

Figure S1. PDGF significantly increases Akt/mTORC1 activity compared to TGFβ alone. Mesangial cells were incubated with TGFβ (2 ng/ml) or PDGF (20 ng/ml) for 2 hours. The cell lysates were immunoblotted with indicated antibodies.

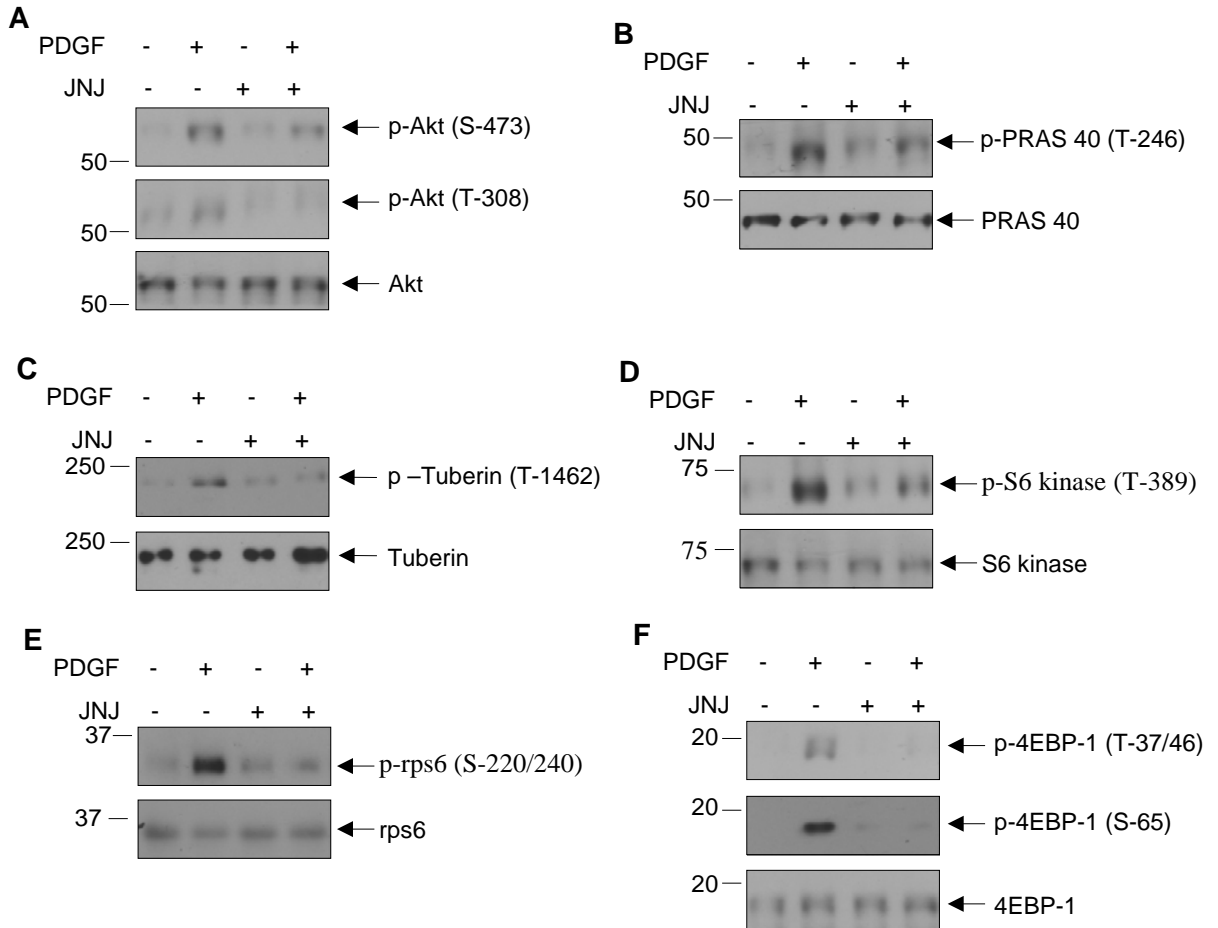


Figure S2. Inhibition of PDGFR β blocks Akt/mTORC1 activity. Mesangial cells were treated with 0.1 μ M JNJ for one hour prior to incubation with PDGF (20 ng/ml) for 2 hours. The cell lysates were immunoblotted with indicated antibodies.

