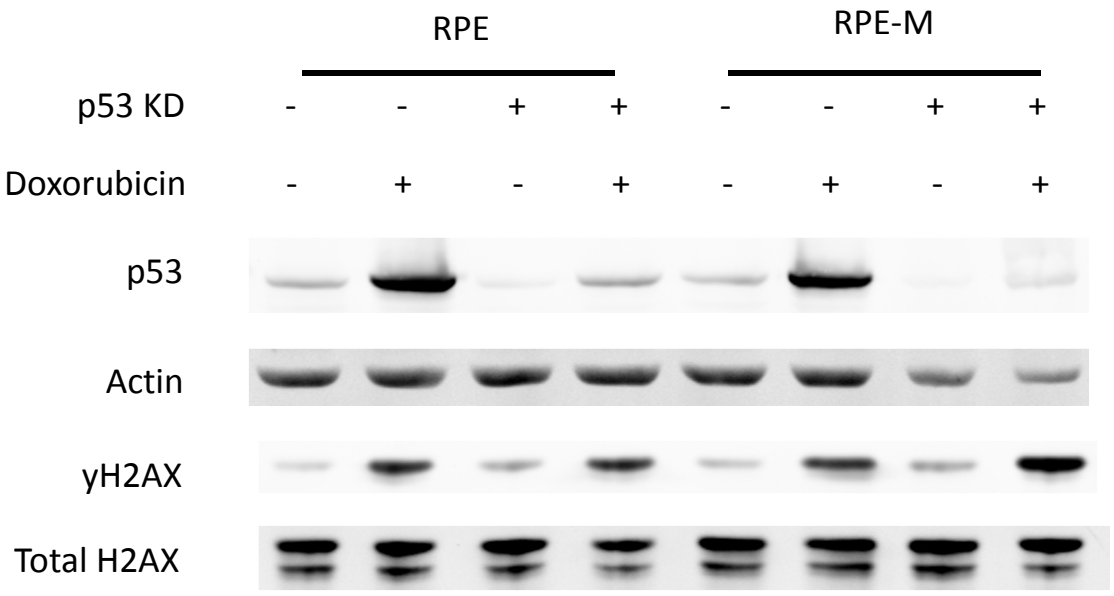
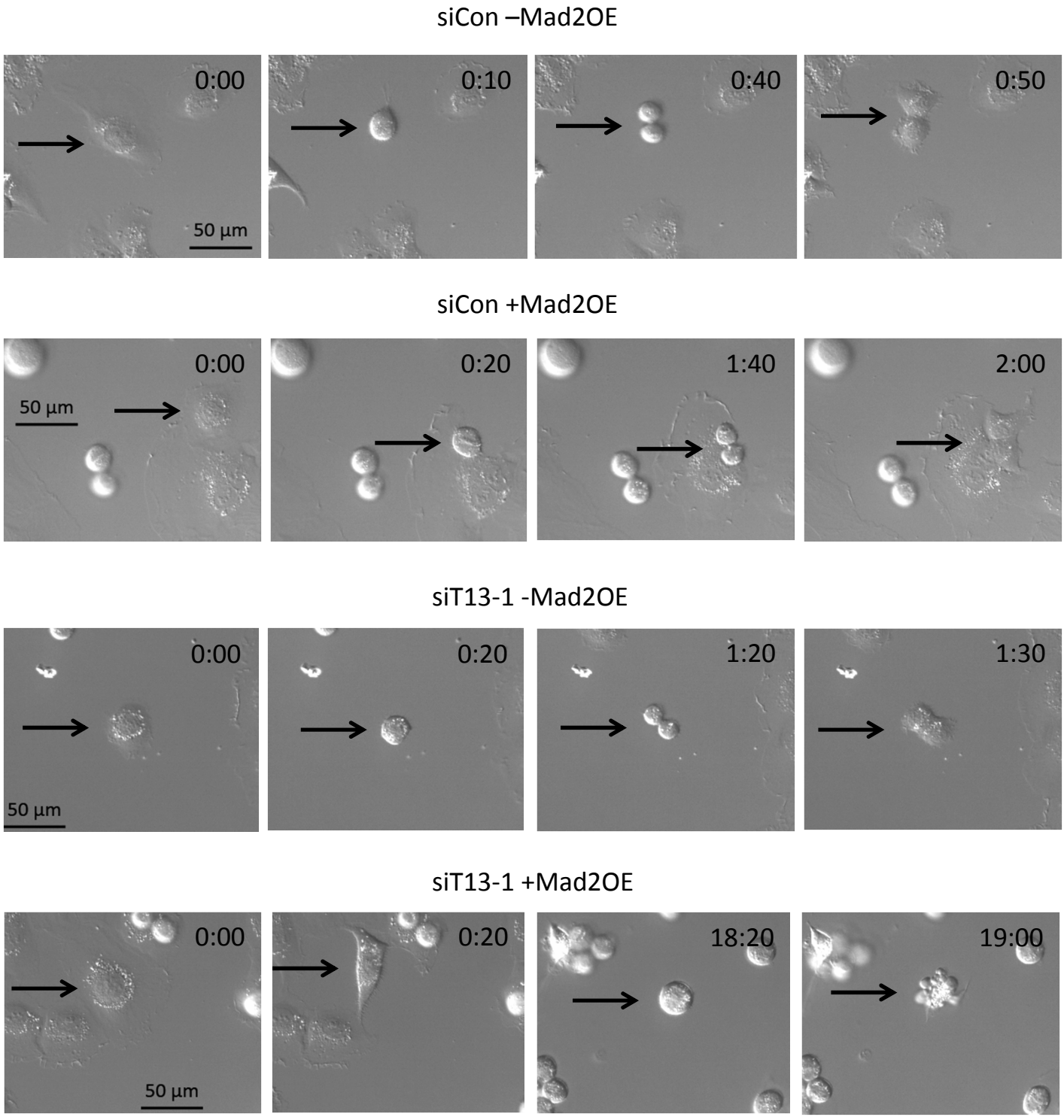


