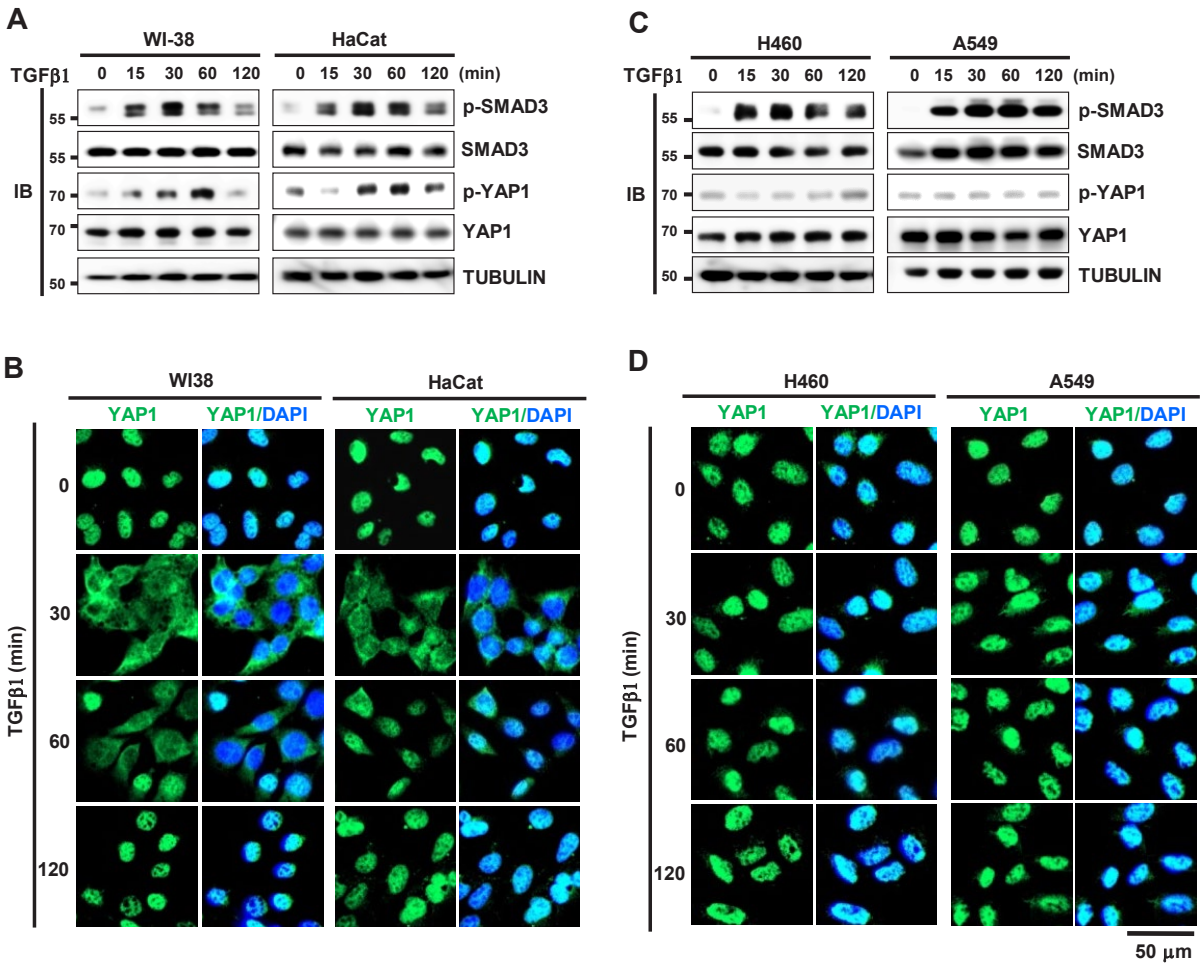

Fig. S1. TGFβ stimulation triggers nuclear-cytoplasmic shuttling of YAP1.

(A) Immortalized normal cell lines [WI-38 (lung fibroblast) and HaCaT (keratinocyte)] were stimulated with TGFβ1. Time-dependent phosphorylation of SMAD3 and YAP1 at various time points was analyzed by immunoblotting (IB).

(B) The cell lines were stimulated with TGFβ1 and time-dependent subcellular localization of YAP1 was analyzed by double immunofluorescence staining (green=YAP1; blue=DAPI).

(C) Cancer cell lines [H460 (human lung cancer) and A549 (human lung cancer)] were stimulated with TGFβ1. Time-dependent phosphorylation of SMAD3 and YAP1 were analyzed by IB.

