A. Images showing the localization of Cdc13-GFP and DNA (DAPI) in cells at 2 and 6 hours. 

B. Graph showing the percentage of cells with Cdc13-GFP on SPBs over time after shifting to 18°C, with different genotypes: nda3, nda3 mod2Δ, and nda3 dnt1Δ. 

C. Images showing the localization of Cut2-GFP and Sid4-GFP in different cell cycle stages (Inter, Meta, Ana). 

D. Graph showing the percentage of prometaphase cells with SPBs duplicated or securin positive over time after shifting to 37°C, with different genotypes: psc3-1T, psc3-1T dnt1Δ, psc3-1T sgo2Δ, and psc3-1T bub1Δ.
S2 Fig. Dnt1 is dispensable for activating the SAC in the absence of kinetochore-microtubule attachment or tension.

(A, B) Cyclin B (Cdc13) is accumulated at SPBs in dnt1Δ as efficiently as in wild-type in the absence of kinetochore-microtubule attachment. The indicated strains carrying Cdc13-GFP were arrested at S phase by HU at the permissive temperature (30 °C) for nda3-KM311, and released from the arrest to the restrictive temperature (18 °C). Samples were collected up to 8 hours after release, fixed with methanol and stained with DAPI. Example pictures of nda3-KM311 cells released after 2 or 6 hours are shown in (A). Arrows indicate Cdc13-GFP signals at SPBs. The kinetics of accumulation of Cdc13 at SPBs at each time point was quantified (n > 200 per time point) (B). Scale bar, 5 μm.

(C, D) Dnt1 is not required for activating the spindle checkpoint in the absence of tension. The indicated strains carrying Cut2-GFP and Sid4-GFP were arrested at S phase by HU at 25 °C and released to 37 °C. Examples of Cut2-GFP and Sid4-GFP images at interphase (Inter.), metaphase (Meta.) and anaphase (Ana.) are shown in (C). Prometaphase (SPB duplicated and Cut2-positive) cells were counted at each time point (n > 200) (D). Note that Cut2-GFP, but not Sid4-GFP at SPBs, disappears from the anaphase cells. Scale bar, 5 μm.