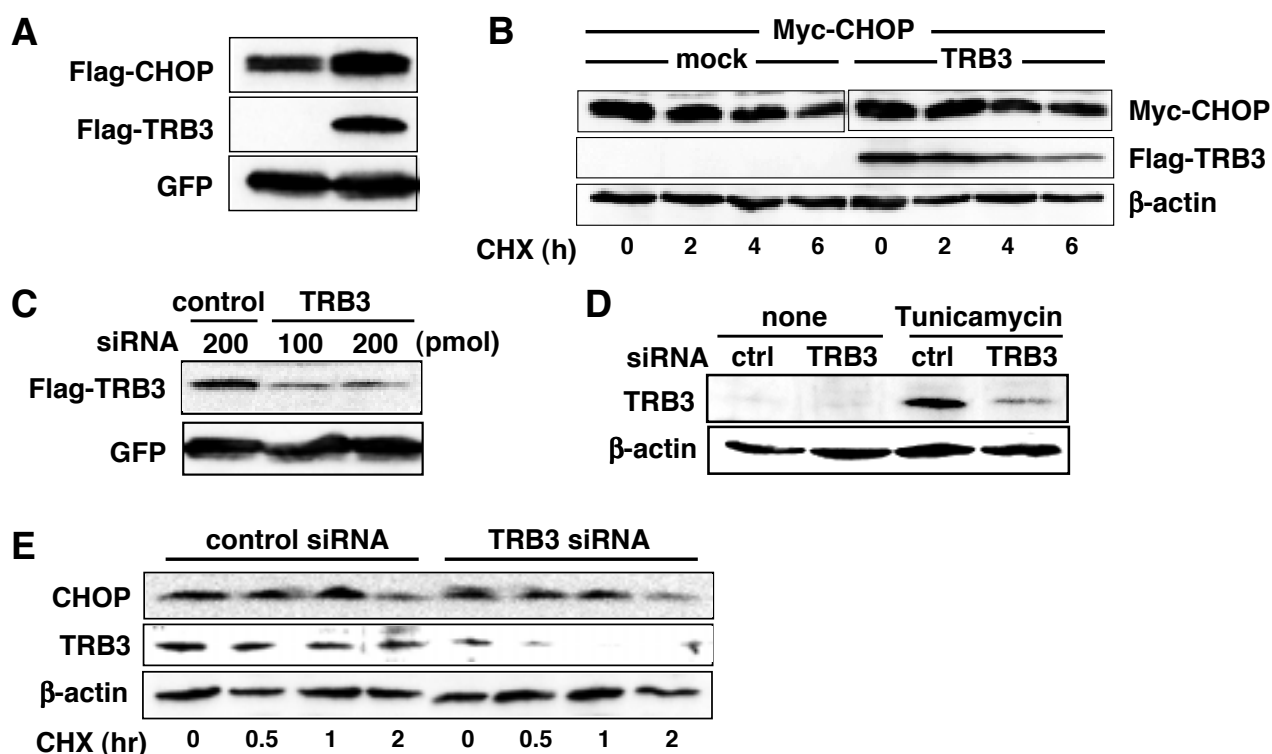


Supplementary Figure S3



TRB3 did not promote the degradation of CHOP.

(A) 293 cells were transiently transfected with Flag-CHOP and pEGFP-C1 with or without Flag-TRB3. After 48hr, the cell lysates were analyzed by immunoblotting using anti-Flag antibody. Transfection efficiency was assessed by detecting the GFP expression level with anti-GFP antibody.

(B) 293 cells were transiently transfected with Myc-CHOP with or without Flag-TRB3. After 36hr, cells were treated with 10 μ g/ml of cycloheximide (CHX) for the indicated periods. And the cell lysates were analysed by immunoblotting using anti-Flag, anti-Myc and anti- β -actin.

(C) 293 cells were transiently transfected with Flag-TRB3, pEGFP-C1 and control siRNA or TRB3 siRNA. After 48hr, the cell lysates were analyzed by immunoblotting using anti-Flag antibody. Transfection efficiency was assessed by detecting GFP expression level with anti-GFP antibody.

(D) 293 cells were transiently transfected with control siRNA or TRB3 siRNA. After 48 hr, cells were treated with 2 μ g/ml tunicamycin for 6hr. The cell lysates were analyzed by immunoblotting using anti-TRB3, and anti- β -actin antibodies.

(E) 293 cells were transiently transfected with control siRNA or TRB3 siRNA. After 48 hr, cells were treated with 2 μ g/ml tunicamycin for 6hr, and then cells were treated with 10 μ g/ml of cycloheximide (CHX) for the indicated periods. The cell lysates were analyzed by immunoblotting using anti-CHOP, anti-TRB3 and anti- β -actin antibodies (right).