(D) The cell lines were stimulated with TGFβ1 and time-dependent subcellular localization of YAP1 was analyzed by double immunofluorescence staining (green=YAP1; blue=DAPI).

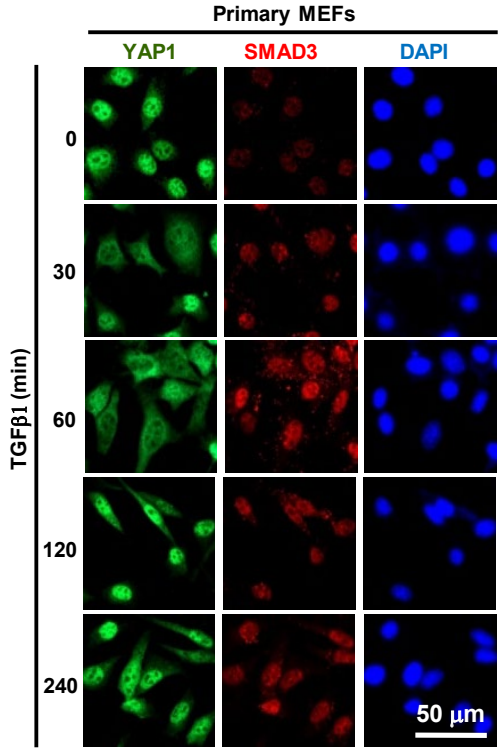

Fig. S2. Subcellular localization of YAP1 and SMAD3 in response to TGFβ stimulation

Primary MEFs were stimulated with TGFβ1. Time-dependent subcellular localization of YAP1 and SMAD3 at the indicated time points was analyzed by triple immunofluorescence staining (green=YAP1/TAZ; red=SMAD3; blue=DAPI).

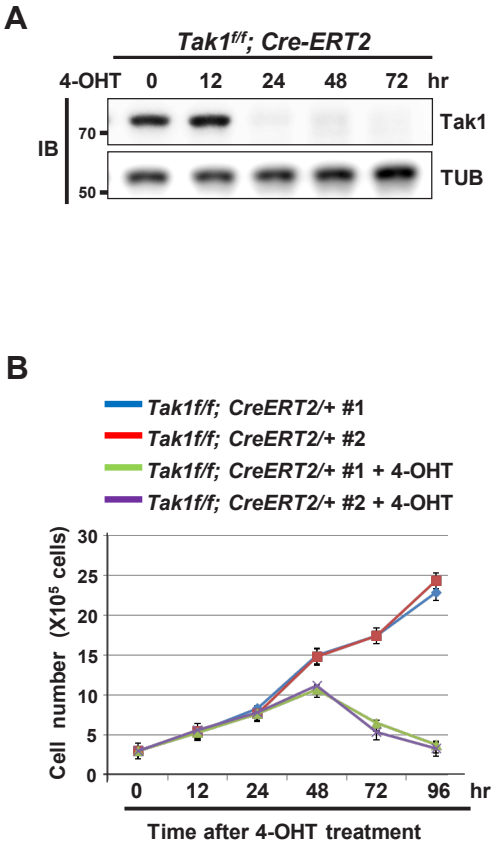

Fig. S3. Deletion of *Tak1* from *Tak1*$^{flox/flox}$*;Cre-ERT2* MEFs.

(A) *Cre-ERT2* is activated by treatment with 4-hydroxy-tamoxifen (4-OHT) and deletes the floxed gene. *Tak1*$^{flox/flox}$*;Cre-ERT2* MEFs were treated with 4-OHT, and deletion of *Tak1* was analyzed by measuring expression of the Tak1 protein by IB at the indicated time points. The results revealed that *Tak1* was completely deleted 24 h after 4-OHT treatment.

(B) *Tak1*$^{flox/flox}$*;Cre-ERT2* MEFs were treated with vehicle or 4-OHT, and cell viability was measured at the indicated time points. MEF clones are denoted as #1 and #2.

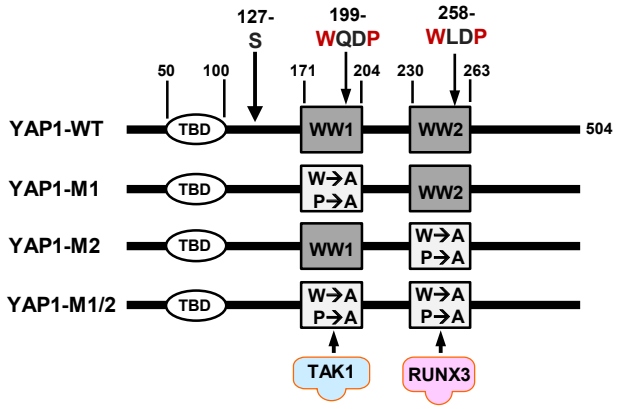

Fig. S4. Schematic illustration of YAP1 mutants.

