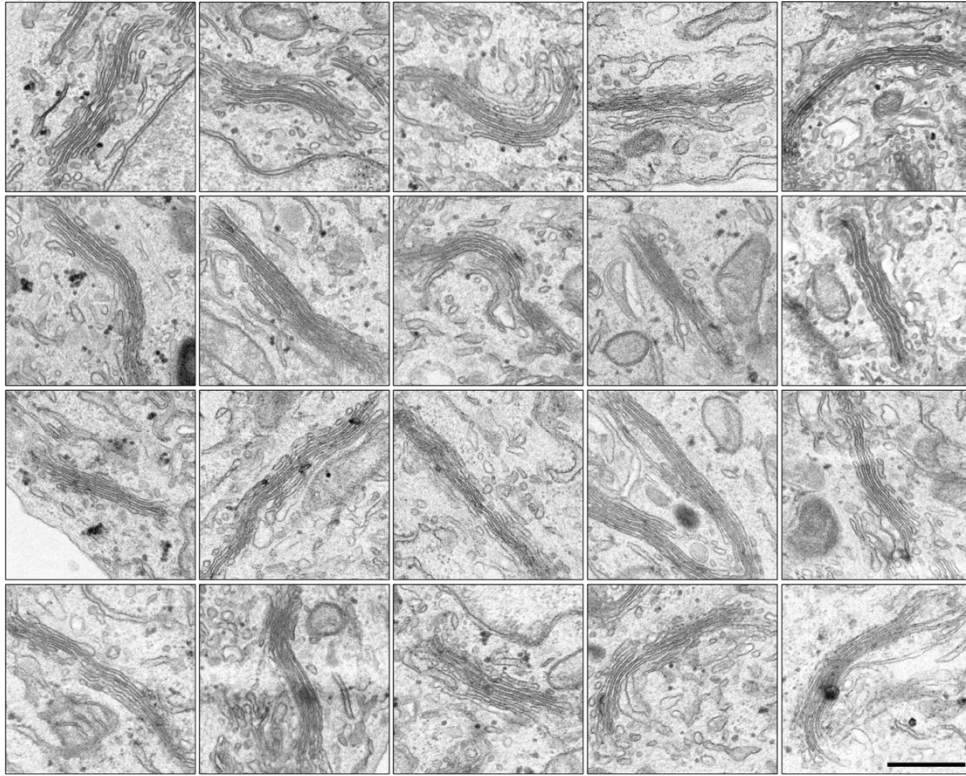


Figure S1. The endogenous SIRT2 protein level is low.

Asynchronous (AS) and mitotic HeLa cells or HeLa cells co-overexpressing GFP-tagged GRASP55 or Golgin-84 and FLAG-SIRT2 were lysed, blotted for GRASP55 (G55 in the figure) and SIRT2. Note that the endogenous SIRT2 level (lane 1 and 2) is very low compared to the overexpressed FLAG-SIRT2 (lane 3-8). Arrows indicate asynchronous SIRT2 bands, while arrowheads indicate mitotic SIRT2 bands.

A. si Control



B. si SIRT2

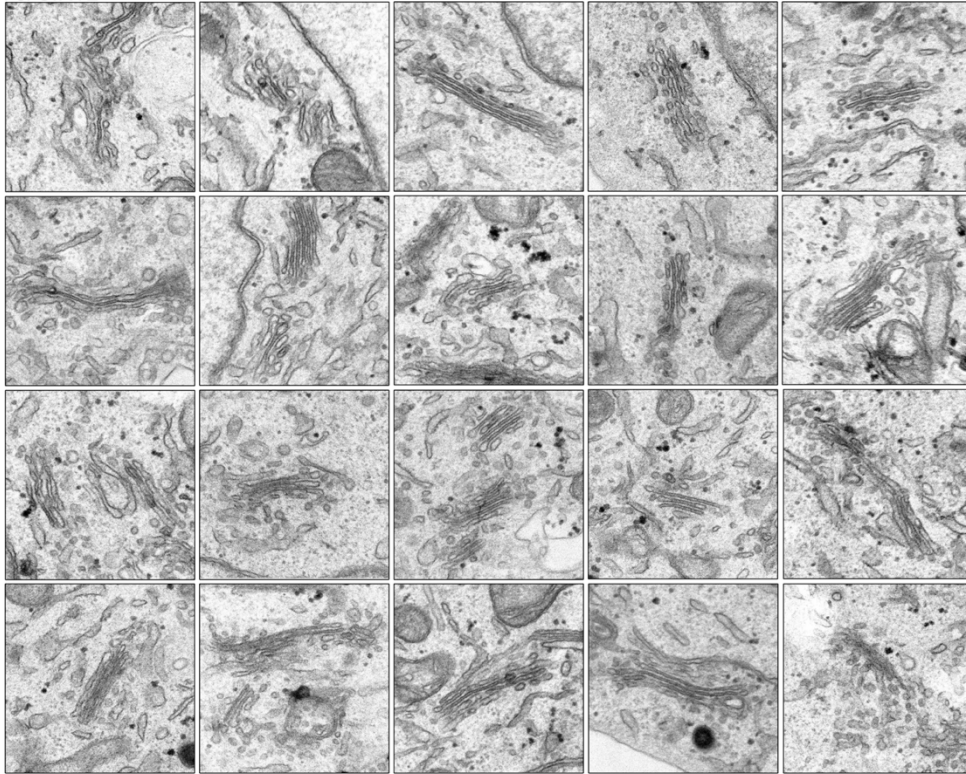


Figure S2. SIRT2 is required for proper Golgi structure maintenance.

