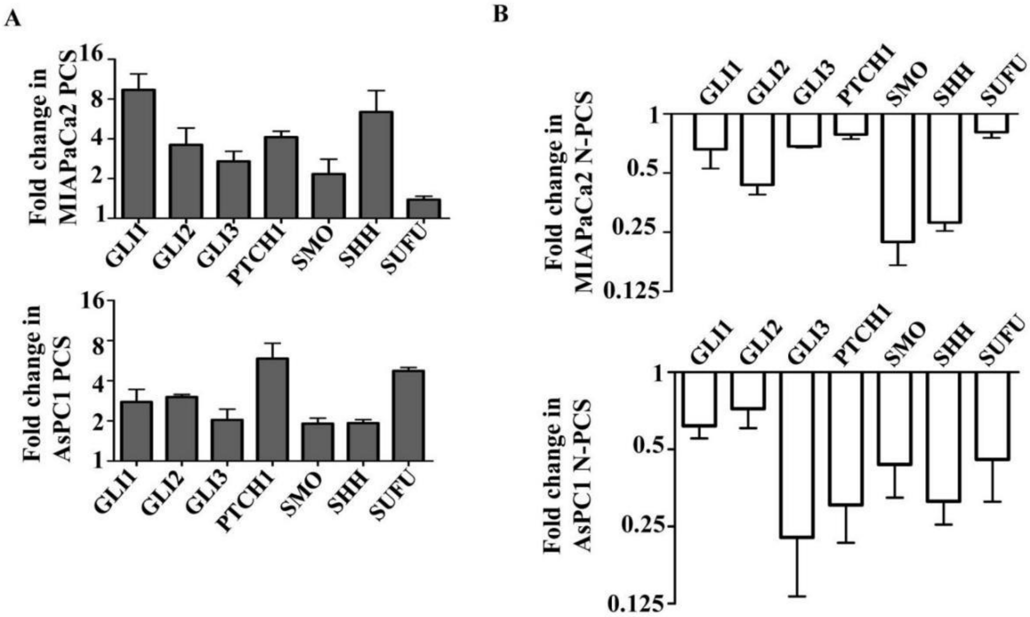
S1



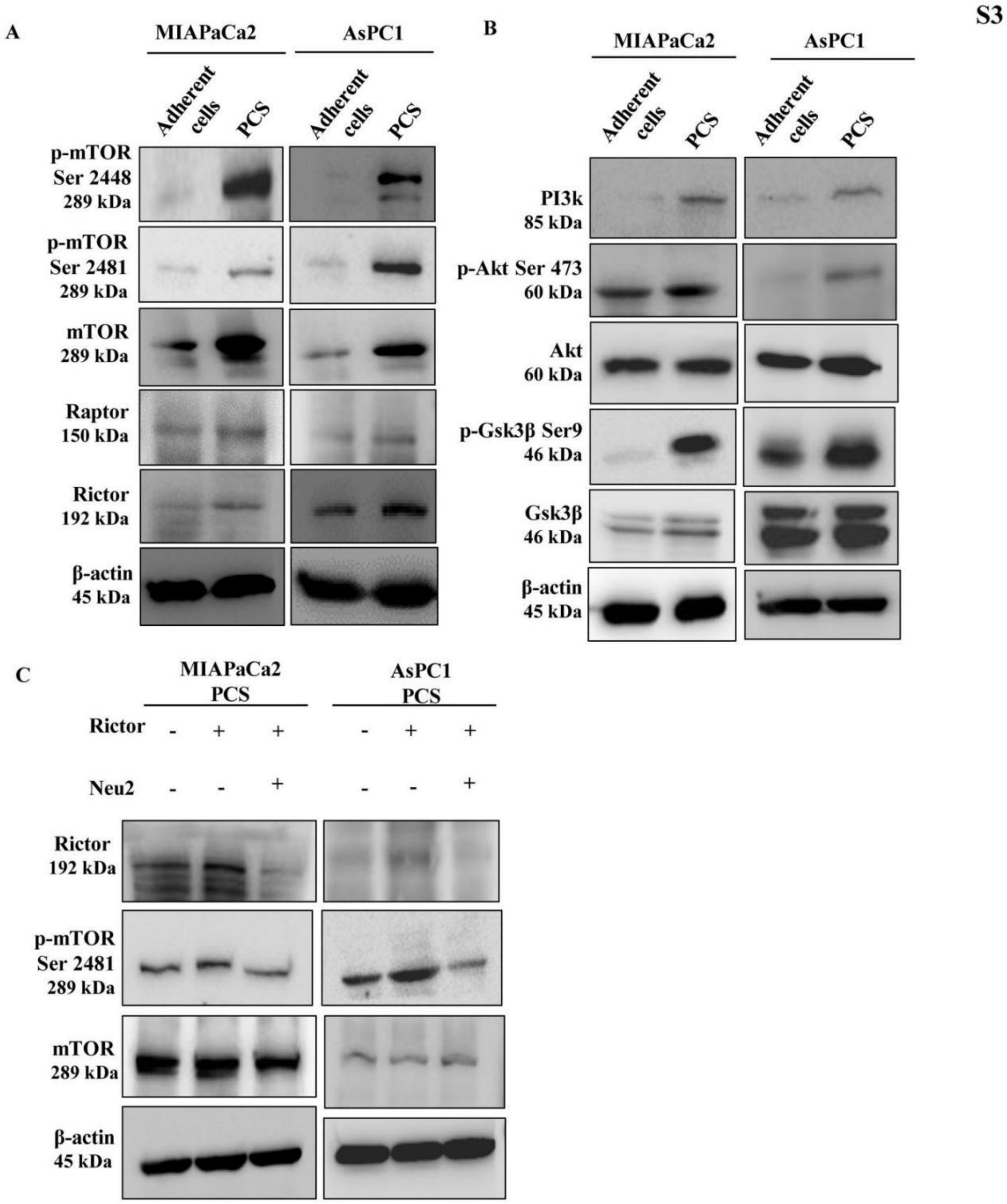

**Supplementary Figure S1:** (**A**) Western blot analysis of whole cell lysate from adherent and PCS. Representative blots show enhanced SNA and MALII binding in PCS. β-actin used as a loading control. (**B**) Representative immunoblots exhibited higher expression of Neu2 upon overexpression in PCS from both MIAPaCa2 and AsPC1 cells. β-actin used as a loading control. (**C**) Western blot analysis demonstrated reduced expression of pluripotent stem cell markers Oct4, Sox2 and Nanog in Neu2-overexpressed PCS. β-actin used as a loading control. (**D**) Western blot analysis of whole-cell lysate from PCS and N-PCS. Representative blots show decreased SNA and MALII binding in

N-PCS. β-actin used as a loading control. **(E)** qPCR analysis shows higher expression of pro-apoptotic genes such as CAS 3, 7, 8 and BAX and reduced expression of anti-apoptotic BCL2 in Neu2-overexpressed PCS from both MIAPaCa2 and AsPC1 cells.

S2




**Supplementary Figure S2:** (**A**) qPCR data represent an enhanced expression of Hedgehog pathway genes such as PTCH1, GLI1, GLI2, GLI3, SMO, SHH and SUFU in pancreatic cancer sphere-forming cells generated from both MIAPaCa2 and AsPC1 cells. (**B**) qPCR analysis showed reduced expression of Hedgehog pathway genes such as PTCH1, GLI1, GLI2, GLI3, SHH, SUFU and SMO in Neu2-overexpressed PCS generated from both MIAPaCa2 and AsPC1 cells.




