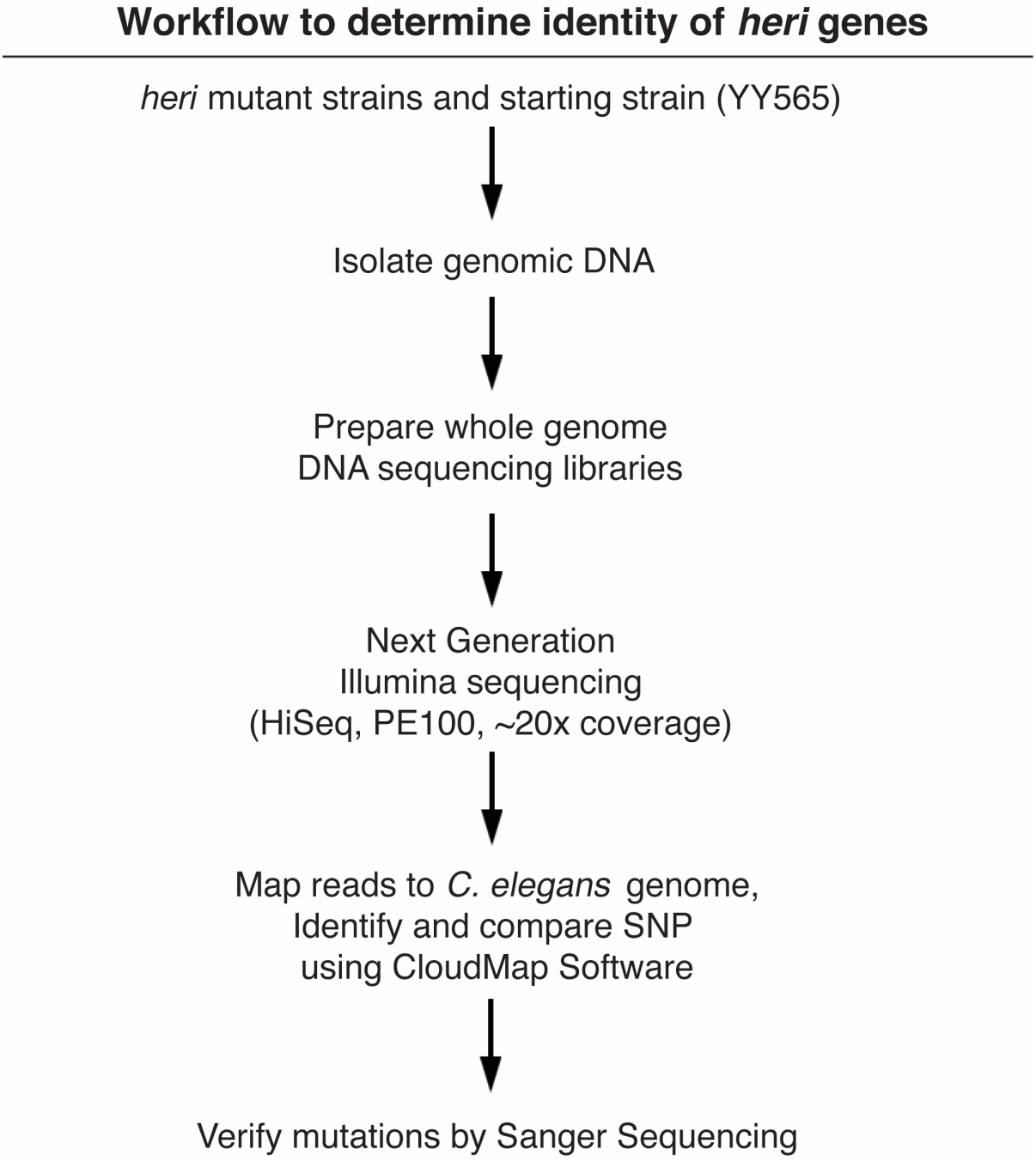


Figure S1. Identifying Heri genes. Workflow used to identify *heri-1.* CloudMap software is described in (Minevich *et al.* 2012).

