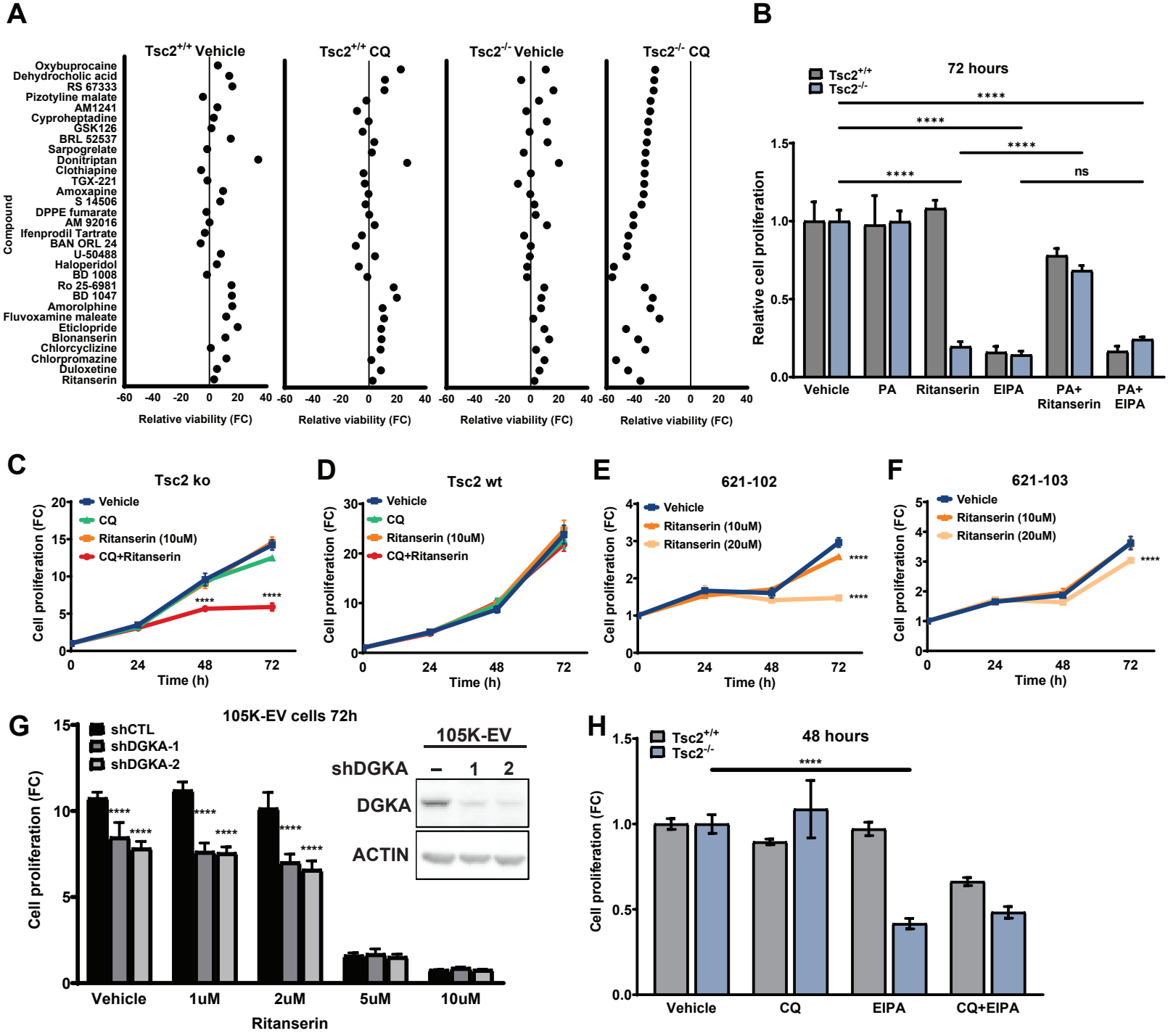
Supplementary Figure 1



Supplementary Figure 1. Ritanserin selectively inhibits proliferation of TSC2-deficient cells by depletion of phosphatidic acid. (A) Identified compounds that selectively inhibit the viability of Tsc2$^{-/-}$ MEFs upon CQ treatment. Viability is represented as fold change (FC) relative to untreated cells. **(B)** Supplementation with PA (100uM) rescues the proliferation of Tsc2$^{-/-}$ MEFs treated with ritanserin (20uM; 72 hours) but not EIPA treated cells (6uM; 72 hours). **(C, D)** CQ (5uM) and ritanserin (10uM) inhibit the proliferation Tsc2-deficient embryonic fibroblasts (MEFs) derived from Tsc2$^{fl/fl}$ Rosa26-CreERT2 mice (Tsc2ko) compared to Tsc2-expressing MEFs (Tsc2wt). **(E, F)** Ritanserin inhibits the proliferation of TSC2-deficient human kidney-derived angiomyolipoma cells in a dose-dependent manner. **(G)** Ritanserin (1-10uM; 72 hours) did not impact the proliferation of shDGKA Tsc2-deficient 105K cells compared to shCtl cells. Immunoblotting confirming DGKA knockdown in 105K cells. **(H)** Macropinocytosis inhibitor EIPA (6uM; 48 hours) selectively inhibits the proliferation of Tsc2$^{-/-}$ MEFs. Data represented as mean +/− standard deviation of six biological replicates. Proliferation was quantified using crystal violet staining. Values are shown as fold-change (FC) normalized to the day of seeding. Statistical significance was assessed using two-way and one-way ANOVAs with Bonferroni correction with ****p < 0.0001.