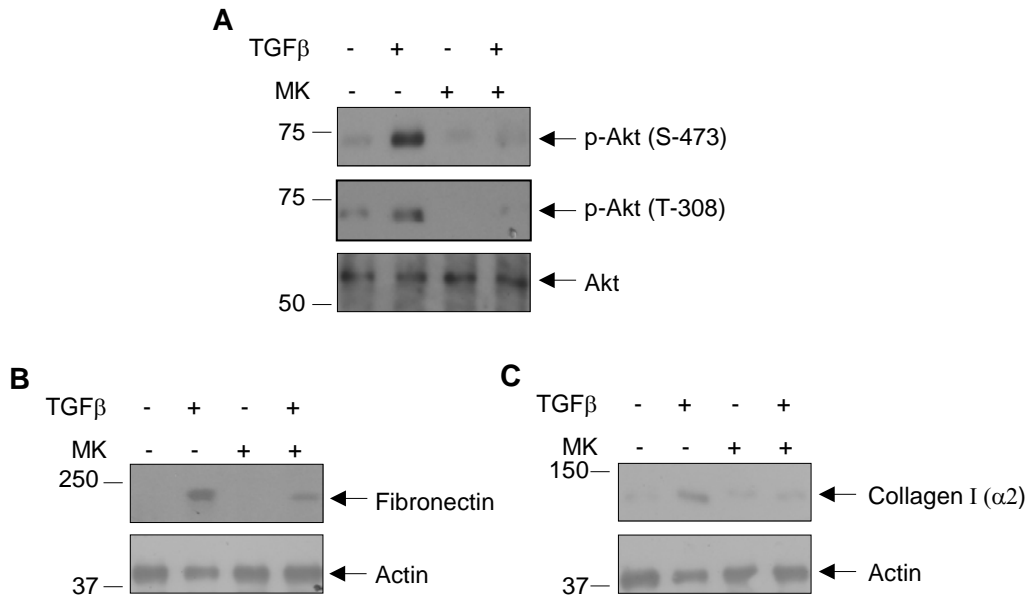


Figure S3. Inhibition of Akt blocks TGF β -induced fibronectin and collagen I (α 2) expression. Mesangial cells were treated with 1 μ M MK 2206 (MK) prior to incubation with TGF β . The cell lysates were immunoblotted with indicated antibodies.