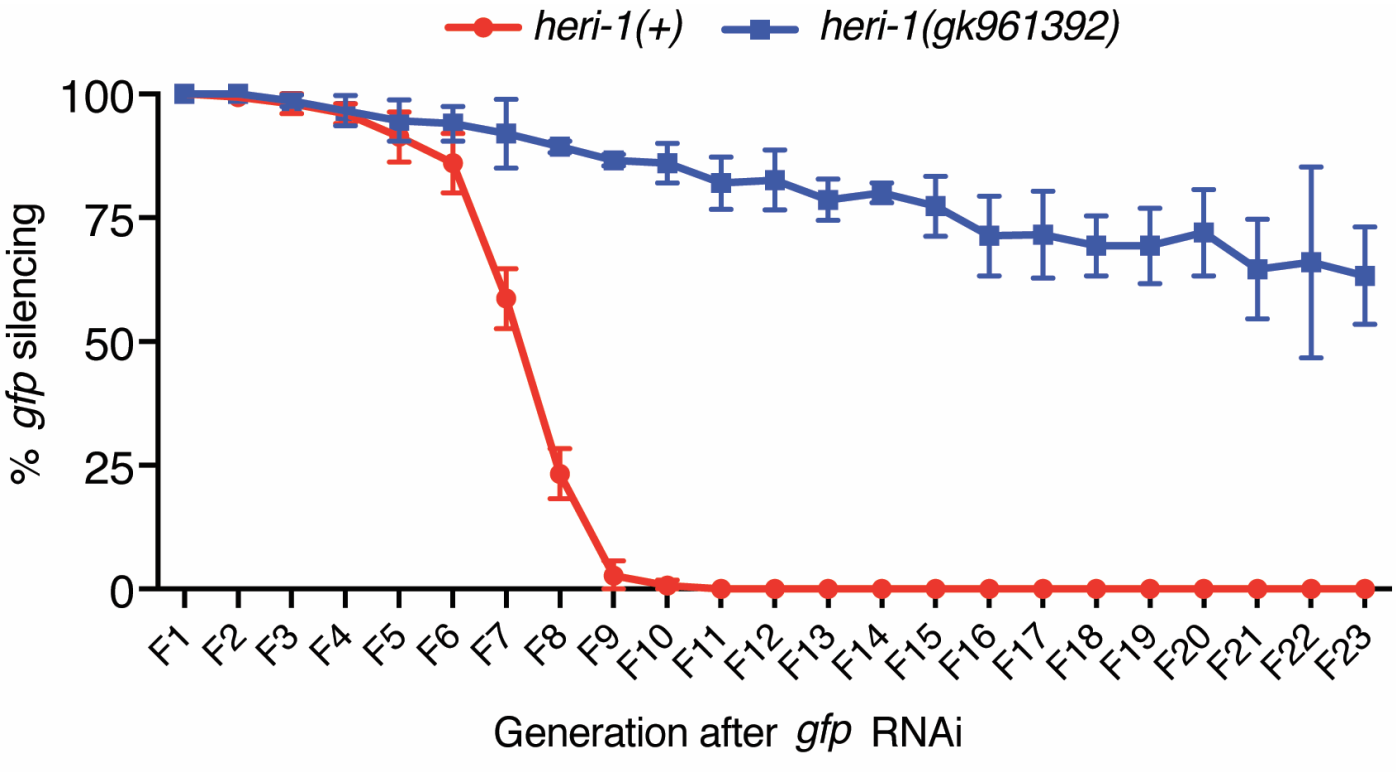
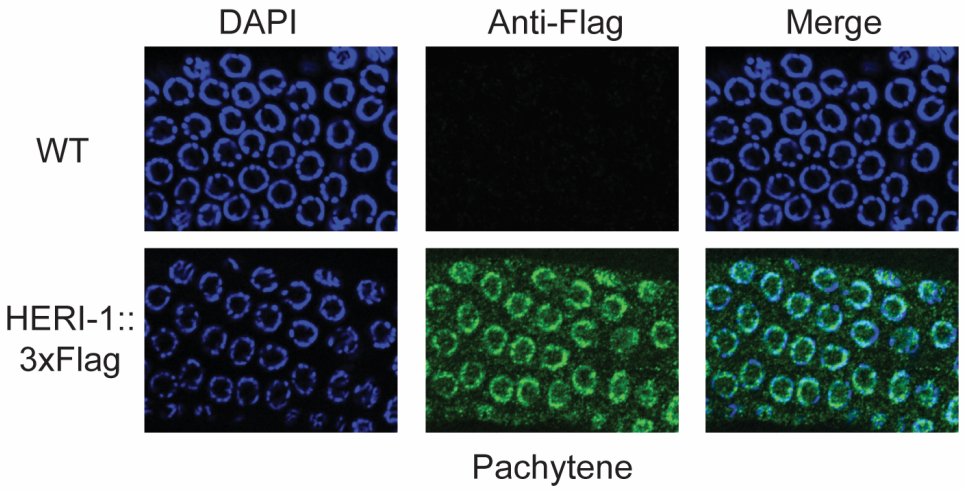


