Supplemental Figure 1

A. Mouse embryonic fibroblast (MEF) cells expressing GFP-tagged Akt-PH domain (a generous gift from Prof. Takehito Sasaki, Akita University School of Medicine, Akita, Japan) were cultured in medium as described for NRK52E cells. For TGFβ1 stimulation experiments, MEF cells (2×10^5) cultured in 6-well plates were made quiescent by culturing in medium containing 0.1 % (v/v) foetal calf serum for 24 hrs prior to 10 ng/ml TGFβ1 treatment. Cells were washed with 1 x PBS and changed to serum free medium containing control diluent or TGFβ1 (10 ng/ml) for 60 min at 37 ^{0}C.

B. NRK52E cells were incubated in the presence or serum (*FBS*) or serum starved overnight and treated with vehicle (*DMSO*) or 10 ng/ml TGFβ1 in the absence or presence of 20 mM LY294002. After 24 h, lipids were extracted and assayed for PIP3 levels using the PIP3 Mass ELISA kit. Absorbance values at 450 nm were converted to pmol of PIP3 and plotted. *, $p<0.05$.