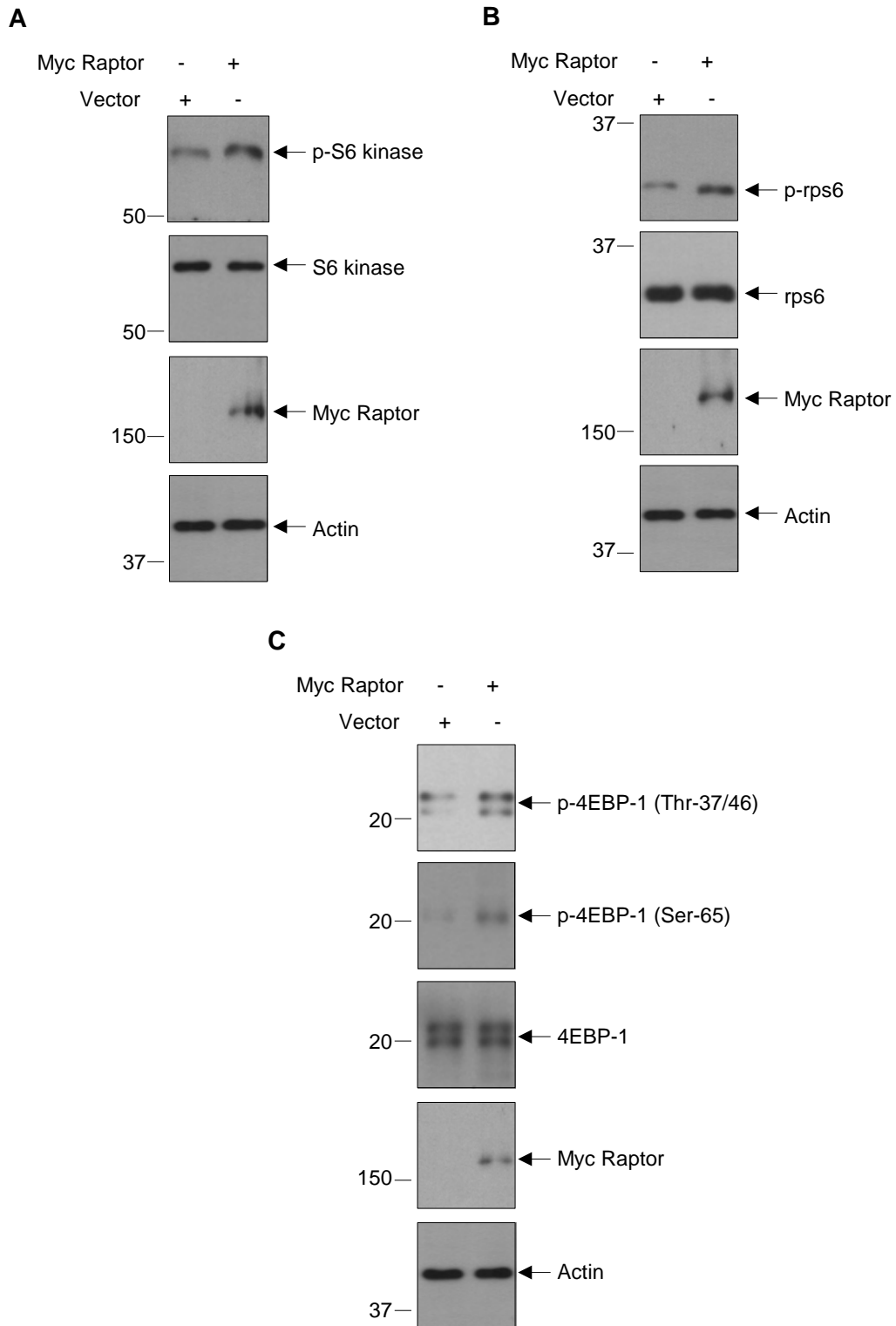


Figure S4. Expression of Raptor increases mTORC1 activity. Mesangial cells were transfected with Myc-tagged raptor or vector alone as indicated. The cell lysates were immunoblotted with indicated antibodies.

