Głuszek et al., http://www.jcb.org/cgi/content/full/jcb.201507006/DC1

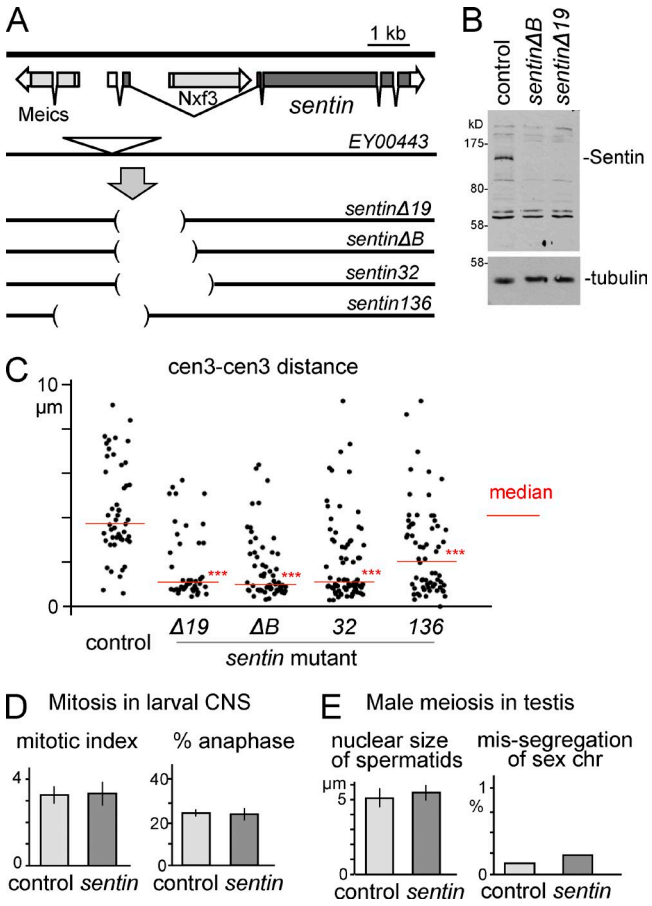


Figure S1. ***sentin* mutants are viable but female sterile.** (A) The genomic region around *sentin* in wild-type, *sentin* mutants (*Δ19*, *ΔB*) and their parental line with a *P*-element insertion (triangle). (B) Western blots of adult females for Sentin and α-tubulin. (C) The distances between homologous centromeres from FISH probed by dodeca satellite (cen 3). ***, $P < 0.001$, significant difference from the medians of wild-type control. (D) Mitotic index and percentage of anaphase among mitosis in central nervous systems (CNS) of wild-type control and *sentin*-mutant larvae ($n = 5$ and 4). (E) Nuclear diameter of spermatids at onion stage and missegregation rate of sex chromosomes in male meiosis ($n = 59$, 213, 1,451, and 434). Error bars indicate the SEM (left) and 95% confidence intervals (right).