Supplementary Figure 1: p53 knockdown in RPE and RPE-M cells, Related to Figure 3

Supplementary Figure 1. **p53 knockdown in RPE and RPE-M cells, Related to Figure 3.**

Western blot of RPE and RPE-M cells with or without transduction with pLVTH-sip53. Cells were treated +/- 0.2 ug/ml doxorubicin for 11 hours to induce DNA damage.

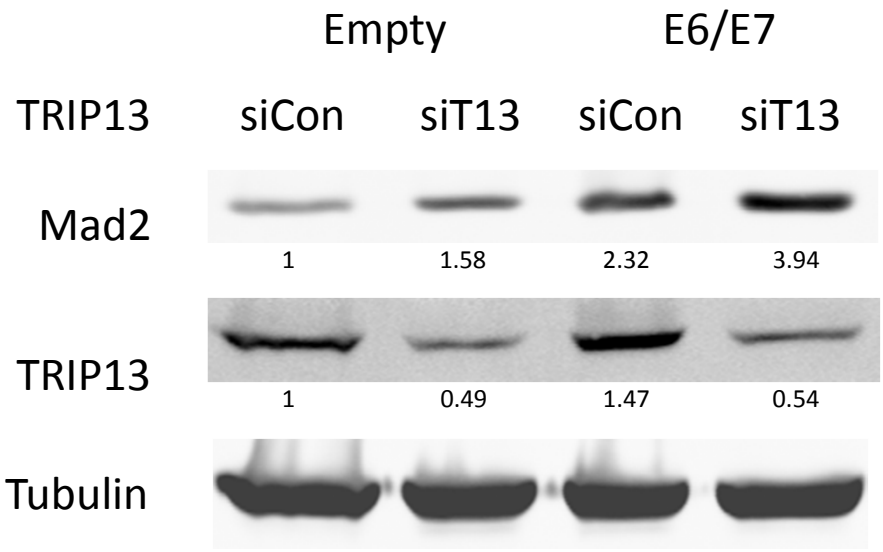


Supplementary Figure 2: Time-lapse sequences of RPE cells,
Related to Figures 3 and 4