Flag-YAP1-M1 (a WW1 domain-disrupted YAP1 mutant; W199A and P202A), Flag-YAP1-M2 (a WW2 domain-disrupted YAP1 mutant; W258A and P261A), or Flag-YAP1-M1/2 (a mutant in which the YAP1 WW1 and WW2 domains are disrupted) are shown.

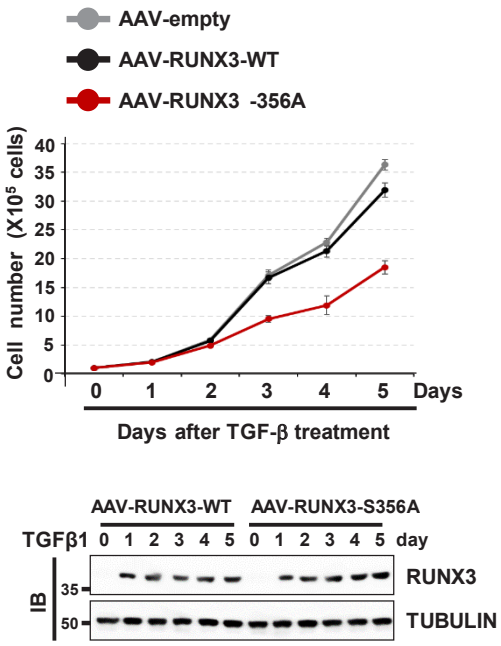

Fig. S5. Role of TGFβ-RPA-AC in cell cycle regulation.

HEK293 cells were infected with AAV-RUNX3-WT, AAV- RUNX3-S356A or control AAV-empty and the proliferation rate of the cells were measured. Expressions of RUNX3-WT and RUNX3-S356A were detected by IB.