**Supplementary Figure S3.:** (**A**) Representative immunoblots exhibited enhanced mTORC1/2 formation and higher expression of Rictor and Raptor in PCS from both cell lines. β-actin used as a loading control. (**B**) Western blots analysis show inhibition of Gsk3β and activation of Akt in PCS from both cell lines. β-actin used as a loading control. (**C**) Representative immunoblots exhibited enhanced mTORC2 formation and Rictor expression at protein levels in Rictor-overexpressed PCS whereas reduced expression of these molecules was found in Rictor and Neu2 co-overexpressed PCS.

**Supplementary Table 1: List of Primers.**

| NAME | FORWARD PRIMER (5′-3′) | REVERSE PRIMER (5′-3′) |
|---|---|---|
| OCT 4 | GAGGAGTCCCAGGACATCAA | ACACTCGGACCACATCCTTC |
| SOX 2 | AGAACCCCAAGATGCACAAC | ATGTAGGTCTGCGAGCTGGT |
| NANOG | ACCAGACCCAGAACATCCAG | CTCGTCGATTAGGCTCCAAC |
| CD133 | TTGTGGCAAATCACCAGGTA | TCAGATCTGTGAACGCCTTG |
| GLI1 | GTGCAAGTCAAGCCAGAACA | ATAGGGGCCTGACTGGAGAT |
| GLI2 | TTTATGGGCATCCTCTCTGG | AAGGCTGGAAAGCACTGTGT |
| GLI 3 | TCCCAGCGCTTTCTACATCT | CTTTGTCGTGGACCCATTCT |
| PTCH1 | TCCCAGCGCTTTCTACATCT | CTTTGTCGTGGACCCATTCT |
| SHH | TACTCGCAGCTGCTCTACCA | TGTCTTTTTGCTTTGCGTTG |
| SMO | CAACCTGTTTGCCATGTTTG | TTTGGCTCATCGTCACTCTG |
| Neu1 | GGAGCAAGGATGATGGTGTT | CATGATCATCGCTGAGGAGA |
| Neu2 | CCTCTTCTTCATTGCCATCC | GTCGTGAAGCTGCAAACAAT |
| Neu3 | CAGATTGTGTCAGGCAGGAA | AGCTGGAAGCAAAAGAACCA |
| Neu4 | CACCGTCTTCCTCTTCTTCA | GGCAGATCTTGCCAAAACA |
| Caspase 8 | AGACTGATTCAGAGGAGCAA | GTGACTGGATGTACCAGGTT |
| Caspase 7 | GATTTGACAGCCCACTTTAG | TTCCACTGGGATCTTGTATC |
| Caspase 3 | AGGAGCAGTTTTGTTTGTGT | CAGGCCTGAATAATGAAAAG |
| Bax | TCACTGAAGCGACTGATGT | GAGGAAAAACACAGTCCAAG |
| Bcl2 | ATTGTGGCCTTCTTTGAGT | TACAGTTCCACAAAGGCATC |
| 18s rRNA | GCTCATTAAATCAGTTATGG | ACTACCATCGAAAGTTGATA |