Figure S2. *heri-1(gk961392)* enhances TEI. Animals of indicated genotypes that expressed a *pie-1::h2b::gfp* transgene were treated with *gfp* RNAi (see methods). Data points represent 3 biological replicates in which 50 animals were scored for GFP expression. Experiment was performed blind and error bars represent standard deviations of the mean.

A


DAPI
Anti-Flag
Merge
WT
HERI-1::
3xFlag
Pachytene

B

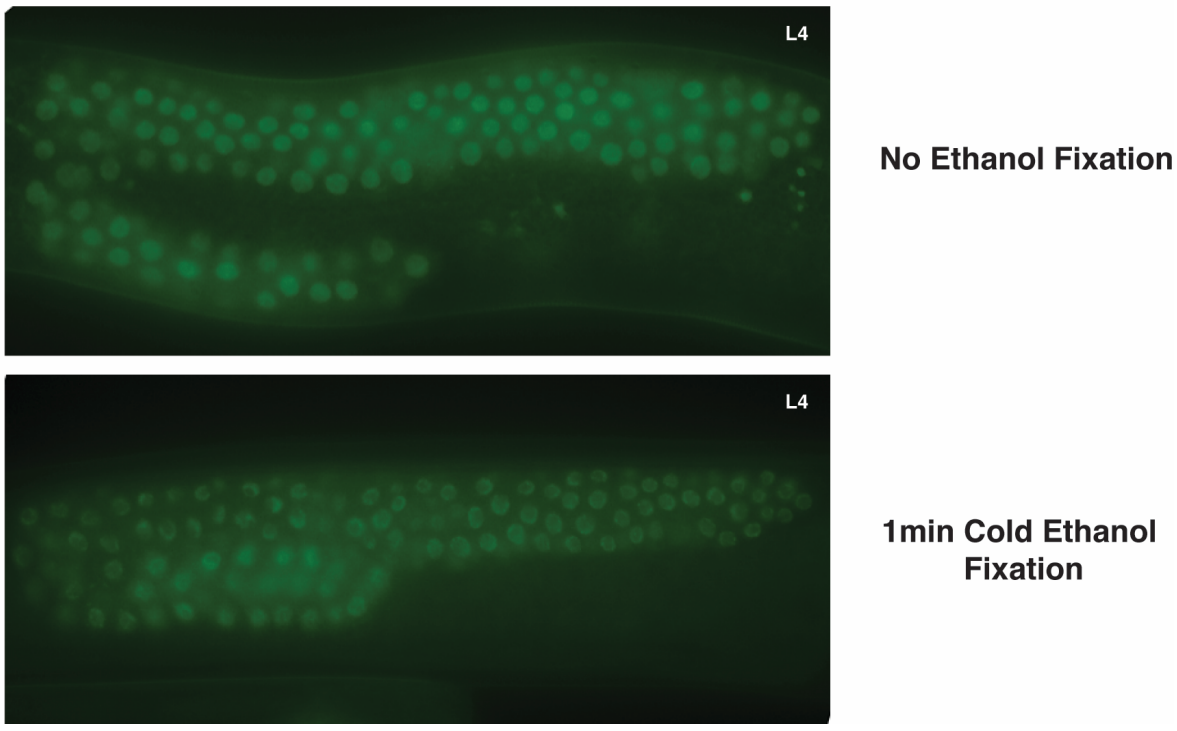

HERI-1::GFP::3xFLAG
L4
No Ethanol Fixation
L4
1min Cold Ethanol
Fixation

Figure S3. HERI-1 localizes to nuclei. A) Micrographs show immunofluorescent (IF) images of fixed adult pachytene germ cell nuclei in wild type and HERI-1::3xFLAG detected with α-FLAG antibodies. DAPI staining (DNA) is shown in blue. Note that the subnuclear distribution of HERI-1 appears more peripheral in these micrographs that when we visualized HERI-1::GFP in the main text (Fig. 3A). The data in (B) hint at why this might be. **B)** Micrographs of larval stage 4 (L4) germlines without ethanol fixation (top panel) or with -20ºC ethanol fixation for 1 min (bottom panel). During ethanol fixation, germlines of L4 animals were dissected onto a subbed slide, placed on a cover slip**,** and snap frozen on dry ice for 10 mins. Cover slips were removed and the gonads were fixed in -20ºC ethanol in a coplin jar for 1 min. Slides were allowed to air dry for 5 mins and excess ethanol was removed and gonads were imaged under our inverted microscope. This fixation process seems to alter the sub-nuclear distribution of HERI-1 in a way that makes HERI-1 appear to colocalize with chromatin. Notice the localization of HERI-1::GFP::3xFlag closer to the nuclear periphery after fixation.