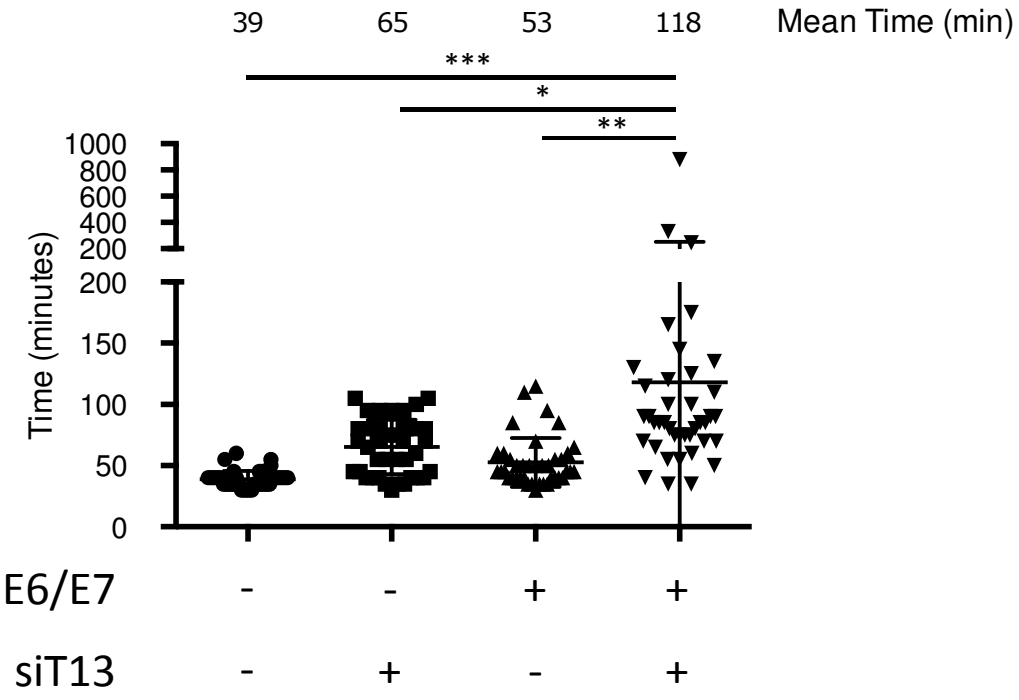
Supplementary Figure 2. **Time-lapse sequences of RPE cells, Related to Figures 3 and 4.**

Cells were transfected with control or TRIP13 siRNA 3 days prior to imaging, Mad2 was induced with doxycycline 1 day prior to imaging. Shown are representative cells entering mitosis for RPE cells treated with control siRNA or TRIP13 siRNA, with or without Mad2 overexpression. Arrows point to cells that enter and exit mitosis.