HeLa cells were transfected with control (**A**) or SIRT2 (**B**) siRNA and analyzed by EM. Shown are galleries of EM images. The reduced average number or length of Golgi cisternae were frequently observed after SIRT2 depletion as displayed in **B**. Scale bar: 500 nm.

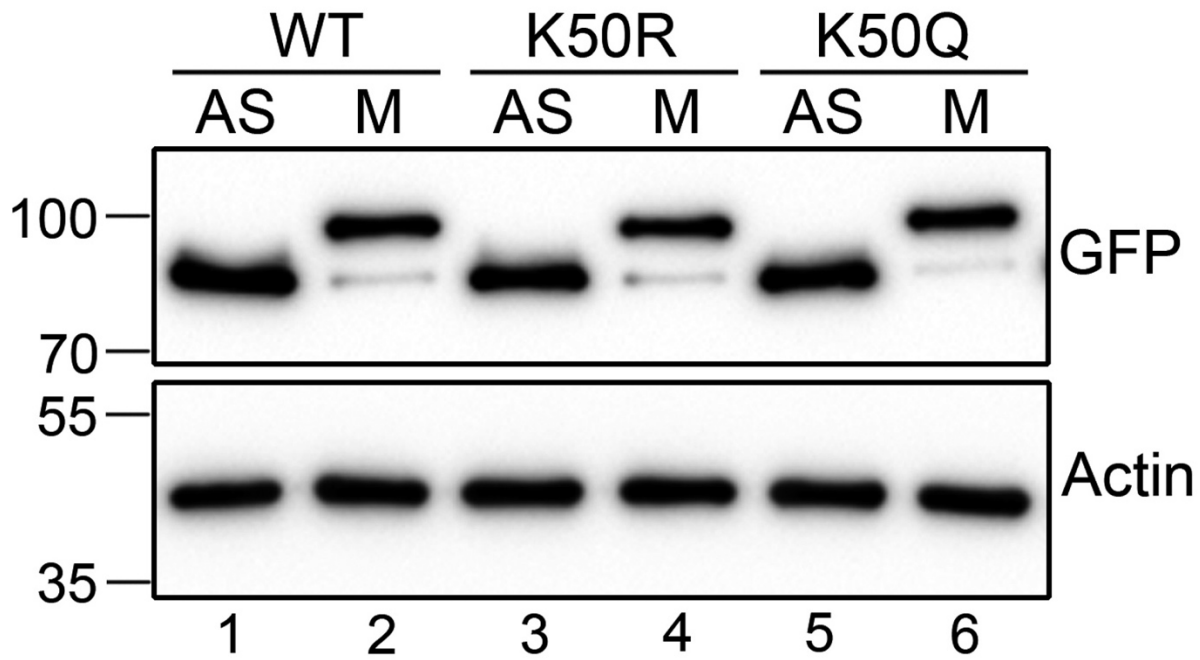


Figure S3. GRASP55 acetylation status does not affect its phosphorylation in mitosis.