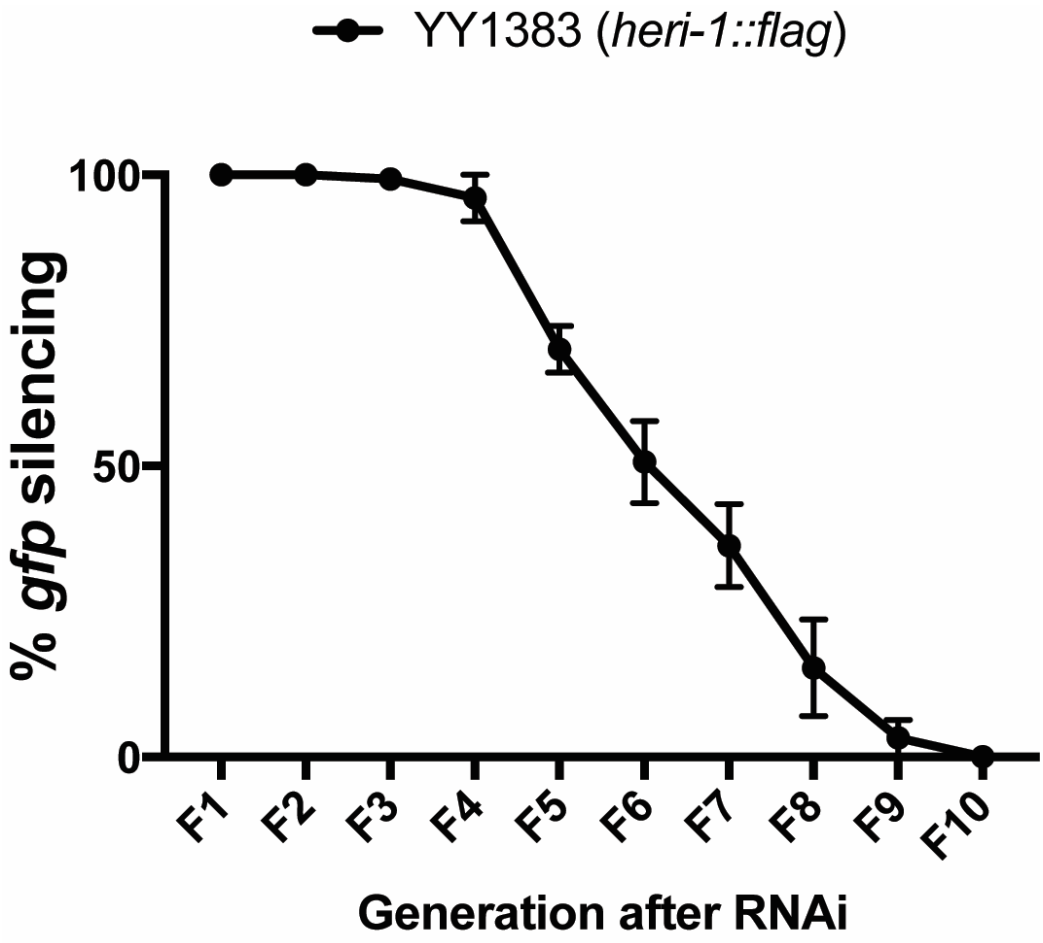


Figure S4. HERI-1::3xFLAG is functional for RNAi inheritance. *heri-1::flag* animals that express the *pie-1::h2b::gfp* transgene were treated with *gfp* RNAi (see methods) and the percentage of animals silencing GFP across generations is indicated. Data points represent 3 biological replicates in which 50 animals were scored for GFP expression. Error bars represent standard deviations of the mean. Because *gfp* inheritance silencing looks similar to what we always see in wild type animals (compare Figure 2 and Figure S3), we conclude that HERI-1::FLAG is functional. Note, we have not tested HERI-1::GFP::FLAG animals in this assay because these animals harbor two *gfp* loci.

