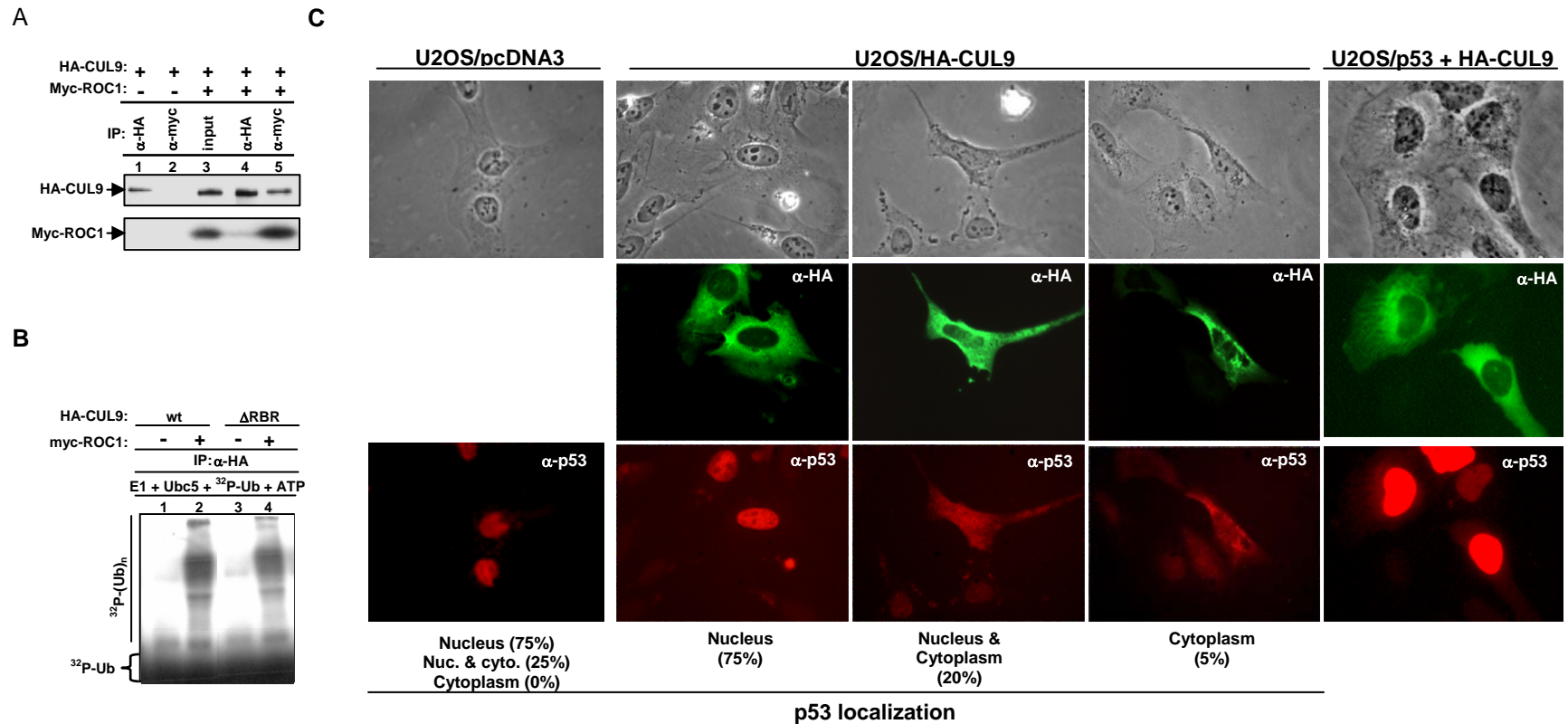


Pei et al. Supplementary Figure S1



**Supplementary Figure S1. CUL9 binds to ROC1, localizes in the cytoplasm, but does not sequesters p53 to the cytoplasm.**

**A.** CUL9 binds with ROC1. HA- CUL9 was co-transfected 293T cells with ROC1 and its association was examined by reciprocal IP-Western.

**B.** CUL9 immunocomplexes contain ubiquitin ligase activity. HA-tagged wild-type or mutant CUL9 with the two C-terminal RING fingers deleted ( $\Delta$  RBR) were transiently transfected into 293T cells, either alone or with ROC1. HA immunocomplexes were incubated *in vitro* with E1, E2 (UbcH5) and  $^{32}$ P-labeled ubiquitin in the presence of ATP. Self-ubiquitinylation ligase activity (E2 activation) was assayed by the synthesis of polyubiquitin chain.

**C.** CUL9 does not sequester p53 in the cytoplasm. Subcellular localization of endogenous or ectopically expressed p53 in the absence or presence of ectopic expression of CUL9 was analyzed by indirect immunofluorescence in U2OS cells. Subcellular localization of p53 was categorized into three groups and the percentage of each group was calculated from microscopic examination of approximately 300 cells.