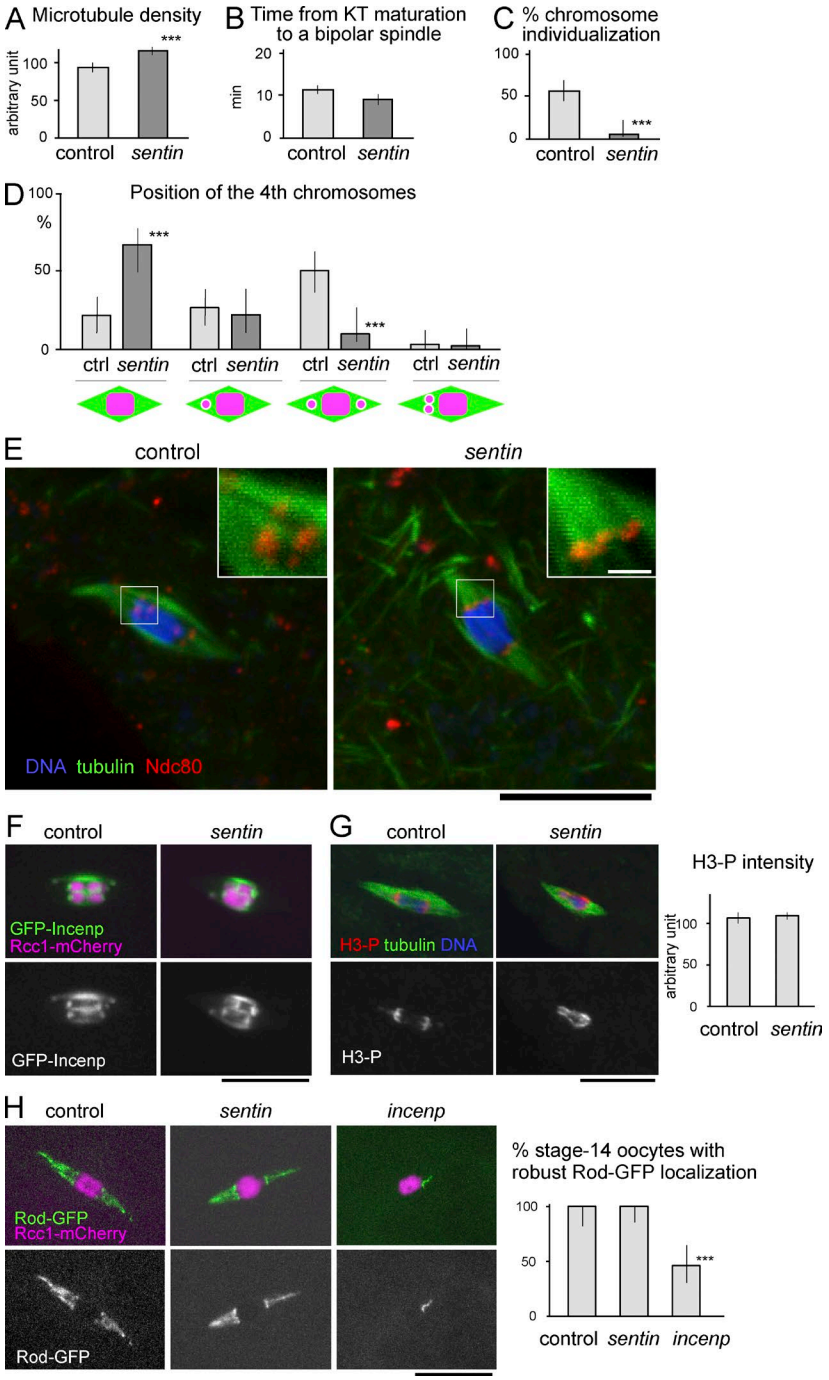


Figure S2. **Phenotype of *sentin*-mutant oocytes with unaltered Aurora B activity and localization.** (A) Microtubule density estimated by the maximum tubulin signal intensity of the spindle in fixed stage 14 oocytes (n = 56 and 64). Error bars represent SEM. (B) Time taken from kinetochore maturation (full recruitment of Rod-GFP) to establishment of the spindle bipolarity in live oocytes expressing Rod-GFP and Rcc1-mCherry. Rod-GFP streams were used to estimate the timing of establishment of spindle bipolarity, which was consistent with the timing from live imaging using GFP-tubulin (n = 5). Error bars represent SEM. (C) The proportion of DAPI-stained mature oocytes in which at least one chromosome can be recognized separately from others (n = 59 and 51). (D) The proportion of DAPI-stained mature oocytes with the following location of the two fourth chromosomes (small dots). From left to right; both are located together with the other chromosomes; only one is located closer to the poles and separated from the other chromosomes; each of the two is located closer to the opposite pole and separated from the other chromosomes; both are located closer to the same pole and separated from the chromosome mass (n = 58 and 41). All error bars in C and D represent 95% confidence intervals. ***, $P < 0.001$, significant difference from the control. (E) High-magnification images of the meiotic spindle in mature oocytes from wild-type control and *sentin*$^{32/Df}$ mutant. The insets (bars, 1 µm) are magnified images of the areas indicated in the boxes. (F) Wild-type control and *sentin*-mature oocytes expressing GFP-Incenp and Rcc1-mCherry driven by *nanos-GAL4*. GFP-Incenp localization to the spindle equator and centromeres was not affected by the *sentin* mutation. (G) Immunostaining images of phospho-H3-Ser10 (H3P), α-tubulin, and DNA in control and *sentin*-mature oocytes. The maximum intensities of H3-P signals were measured. No significant difference was observed. Error bars represent SEM. (H) Wild-type control, *sentin*, and *incenp*^{QA26}-mature oocytes expressing Rod-GFP and Rcc1-mCherry. The proportion of robust Rod-GFP localization in oocytes was shown with each genotype. The results showed that full Rod recruitment requires the Aurora B activity as shown in mammalian mitotic cells, but it is not affected in the *sentin* mutant, implying that Aurora B activity is not compromised in *sentin*-mutant oocytes. The error bars represent 95% confidence intervals. ***, $P < 0.001$ (n = 18, 21, and 30). Bars, 10 µm.

