



**Figure S5 Further Mad1 and Mps1 kinetochore SPI analysis.**

(A-B) Mad1-GBP and mad1-A736T-GBP are both recruited to (A) Mif2-GFP and (B) Cep3-GFP, the latter of which shows unusual localization when associated with either Mad1 or its mutant version.

(C) The growth effects of the Mps1-KMN network SPIs (and controls) are shown both in wild-type and *mad3Δ* cells.

(D) We deleted the *MAD3* gene in 21 GFP-tagged kinetochore strains and repeated the assay with Mps1-GBP compared with mps1-kd-GBP. The KMN-Mps1 interactions are all suppressed by deletion of *MAD3*.

(E-G) Fluorescence imaging of cells encoding Spc105-GFP (E), Kre28-GFP (F) and Ndc80-GFP (G), shows Mps1-GBP and mps1-kd-GBP are colocalized with the GFP-tagged kinetochore protein. Fluorescence contrast differs between images and the scale bar is 5  $\mu\text{m}$ .