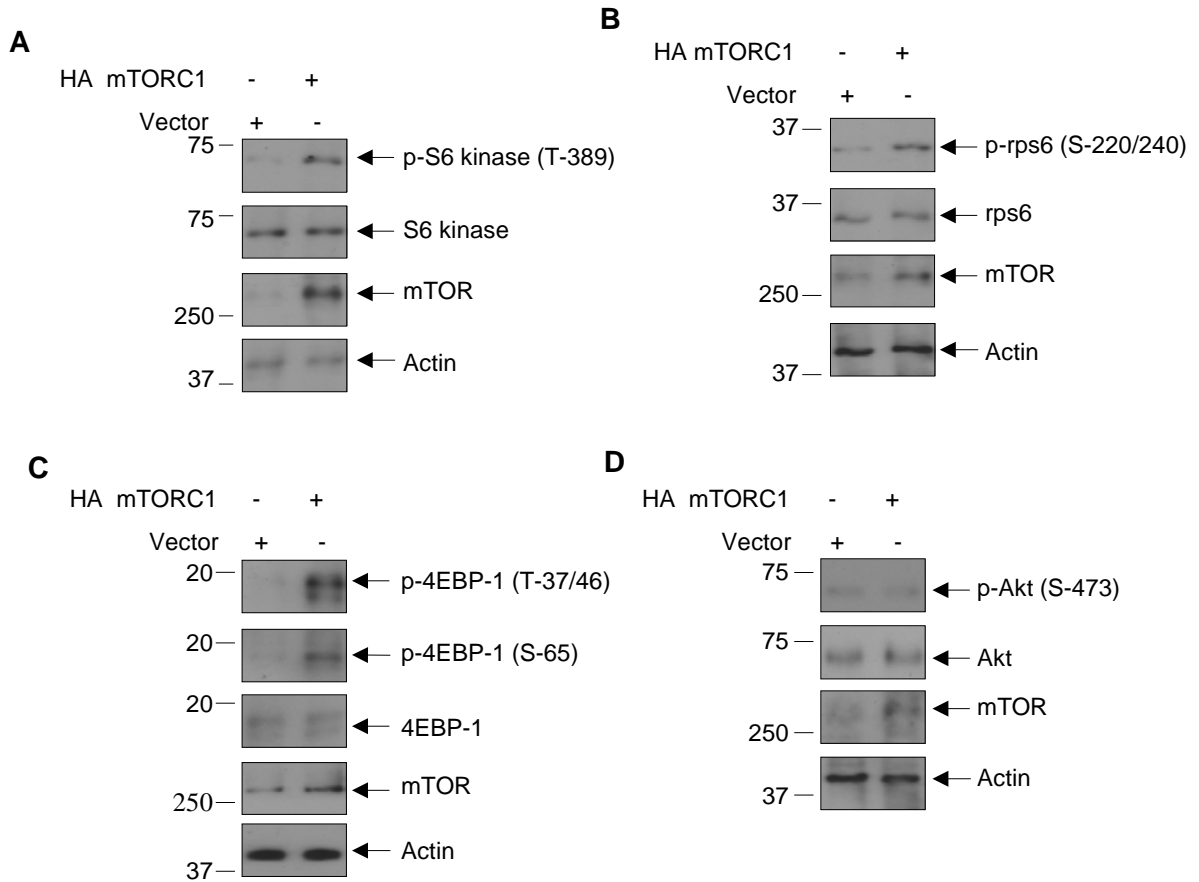


Figure S5. Expression of hyperactive (HA) mTOR mutant increases mTORC1 activity in the absence of mTORC2 activity. Mesangial cells were transfected with indicated plasmids. The cell lysates were immunoblotted with phospho-S6 kinase, phospho-rps6, phospho-4EBP-1 as indication of mTORC1 activity (panels A – C) and with phospho-Akt (Ser-473) as indication of mTORC2 activity (panel D).