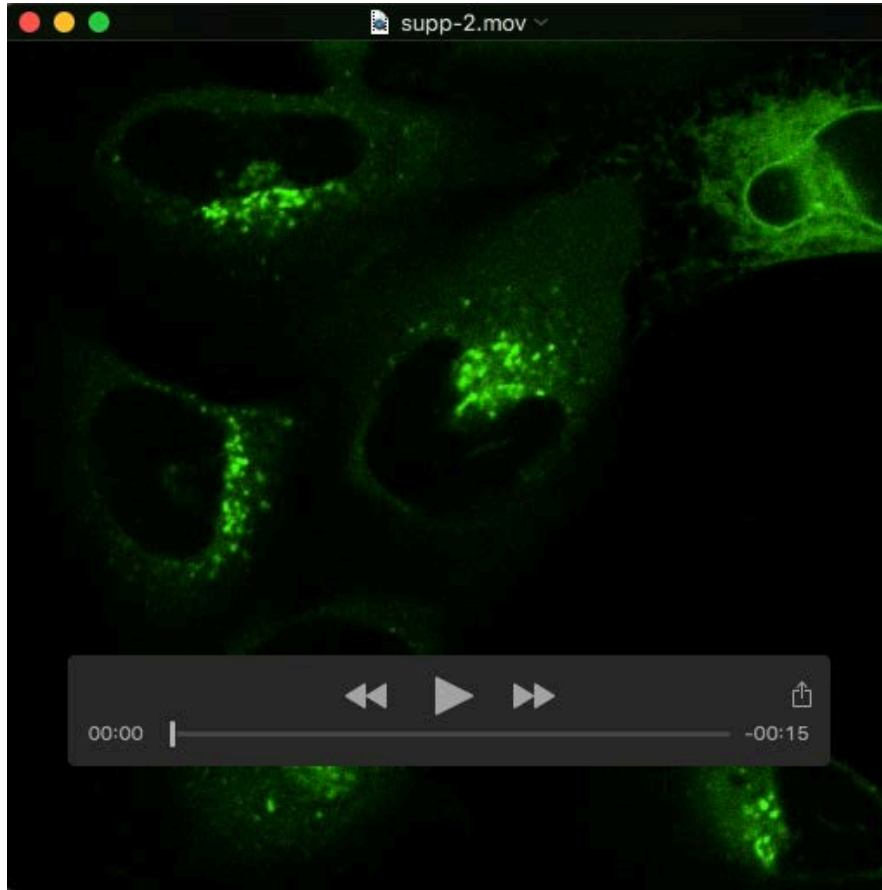
HeLa cells were transfected with GFP-tagged GRASP55 WT, K50R or K50Q. Non-synchronized (AS) or mitotic (M) cells synchronized with nocodazole were lysed and blotted for GFP. Note that the GRASP55 phosphorylation status indicated by the band-shift is not affected by the acetylation mutations.

Table S1. Mass spectrometry analysis of GRASP55 post-translational modifications (acetylation, phosphorylation and ubiquitination) in asynchronous and mitotic HEK293T cells expressing GRASP55-GFP.

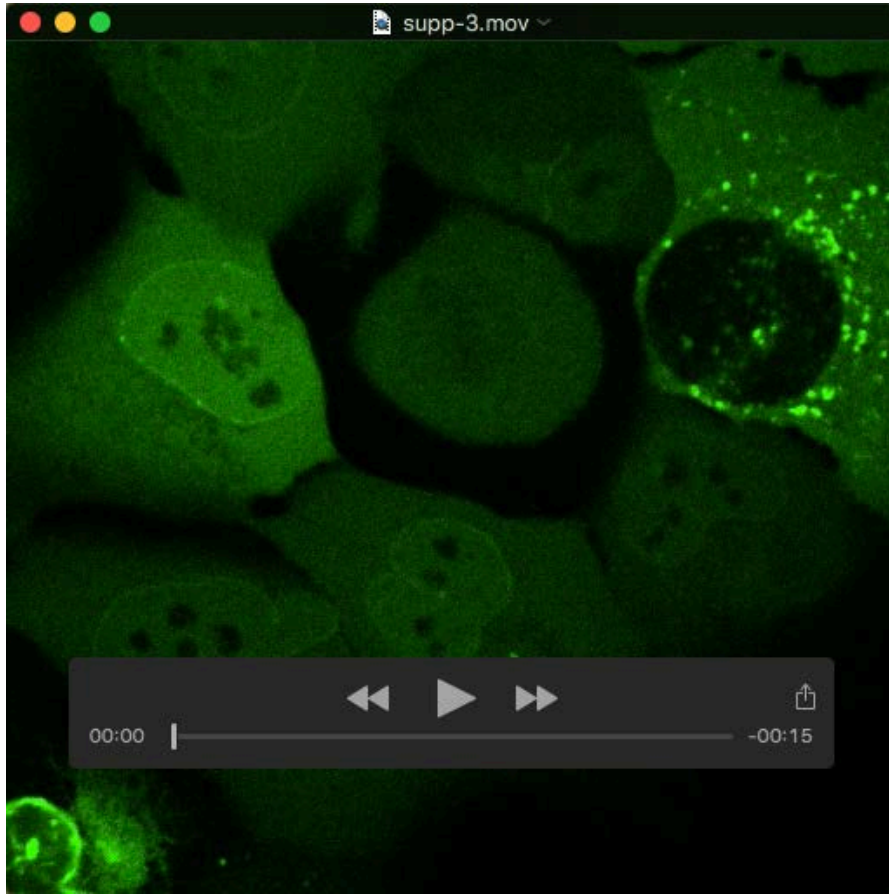
[Click here to Download Table S1](#)

Table S2. PRM (parallel reaction monitoring) mass spectrometry analysis of GRASP55-GFP K50 acetylation in mitotic HEK293T cells after silencing or overexpressing SIRT2.

[Click here to Download Table S2](#)



Movie 1. Live-cell imaging of GRASP55-GFP U2OS cells 72 h after transfection with si Control.



Movie 2. Live-cell imaging of GRASP55-GFP U2OS cells 72 h after transfection with si SIRT2.