

SUPPLEMENTAL FIGURE LEGENDS

Supplemental Figure 1. NUSAP1 mitotic localization and cell cycle regulation. A) NUSAP1 localization through mitosis. NUSAP1 localization to the spindle during mitosis in U2OS cells. Metaphase and Anaphase images are replicated from those shown in Figure 1B. Scale bars indicate 10 μ m. B) U2OS Nocodazole release. Cells were arrested in 150ng/mL Nocodazole overnight and released by mitotic shake-off. Time points were taken every 2 hours until 10h.

Supplemental Figure 2. RanBP2 co-precipitates with NUSAP1 in both Nocodazole and Taxol arrested cells. U2OS cells were either arrested using 200ng/mL Nocodazole or 500nM Taxol overnight before collection. Cells were then used for endogenous NUSAP1 IP.

Supplemental Figure 3. NUSAP1 depletion in U2OS cells does not alter RanBP2 or RanGAP1 localization. Control and NUSAP1 depleted cells were fixed and stained with either RanBP2, Tubulin, and CENP-C or with RanGAP1, Tubulin and NUSAP1. (Scale bars indicate 10 μ M.)

Supplemental Figure 4. NUSAP1 contains a SAP domain in its N-terminus. A) Schematics of NUSAP1 and PIAS1 as a representation of the PIAS family members, which all contain SAP domains approximately 10 AA away from their N-termini. B) Alignment of SAP domains of NUSAP1 and four PIAS protein family members.

Supplemental Figure 5. NUSAP1 depletion does not affect RanBP2 complex formation. HEK-293T cells were depleted of NUSAP1 using one of two different siRNAs or a pool of siRNAs. Endogenous RanBP2 IPs were then performed, indicating RanBP2 is still able to associate with SUMOylated RanGAP1 when NUSAP1 is depleted.