A



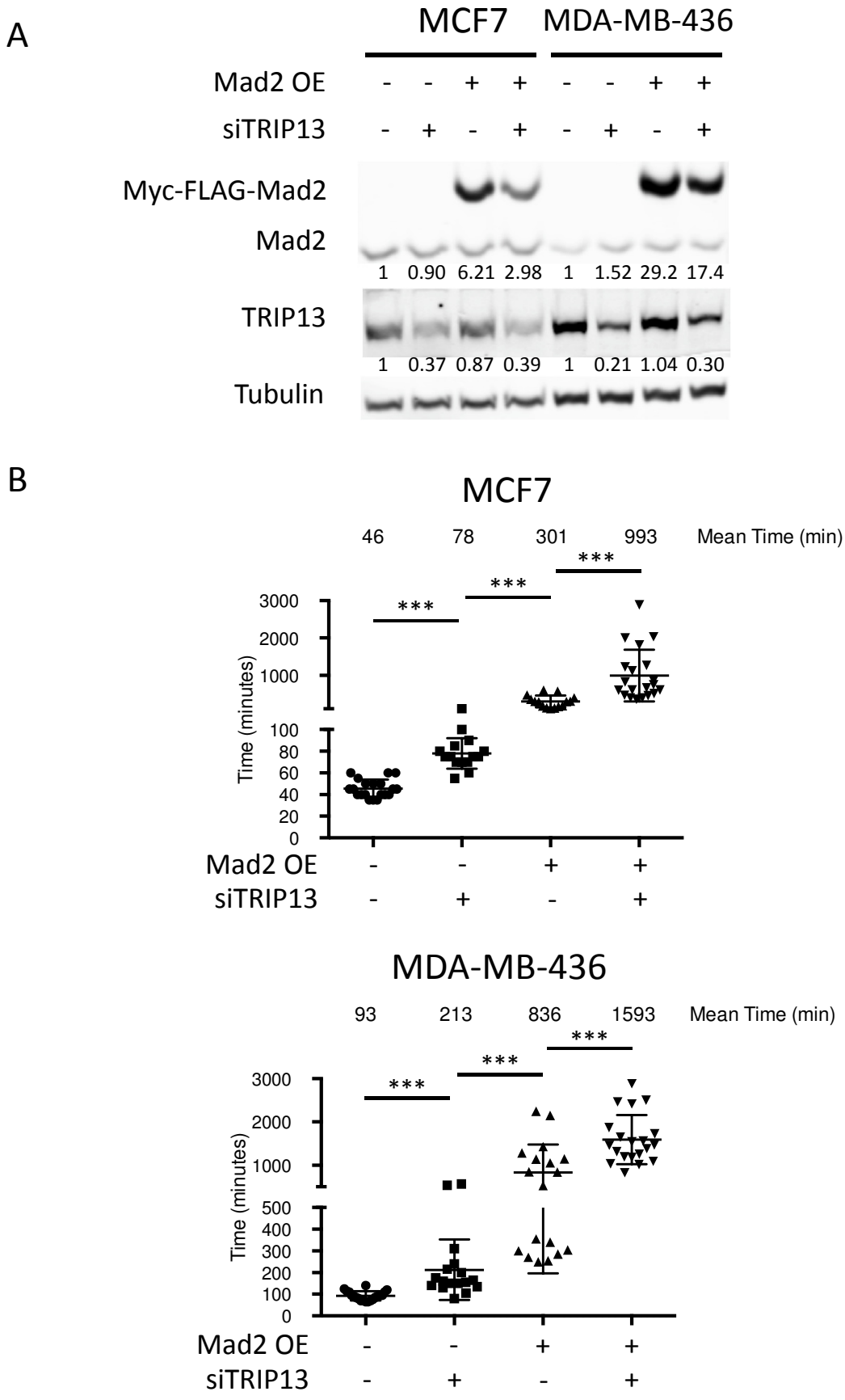
B



Supplementary Figure 3: TRIP13 knockdown in E6/E7 transduced cells causes mitotic delay, Related to Figure 3

Supplementary Figure 3. **TRIP13 knockdown in E6/E7 transduced cells causes mitotic delay, Related to Figure 3.**