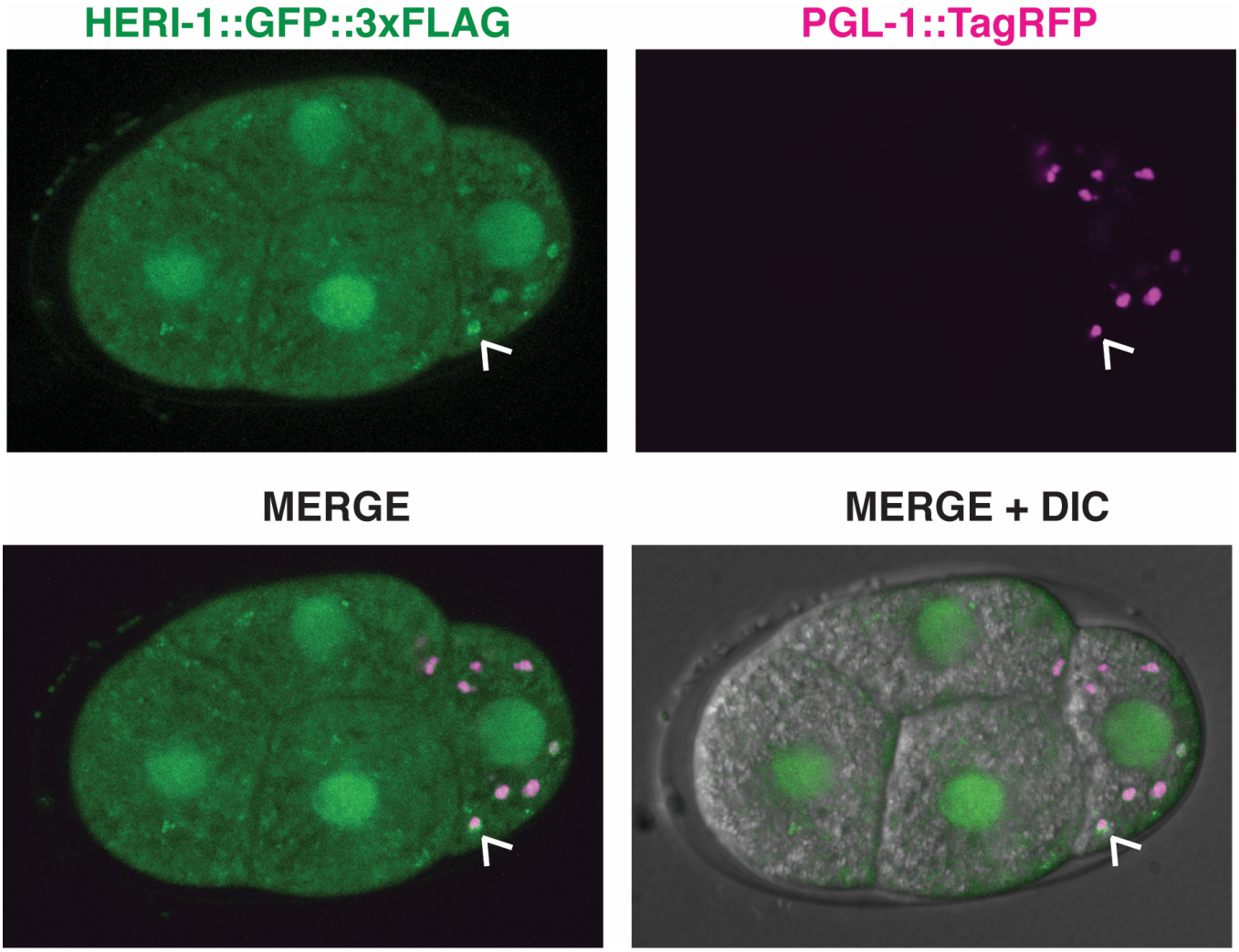


Figure S5. HERI-1 is expressed in somatic and germline blastomeres during early development. Micrograph of a 4 cell embryo expressing HERI-1::GFP::3xFLAG. HERI-1 is present in all four nuclei. In the P2 cell, some HERI-1::GFP::3xFLAG is present in the cytoplasm and this cytoplasmic signal co-localizes with PGL-1::TagRFP, which is a marker of P granules (arrowhead).

