

Supplemental material

Martinez-Garcia et al., <https://doi.org/10.1083/jcb.201803019>

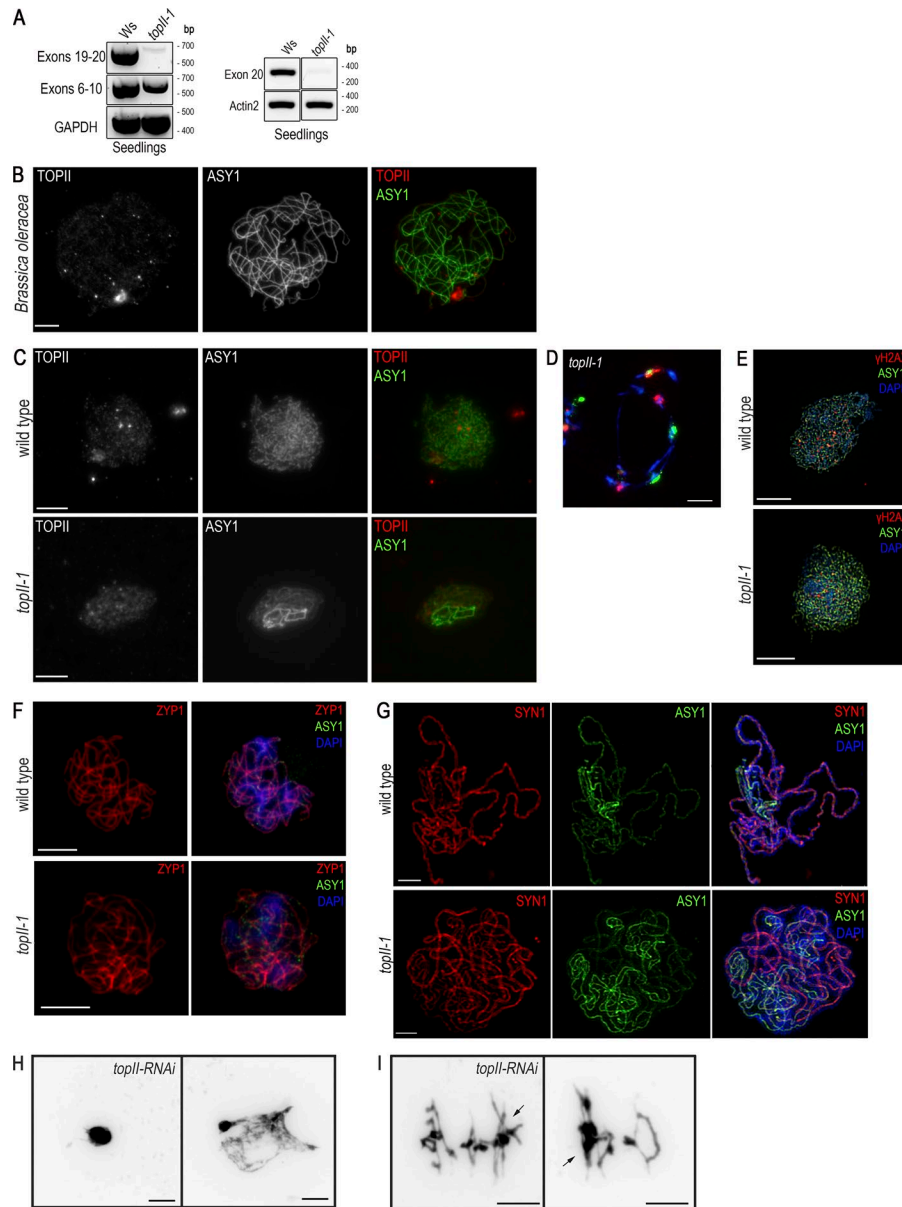


Figure S1. **Cytogenetic analysis of *topII-1* and *topII-RNAi* lines.** (A) Expression of *TOPII* assessed by RT-PCR of seedlings of *topII-1* and WT. (B) Immunolocalization of TOPII (mouse) and ASY1 in prophase I cells of *B. oleracea*. (C) Immunolocalization of TOPII (rabbit; Agrisera) in WT (Ws) and *topII-1* meocytes. (D) Anaphase I cell of *topII-1* with FISH probes. Bivalents 2 and 4 reveal chromatin bridges between homologues. (E) Immunolocalization in early prophase I cells of  $\gamma$ H2AX and ASY1 in WT and *topII-1*. (F) Immunolocalization in fully synapsed cells of ZYP1 and ASY1 in WT and *topII-1*. (G) Immunolocalization in zygotene cells of SYN1 and ASY1 in WT and *topII-1* by SIM. (H) Representative chromosome spread images of *topII-RNAi* PMCs with highly condensed nuclei during zygotene and diplotene. (I) Representative chromosome spreading images of *topII-RNAi* metaphase I cells with interlocks (arrows). Bars: 5  $\mu$ m (B–E and G–I); 2  $\mu$ m (F).