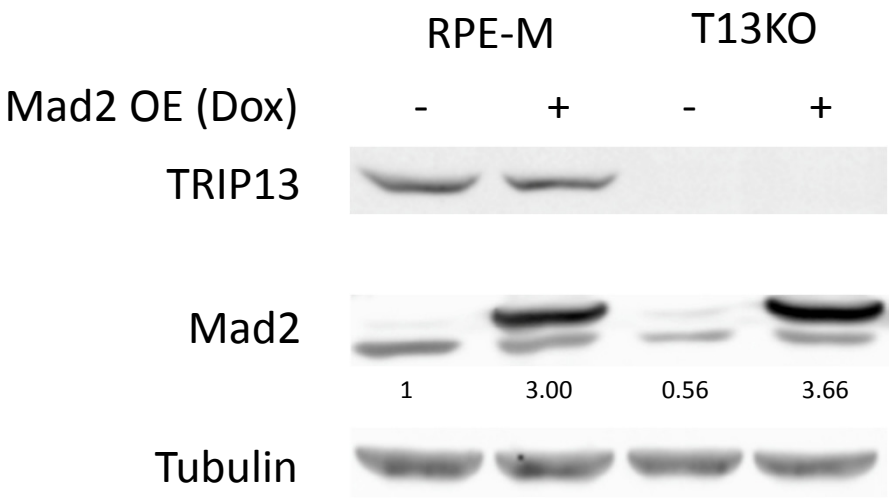
(A) Western blot of cells transduced with empty or E6/E7 expressing pLXSN vectors and transfected with control or TRIP13 siRNA #1. Quantification shown below corresponding bands. (B) Quantification of mitotic duration of pLXSN-Empty and E6/E7 transfected with control or TRIP13 siRNA #1. * indicates $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$; Unpaired t test. Data are represented as mean± SD.



Supplementary Figure 4: Mad2 overexpression and TRIP13 knockdown causes prolonged mitotic arrests in MCF7 and MDA-MB-436 cells, Related to Figure 3

Supplementary Figure 4. **Mad2 overexpression and TRIP13 knockdown causes prolonged mitotic arrests in MCF7 and MDA-MB-436 cells, Related to Figure 3.**

(A) Western blot of MCF7 and MDA-MB-436 cells 3 days after transfection with TRIP13 siRNA #1 or control siRNA and Myc-FLAG-Mad2 or empty vector. Total quantification (endogenous + exogenous) is shown below corresponding bands. (B) Mitotic timing of cells in (A). n ≥15 cells for each condition. *** indicates $p < 0.001$; Unpaired t test. Data are represented as mean± SD.

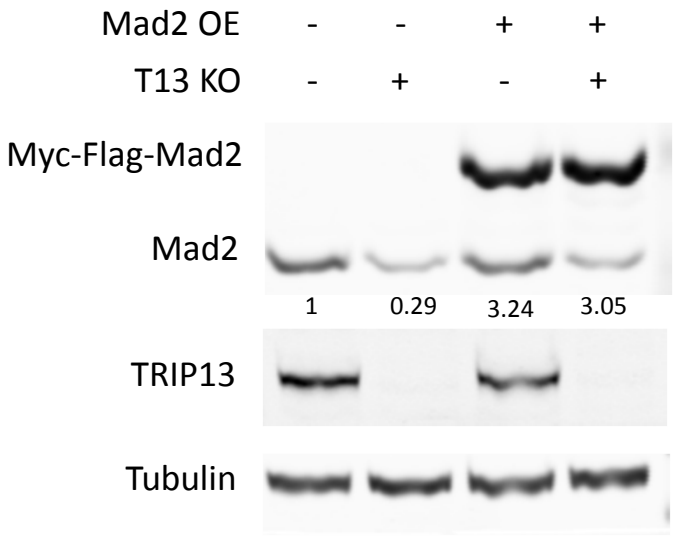


Supplementary Figure 5: Mad2 overexpression in TRIP13 knockout cells, Related to Figure 5