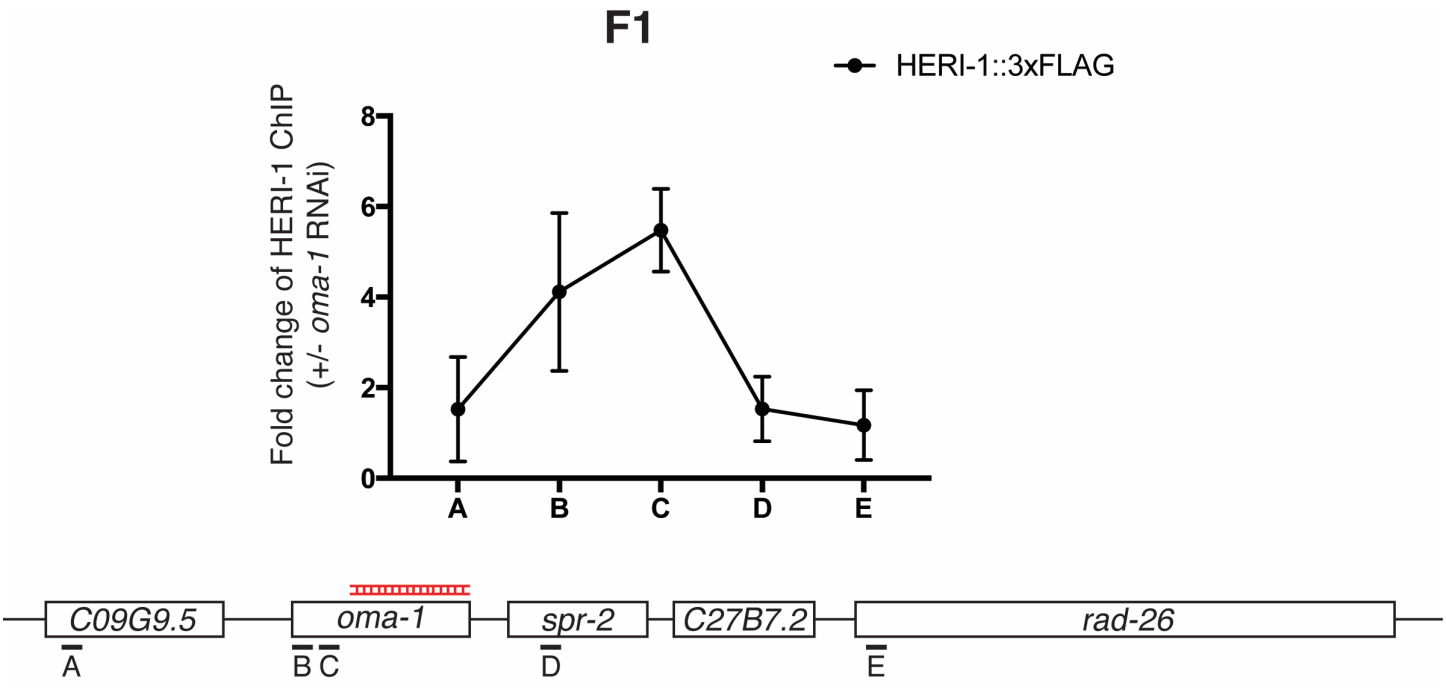


Figure S6. HERI-1::3xFLAG binds to chromatin in response to RNAi. FLAG ChIP-qPCR on progeny of HERI-1::3xFLAG animals treated +/- *oma-1* RNAi. Data is expressed as a ratio of FLAG ChIP signals in animals treated with *oma-1* RNAi over non-RNAi control animals. Data is from 3 biological replicates and error bars are standard deviations of the mean. Pattern is similar to what we observed with HERI-1::GFP in Fig. 5.