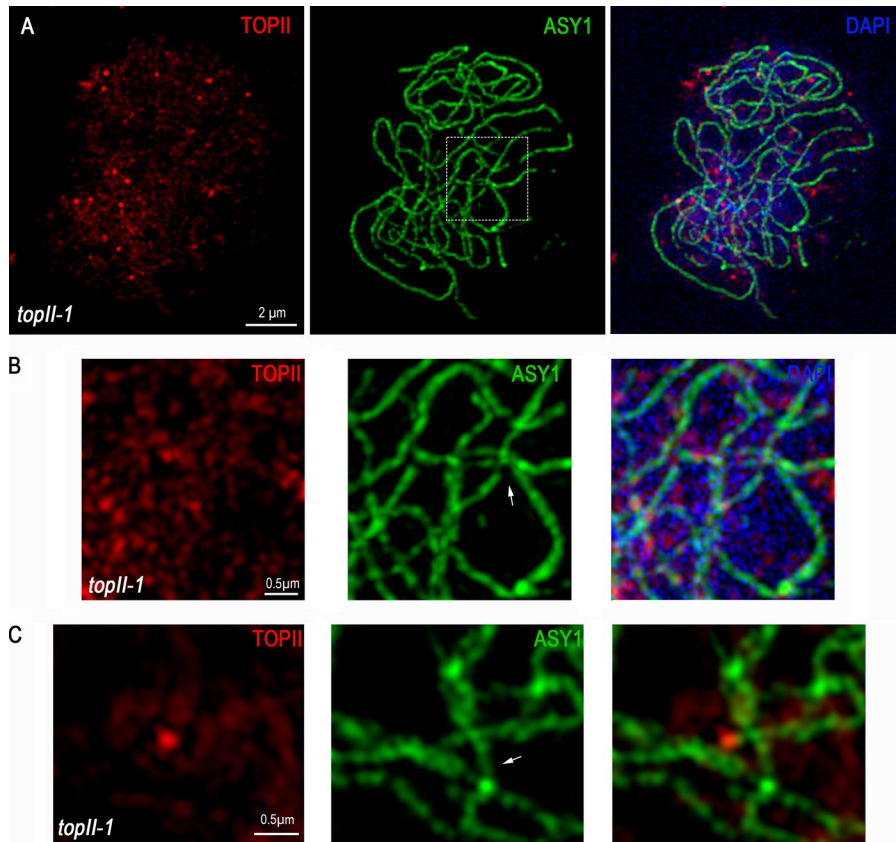


Figure S2. Immunolocalization of TOPII (red) and ASY1 (green) in prophase I of *topII-1* meicytes using SIM. (A) Late zygotene stage. (B) Zoomed region with axis entanglements (arrow) without TOPII (red) accumulation. (C) Zoom in an interlock (arrow) with a dot of TOPII signal.

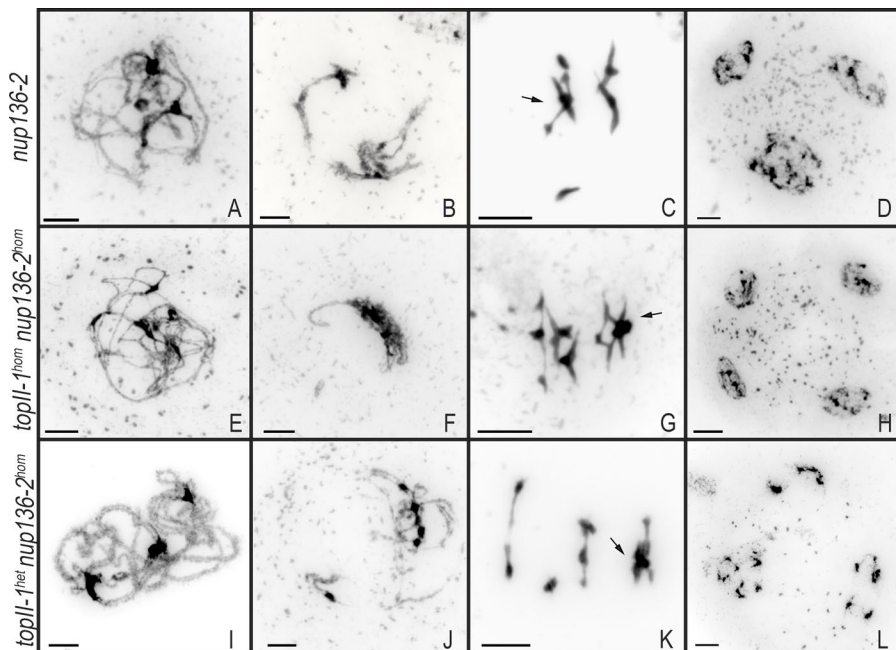
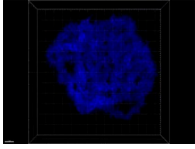
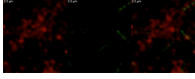


Figure S3. Representative chromatin spreading images of *nup136-2*, *topII-1<sup>hom</sup> nup136-2<sup>hom</sup>*, and *topII-1<sup>het</sup> nup136-2<sup>hom</sup>* PMCs. (A–D) *nup136-2* PMCs. (E–H) *topII-1<sup>hom</sup> nup136-2<sup>hom</sup>* PMCs. (I–L) *topII-1<sup>het</sup> nup136-2<sup>hom</sup>* PMCs. (A, E, and I) Pachytene-stage cells. (B, F, and J) Diakinesis stage. (C, G, and K) Metaphase I cells with interlocks (arrows). (D, H, and L) Tetrad stages.



Video 1. **3D rendering of a SIM image stack from a *Ws* late zygotene cell with TOPII (red) and ASY1 (green) immunolocalization.** Chromatin (blue) was counterstained with DAPI. Bar, 1  $\mu$ m. Frame rate, 24 frames per second.



Video 2. **3D-SIM image stack of a *Ws* meiocyte with an axis break (ASY1 in green) and TOPII accumulation (red).** Chromatin (blue) was counterstained with DAPI. Frame rate, three frames per second.