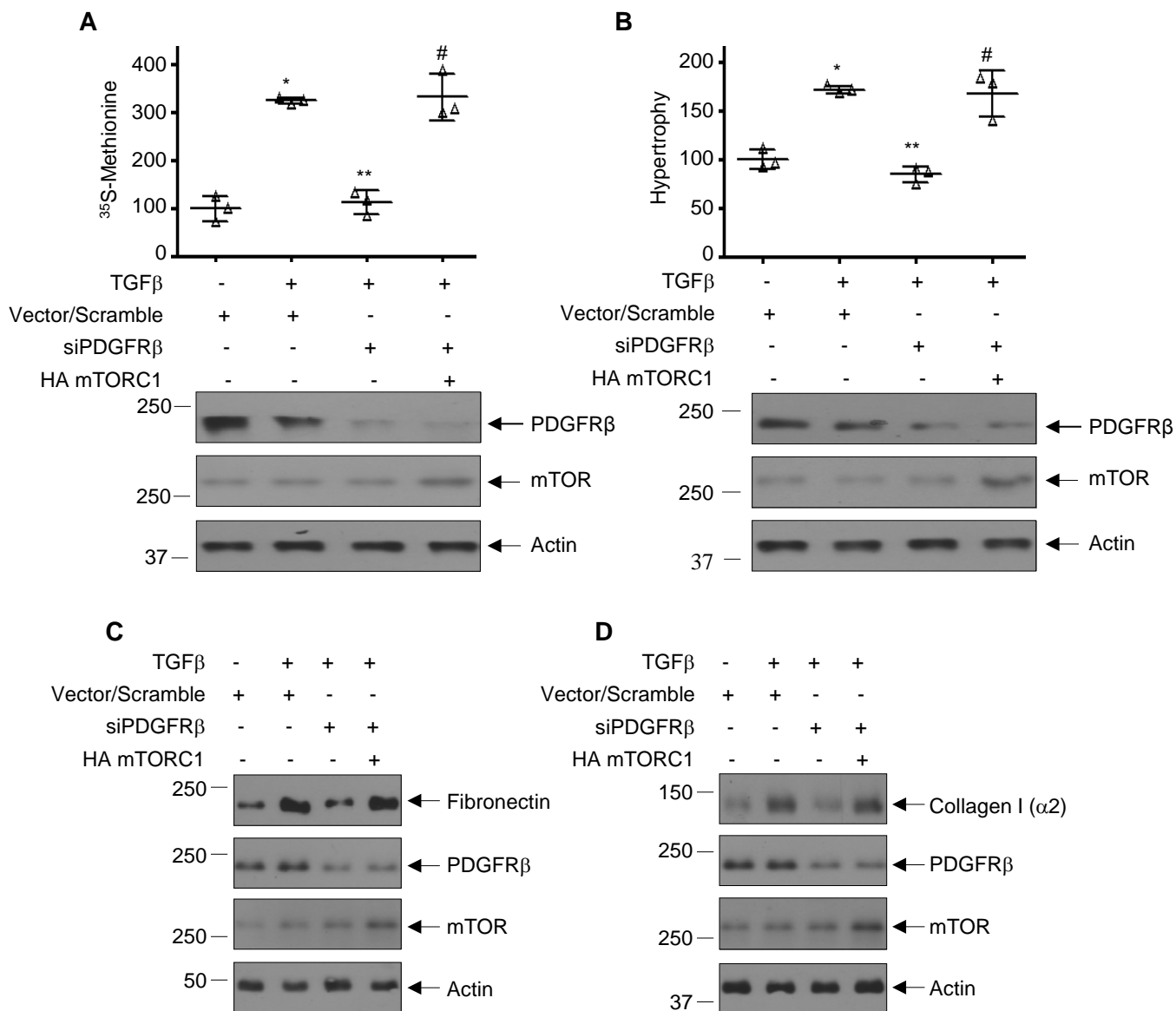


Figure S6. PDGFR β -stimulated mTORC1 regulates TGF β -induced mesangial cell pathology. Mesangial cells were transfected with siRNAs against PDGFR β or scrambled RNA along with hyperactive (HA) mTOR mutant conferring mTORC1 activity as indicated. Transfected cells were incubated with 2 ng/ml TGF β . In panels A and B, respectively, protein synthesis and hypertrophy of mesangial cells were measured as described in the experimental procedures. Mean \pm SD of triplicate measurements is shown. * $p < 0.0001$ vs control; ** $p < 0.0001$ vs TGF β ; # $p < 0.0001$ vs TGF β + siPDGFR β . In panels C and D, the cleared cell lysates were immunoblotted with indicated antibodies.