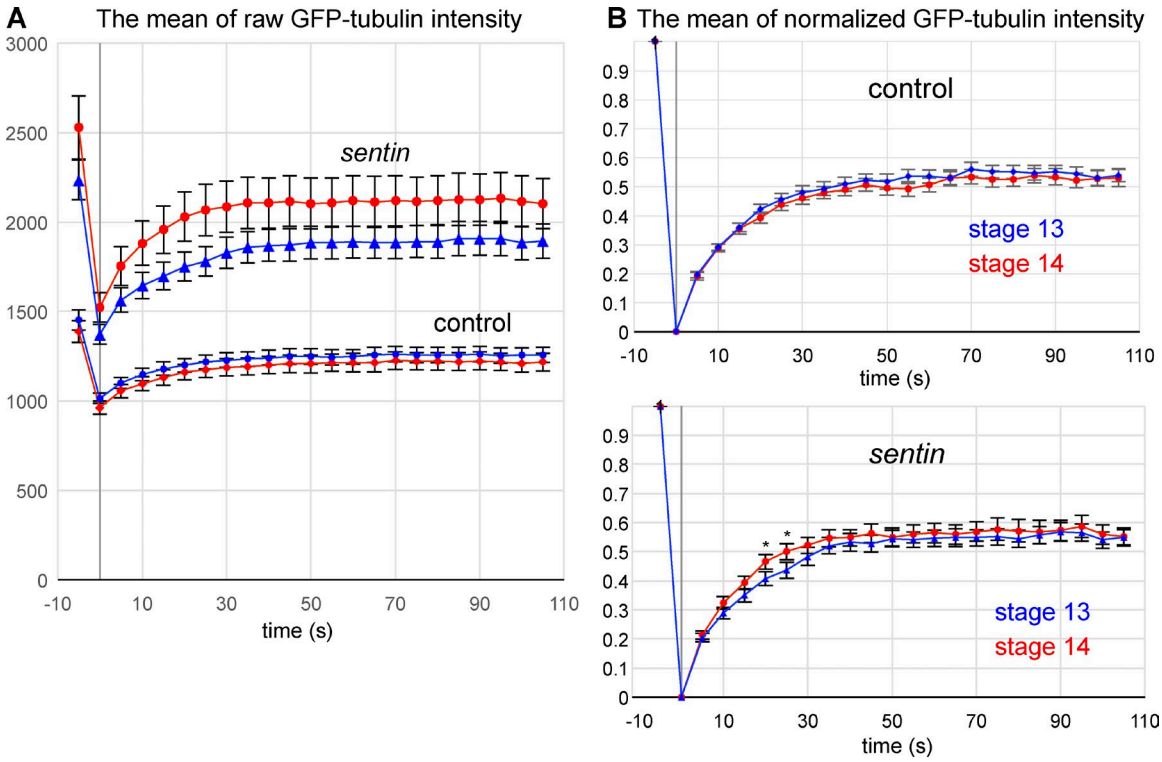
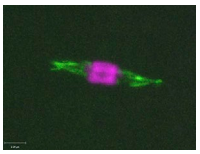
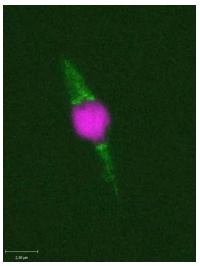


Figure S3. **FRAP analysis of GFP–α-tubulin in control and *sentin*-mutant oocytes.** Oocytes at stages 13 and 14 expressing GFP–α-tubulin and Rcc1-mCherry driven by n*anos-GAL4* were used. GFP-tubulin of a half spindle was photobleached, and the recovery was followed. (A) Raw intensity values are plotted against time. GFP-tubulin intensity in the *sentin* mutant is higher than the control, consistent with immunostaining results. (B) The signal intensity was normalized so that one corresponds to the prebleached value and zero corresponds to the value at the first time point after bleaching (n = 21–27). Error bars represent the SEM. *, P < 0.05, significant difference between the two stages of *sentin* (one-tailed t test).

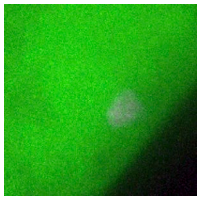


Video 1. **Wild-type stage 14 oocyte expressing Rod-GFP Rcc1-mCherry.** 20 µm × 15 µm × 20 min.

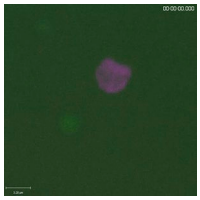


Video 2. ***sentin* stage 14 oocytes expressing Rod-GFP Rcc1-mCherry.** 15 µm × 20 µm × 20 min.

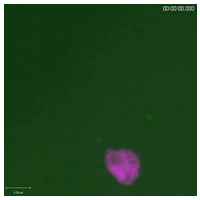
Video 3. **Wild-type stage 13 oocyte expressing GFP-Mis12 Rcc1-mCherry.** 18 µm × 18 µm × 90 min.



Video 4. ***sentin* stage 13 oocyte expressing GFP-Mis12 Rcc1-mCherry.** 18 µm × 18 µm × 90 min.



Video 5. **Wild-type stage 13 oocyte expressing Rod-GFP Rcc1-mCherry.** 25 µm × 25 µm × 60 min.



Video 6. ***sentin* stage 13 oocyte expressing Rod-GFP Rcc1-mCherry.** 25 µm × 25 µm × 60 min.