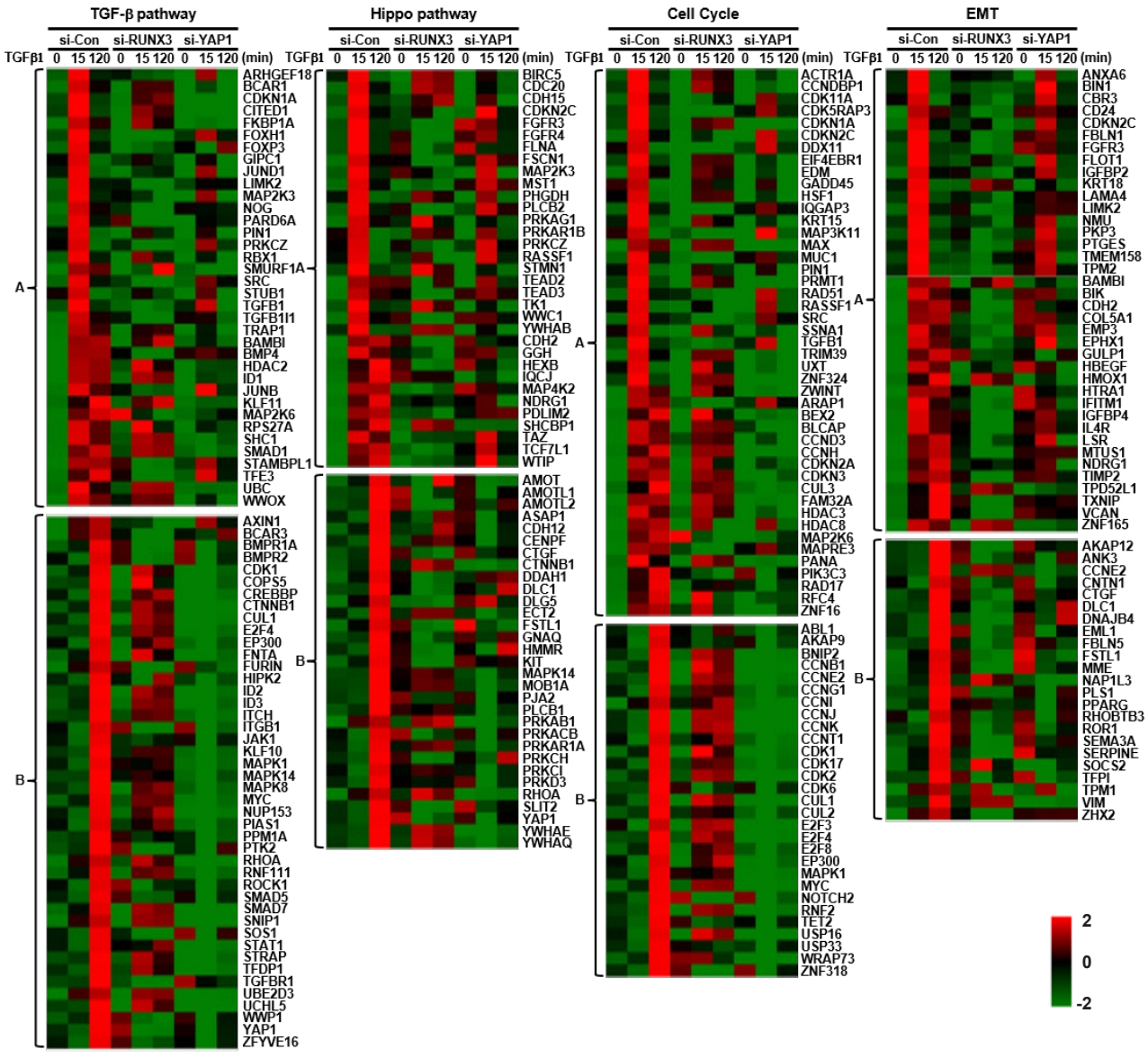

Fig. S6. Biphasic roles of YAP1 in regulating target genes of TGFβ signaling.

Heatmap showing genes upregulated after TGFβ1 stimulation of HEK293 cells, and gene expression levels in si-*RUNX3* or si-*YAP1*-treated cells. The categories of TGFβ-induced YAP1-dependent genes include the "TGFβ pathway", the "Hippo pathway", "cell cycle regulation", and "epithelial-mesenchymal transition (EMT)". Early and late genes are indicated by A and B, respectively

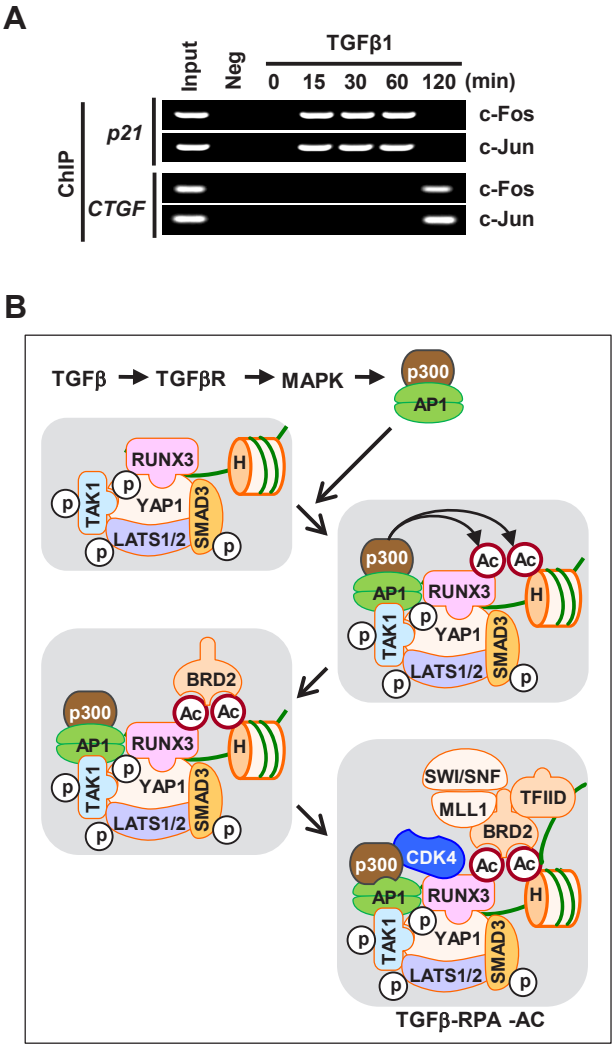

Fig. S7. AP1 is an essential component of the TGFβ-RPA-AC complex.

(A) HEK293 cells were stimulated with TGFβ1. The binding of c-FOS and c-JUN to the *p21* or *CTGF* locus was measured by ChIP at the indicated time points. One-thirtieth of the lysates was used as the as input for PCR.

(B) Schematic illustration of the role of AP1/p300 in TGFβ-RPA-AC complex formation.