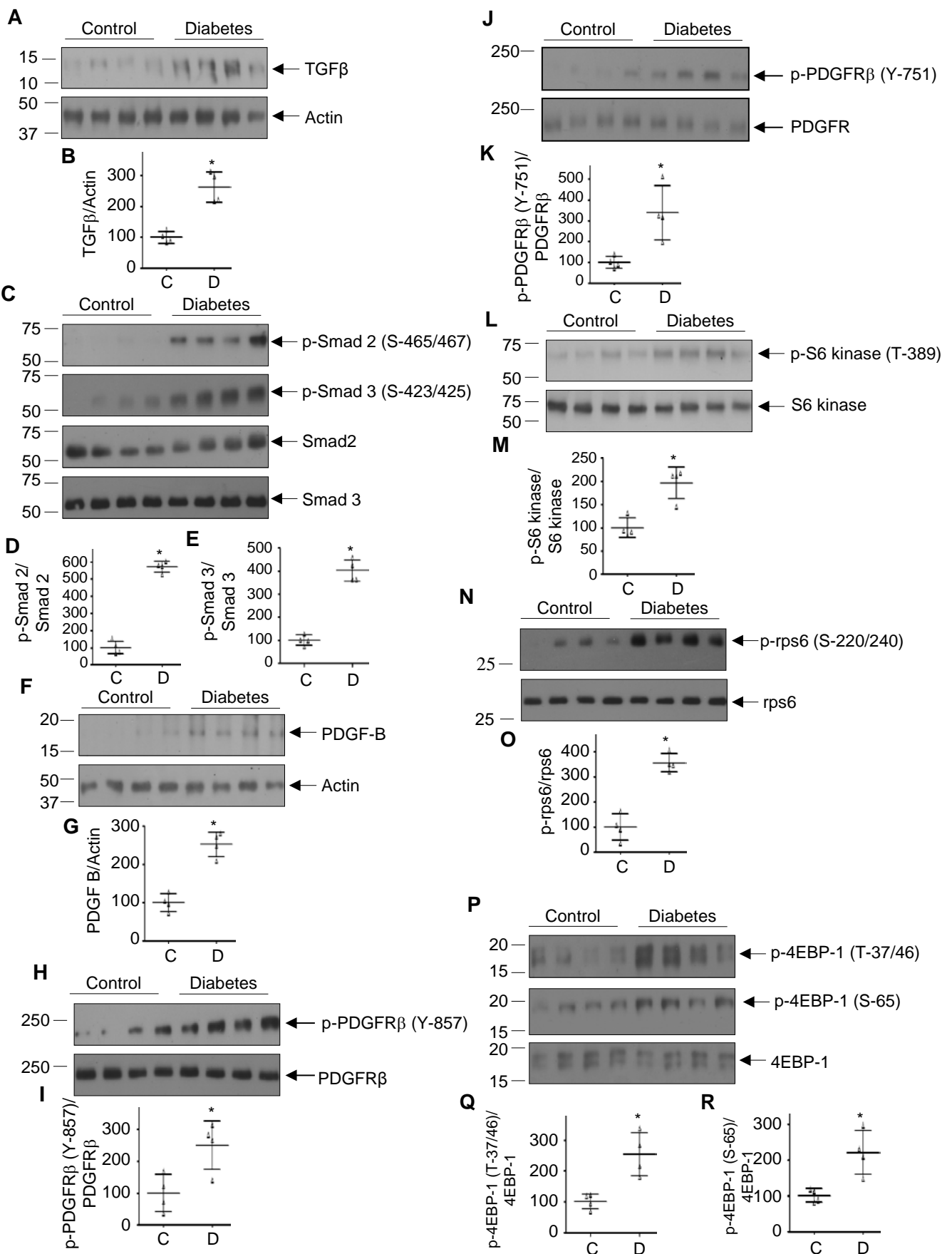


Figure S7. Increased level of TGFβ is associated with activation of PDGFRβ and mTORC1 signaling in STZ diabetic rat glomeruli. Renal glomerular lysates from control and STZ-induced diabetic rats were immunoblotted with indicated antibodies. Bottom panels in each blot represent quantification of protein bands. Mean ± SD of 4 animals is shown. **p* < 0.0007 – 0.03 vs control animals.