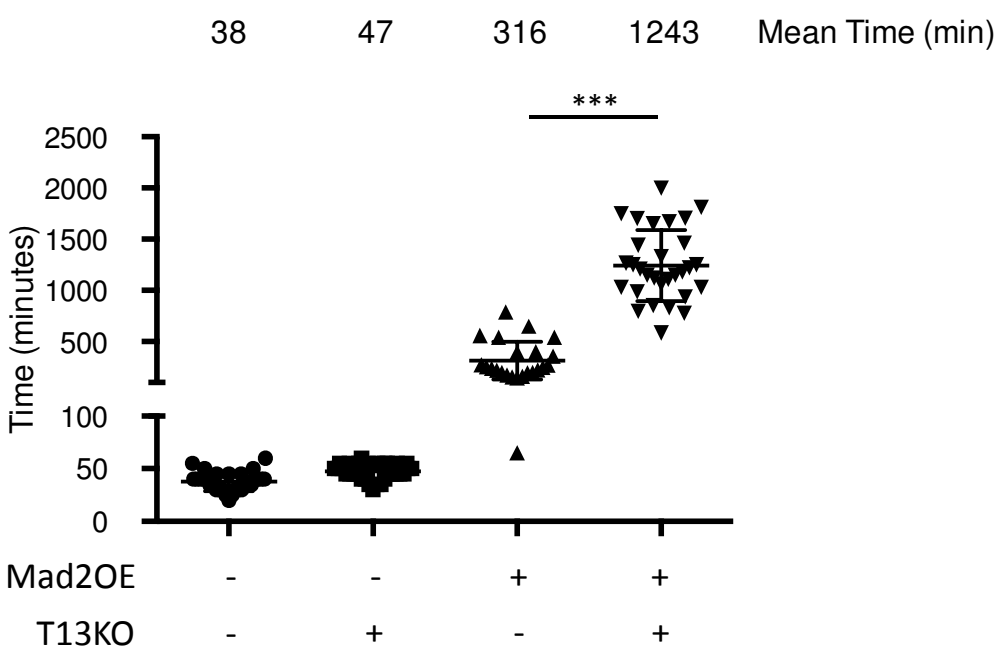
Supplementary Figure 5. **Mad2 overexpression in TRIP13 knockout cells, Related to Figure 5.**

Western blot of RPE-M and T13KO cells induced to express HA-Mad2 with doxycycline for 24 hours. Total quantification (endogenous + exogenous) is shown below corresponding bands.

A



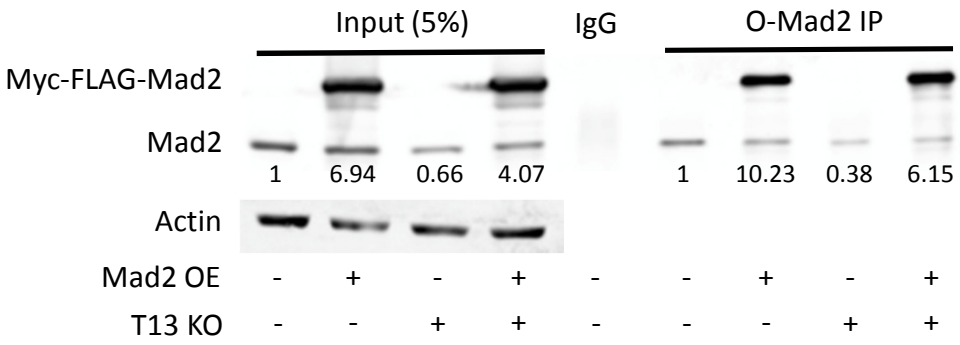
B



Supplementary Figure 6: Human Mad2 combined with TRIP13 knockdown induces prolonged mitotic arrest, Related to Figure 5

Supplementary Figure 6. **Human Mad2 combined with TRIP13 knockdown induces prolonged mitotic arrest, Related to Figure 5.**

(A) Western blot of RPE-M and T13KO cells 3 days after transfection with TRIP13 siRNA #1 or control siRNA and Myc-FLAG-Mad2 or empty vector. Total quantification (endogenous + exogenous) is shown below corresponding bands. (B) Mitotic timing of cells in (A). n ≥20 cells for each condition. *** indicates $p < 0.001$; Unpaired t test. Data are represented as mean± SD.



Supplementary Figure 7: Mad2 overexpression provides a source of O-Mad2 in TRIP13 knockout cells, Related to Figure 6

Supplementary Figure 7. **Mad2 overexpression provides a source of O-Mad2 in TRIP13 knockout cells, Related to Figure 6.**

Western blot of O-Mad2 IPs from asynchronous RPE cells with or without transfection with Myc-FLAG-Mad2 (2 days post transfection). Total quantification (endogenous + exogenous) is shown below corresponding bands.