SUPPLEMENTAL FIGURES

FIGURE LEGENDS

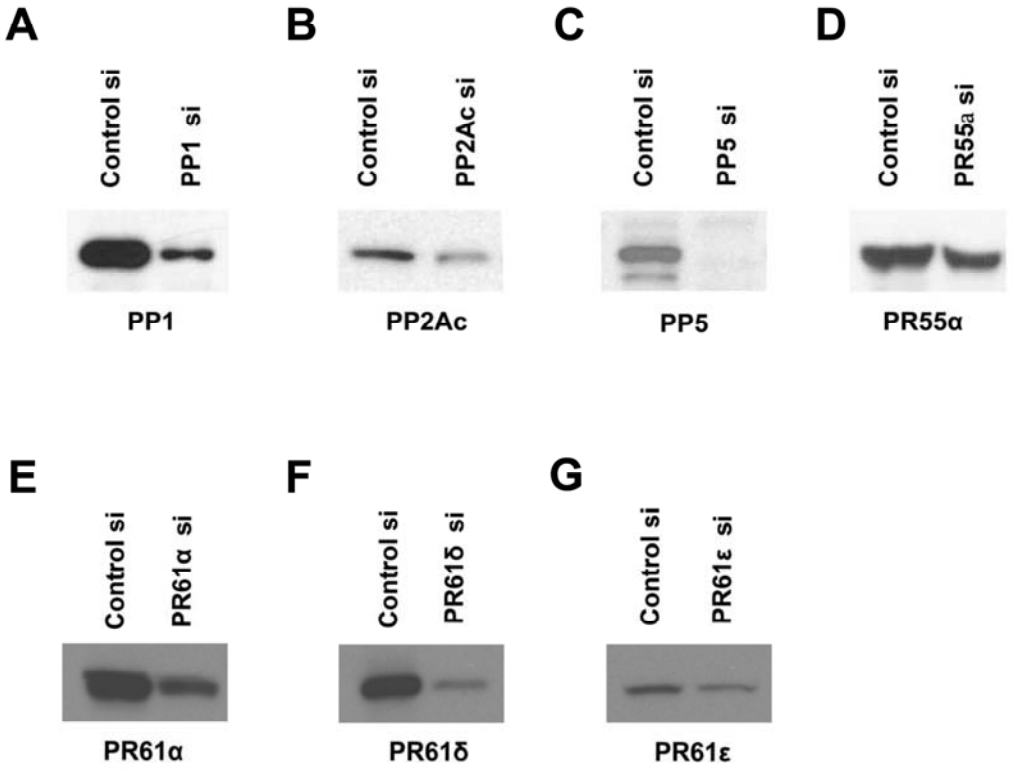
Supplemental Fig. 1. Effects of siRNAs. siRNAs for PP1, PP2Ac, PP5, PR55α, PR61α, PR61δ and PR61ε were transfected into HEK293T cells. The effects of these siRNAs on endogenous proteins (PP2Ac and PR55α) or overexpressed proteins were analyzed by Western blot.

Supplemental Fig. 2. Activities of PP1, PP2A and PP5. Equal amount of Phosphorylated β-catenin was incubated with purified PP1, PP2A and PP5 for 2h. Phosphatase buffer alone was used as a control. β-catenin was analyzed by Western blot using Abs that recognize total β-catenin or phosphorylated β-catenin.

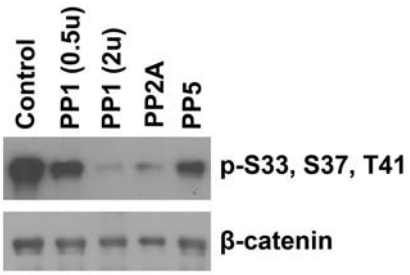
Supplemental Fig. 3. Screening of PR55α shRNAs. HEK293T cells were cotransfected with a Myc-tagged PR55α plasmid and each of these shRNA constructs. PR55α protein was analyzed by Western blot using an anti-Myc Ab. The results from duplicated experiments suggest that TRCN0000002492 and TRCN0000002493 can efficiently knock down PR55α. TRCN0000002493 was used for generating lentivirus and stable cell lines.

Supplemental Fig. 4. Coomassie staining of GST fusion proteins. GST, GST-β-catenin, GST-Axin(Cat+S45K) and GST-Axin(G3+Cat) were purified from E.coli BL21 (DE3) using Glutathione beads. These proteins were used in GST pull-down assays for protein-protein interactions.

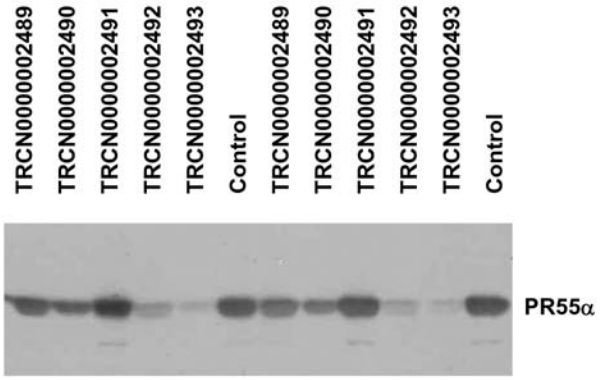
Supplemental Fig. 5. Axin (Cat+S45K) but not Axin (Cat) alone does binds PR55α. Flag-tagged PR55α was cotransfected into HEK293T cells with Myc-tagged Axin (Cat+S45K) or Axin (Cat). These Axin constructs have been described previously (4). Axin fragments were immunoprecipitated with 9E10 beads and PR55α was analyzed by western blot with polyclonal anti-Flag Ab.



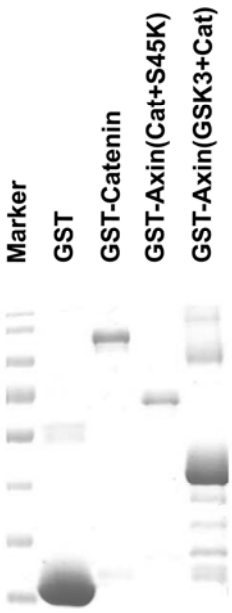
Supplemental Figure 1



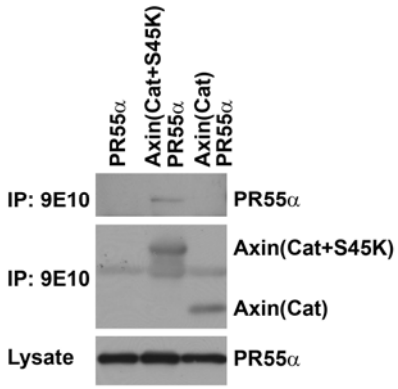
Supplemental Figure 2



Supplemental Figure 3



Supplemental Figure 4



Supplemental Figure 5