



**Figure S3. Variation in *Mutator* foci intensity across gonad regions.**

(A) Schematic of a wild-type adult hermaphrodite gonad. (B) Gonad divided by length into 10 equal regions ending at the first single-file diplotene/diakinesis nucleus (yellow borders). After identical thresholding, image shows only above-threshold foci (white) which were quantified using the analyze particles function in FIJI. (C) *mut-16::gfp::3xFLAG; him-8(tm611)* mutants display an extended transition zone (white dashed line) indicated by DAPI-visualized transition nuclei throughout the gonad (white arrows). MUT-16 foci (green) intensity pattern remains similar to wild-type. (D) *chk-2 RNAi* disrupts polarization of DAPI-visualized transition zone nuclei (yellow dashed line). MUT-16 foci (green) intensity pattern remains similar to the wild-type. (E) Double *gld-2 gld-1 RNAi* on *mut-16::gfp::3xFLAG* animals produces a large proximal mitotic tumor (asterisks). Numerous bright MUT-16 foci (green) are found throughout the mitotic tumor. Some SYP-1 (red) staining is present in the more distal region (diamond). DAPI-visualized DNA (blue) reveals an abundance of small mitotic nuclei in the proximal tumor. Note that the complete distal tip of the gonad is not shown. (F) Quantification of total foci per gonad region in wild-type (n = 3), *gld-2 gld-1 RNAi* (n = 4), and *atx-2 RNAi* (n = 4) germlines. Each line represents one gonad. All scale bars, 15  $\mu$ m.