Supplemental Information

Tetraspanin TM4SF5 mediates epithelial-mesenchymal transition leading to loss of contact inhibition

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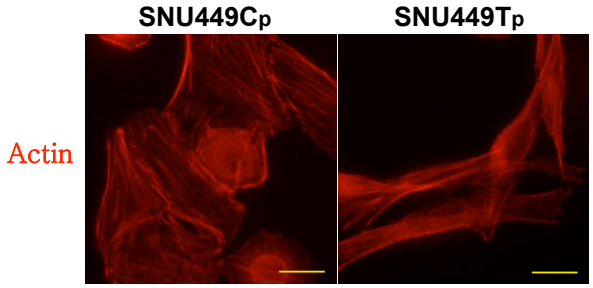
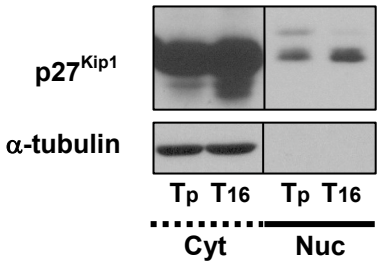
Supplemental Figures

Figure S1. Differential actin organization in TM4SF5-null or -expressing cells.



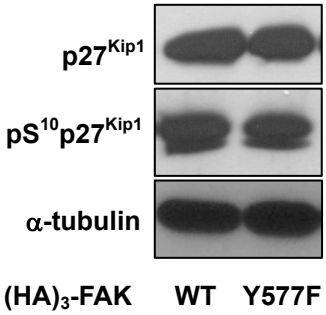
Control (SNU449Cp) or TM4SF5-expressing (SNU449Tp) cells on glass coverslips precoated with 10% FBS/RPMI-1640 were stained with phalloidin-conjugated with rhodamine. Scale bar for 20 μm.

Figure S2. Dominant $p27^{Kip1}$ level in cytosol.



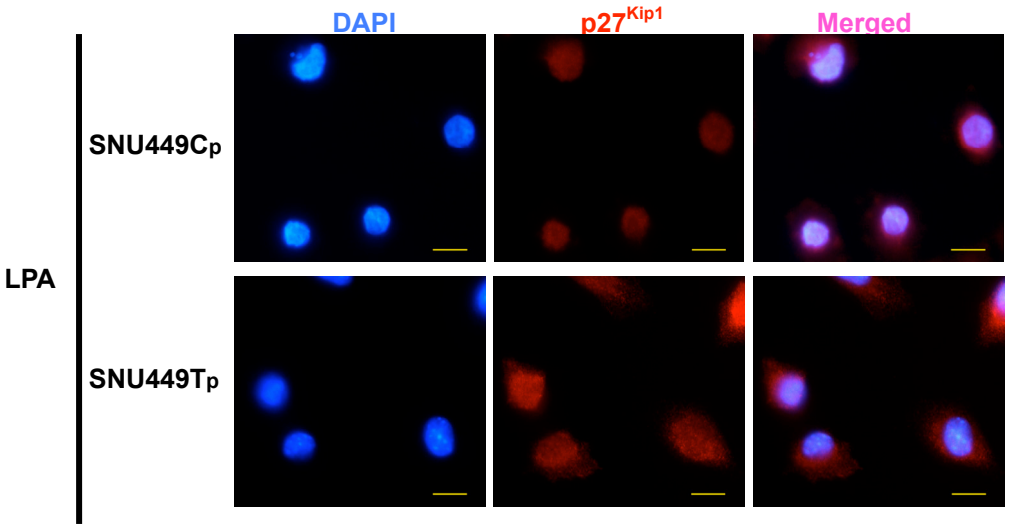
Nucleus and cytosol fractions were prepared from subconfluent SNU449Tp and SNU449T16 cells, as described in Supplemental Experimental Procedures, prior to immunoblotts against anti-$p27^{Kip1}$ or α-tubulin antibody.

Figure S3. Y577F FAK mutation did not affect $p27^{Kip1}$ level and Ser10 phosphorylation in SNU449Tp cells.



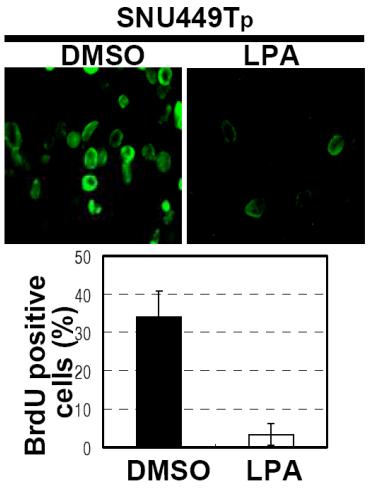
Whole cell lysates prepared from SNU449Tp cells transfected with $(HA)_3$-tagged FAK WT or Y577F mutant, as in Figure 2H, were immunoblotted for indicated molecules.

Figure S4. Reduction of cytosolic $p27^{Kip1}$ in SNU449Tp cells via lysophosphatidic acid (LPA) treatment.



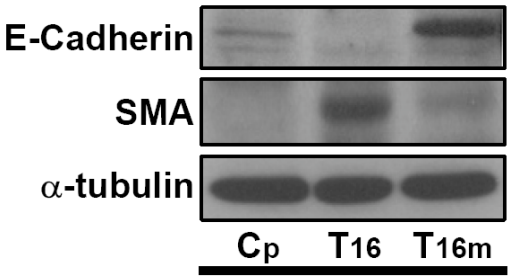
SNU449Cp and SNU449Tp cells on coverslips precoated with 10% FBS/RPMI-1640 were treated with 10 mM LPA for 1 h, before double-staining for DAPI and $p27^{Kip1}$, as in Figure 3B. Note that LPA-treated SNU449Tp cells showed retracted morphology with less $p27^{Kip1}$ in cytoplasm, compared to untreated cells shown in Figure 3B (bottom). Scale bar for 20 μm.

Figure S5. TM4SF5-enhanced S-phase progression is blocked by LPA treatment.



SNU449Tp cells treated with DMSO or LPA were analyzed for S-phase progression via BrdU incorporation assay, as described in Materials and Methods.

Figure S6. Reconstitution of E-cadherin in TM4SF5-expressing cells.



SNU449Cp (Cp), SNU449T16 (T16), and SNU449T16m (T16m) cells were harvested for whole cell lysates, prior to immunoblots for indicated molecules.