

Figure S6. FUNDC1 is required for ULK1 translocation to mitochondria upon mitophagy induction

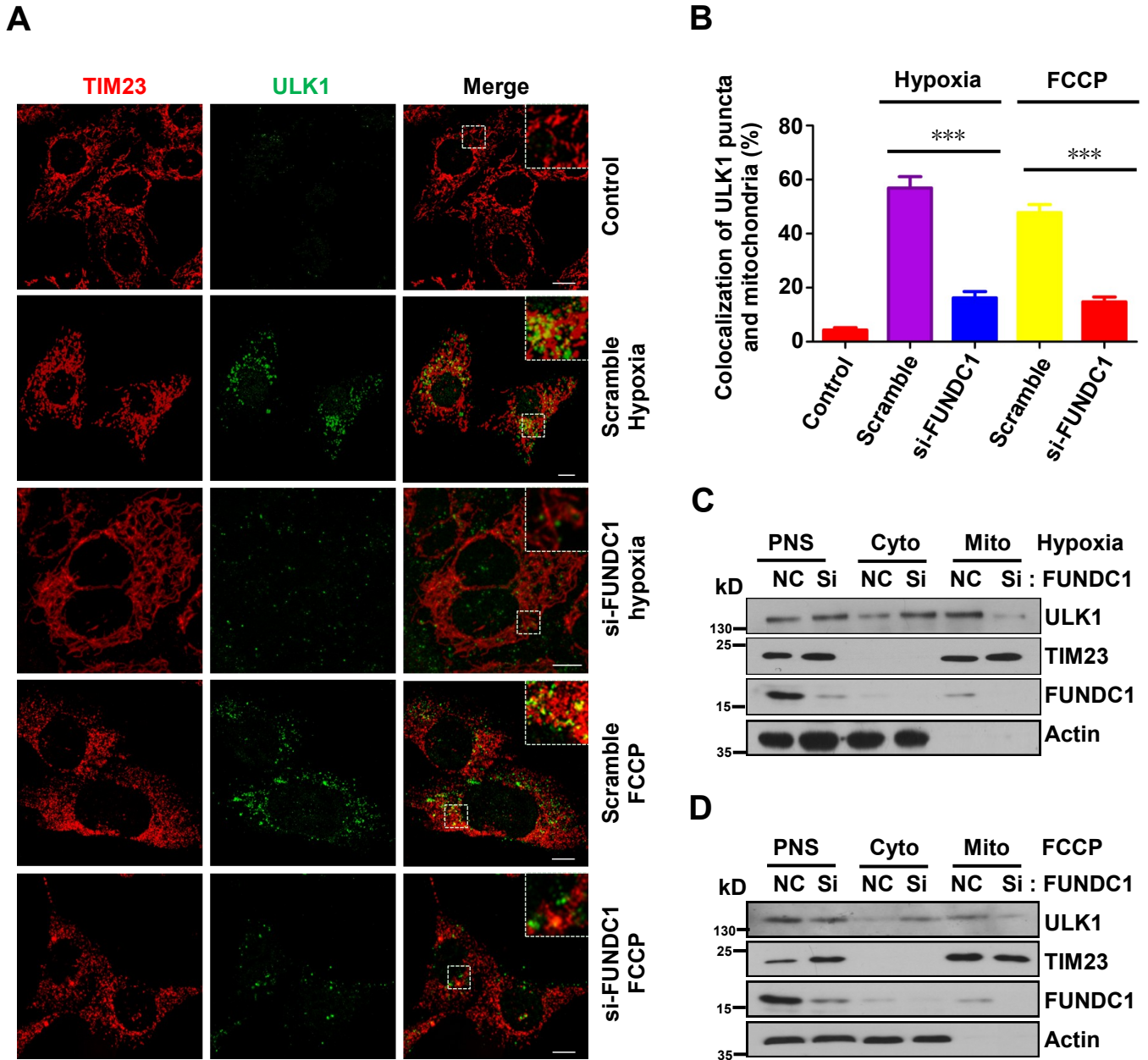


Figure S6. FUNDC1 is required for ULK1 translocation to fragmented mitochondria upon hypoxia or FCCP treatment

- (A) MEFs were exposed to hypoxia (1% O₂) for 12 h or FCCP (20 μM) for 6 h after the endogenous FUNDC1 was knocked down with siRNA. Cells were fixed by 4% paraformaldehyde and stained with anti-TIM23 (mouse) and anti-ULK1 (rabbit) primary antibodies, then Alexa fluor-488 labeled Donkey anti-Rabbit IgG and Alexa fluor-555 labeled Donkey anti-Mouse IgG secondary antibodies before analysis by immunofluorescence microscopy. Bar, 10 μm. All results are from 3–4 independent experiments.
- (B) Quantification of colocalization of ULK1 puncta with mitochondria in (A). Data represent mean ± SEM of 50 cells. *** P<0.001.
- (C) Immunoblots of subcellular fractions isolated from MEFs cultured in hypoxic (1% O₂) conditions for 12 h after the endogenous FUNDC1 was knocked down (Si) or not (NC). PNS, post-nuclear supernatant; Cyto, cytosol; Mito, mitochondria. All results are from 3–4 independent experiments.
- (D) Immunoblots of subcellular fractions isolated from MEFs treated with FCCP (20 μM) for 6 h after the endogenous FUNDC1 was knocked down (Si) or not (NC). PNS, post-nuclear supernatant; Cyto, cytosol; Mito, mitochondria. All results are from 3–4 independent experiments.