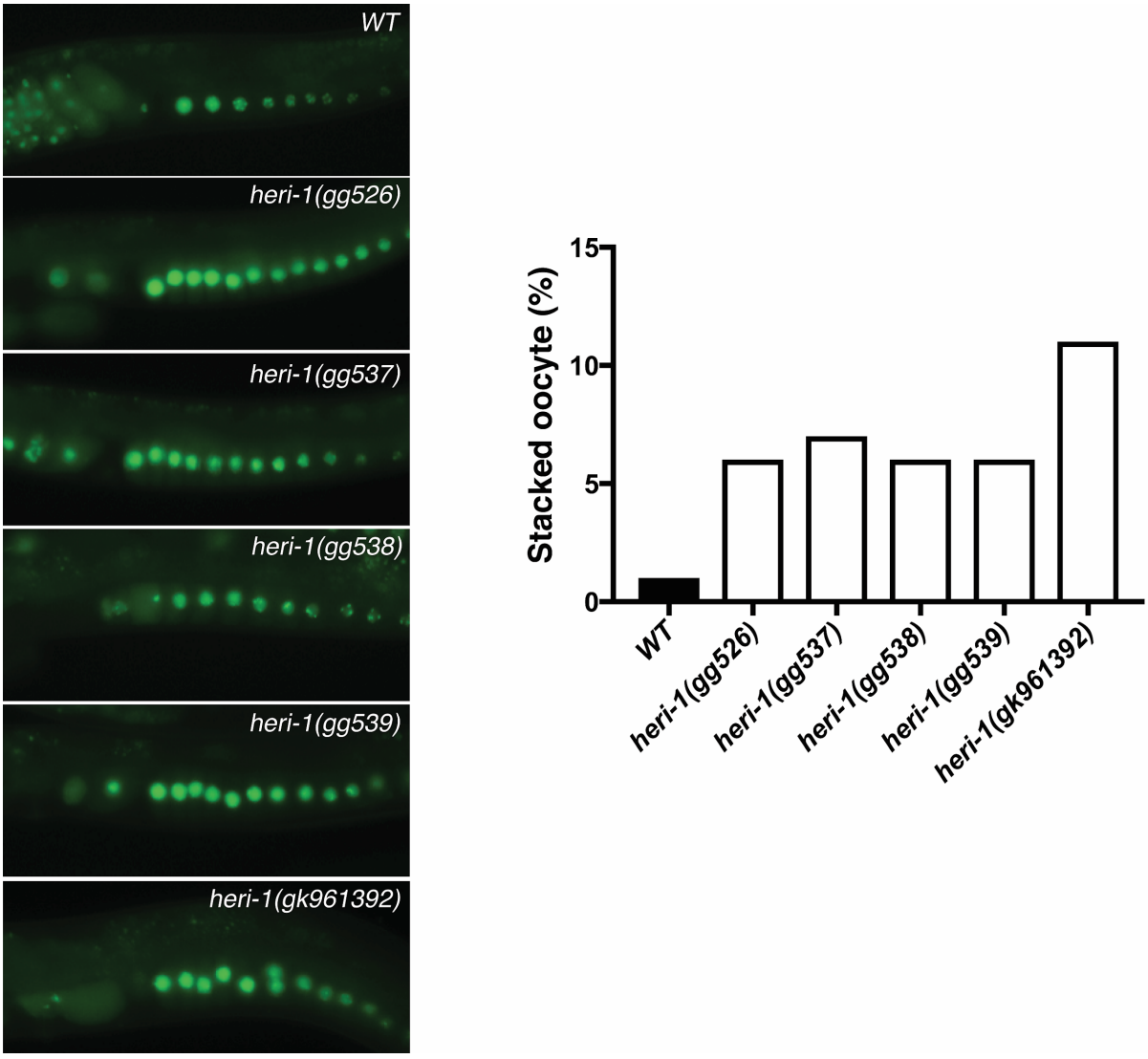


Figure S7. *heri-1* mutant animals show a "stacked oocyte" phenotype. Micrographs of stacked oocyte defects in five different *heri-1* mutants strains. *gg526*, *gg537*, *gg538* and *gg539* were from Heri screen. *gk961392* was from the million mutation project. All animals are expressing a *pie-1::gfp::h2b* reporter gene. Quantification (50 animals scored/strains) is shown on the right.

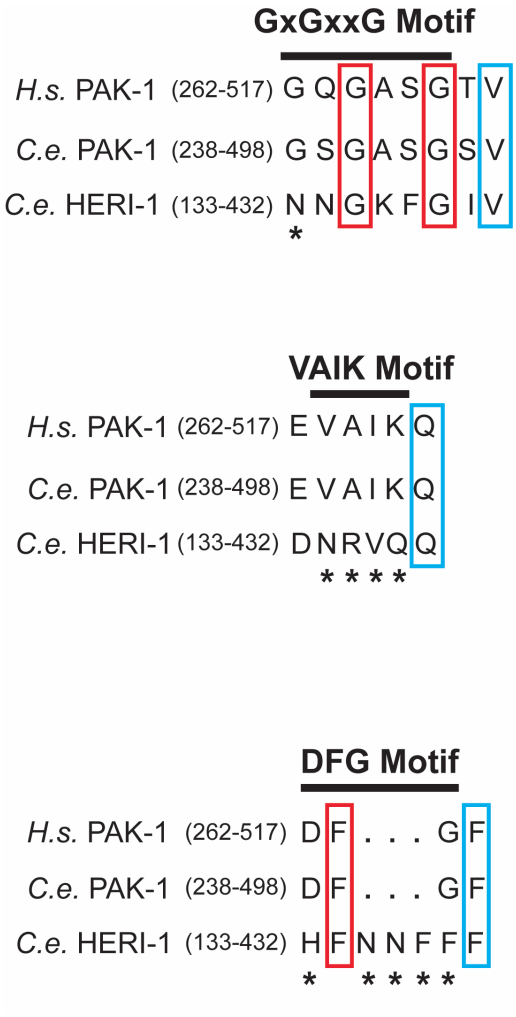


Figure S8. HERI-1's kinase motifs are degenerate. Sequence alignments of three motifs (GxGxxG, VAIK and DFG), which contribute to kinase activity in most protein kinases, from human PAK-1, C. *elegans* PAK-1 and HERI-1. Motif location is indicated by the black bar. Amino acids in red rectangles indicate conservation within the motif, while amino acids in blue rectangles indicate conservation outside the motif. Asterisks indicate lack of conservation.

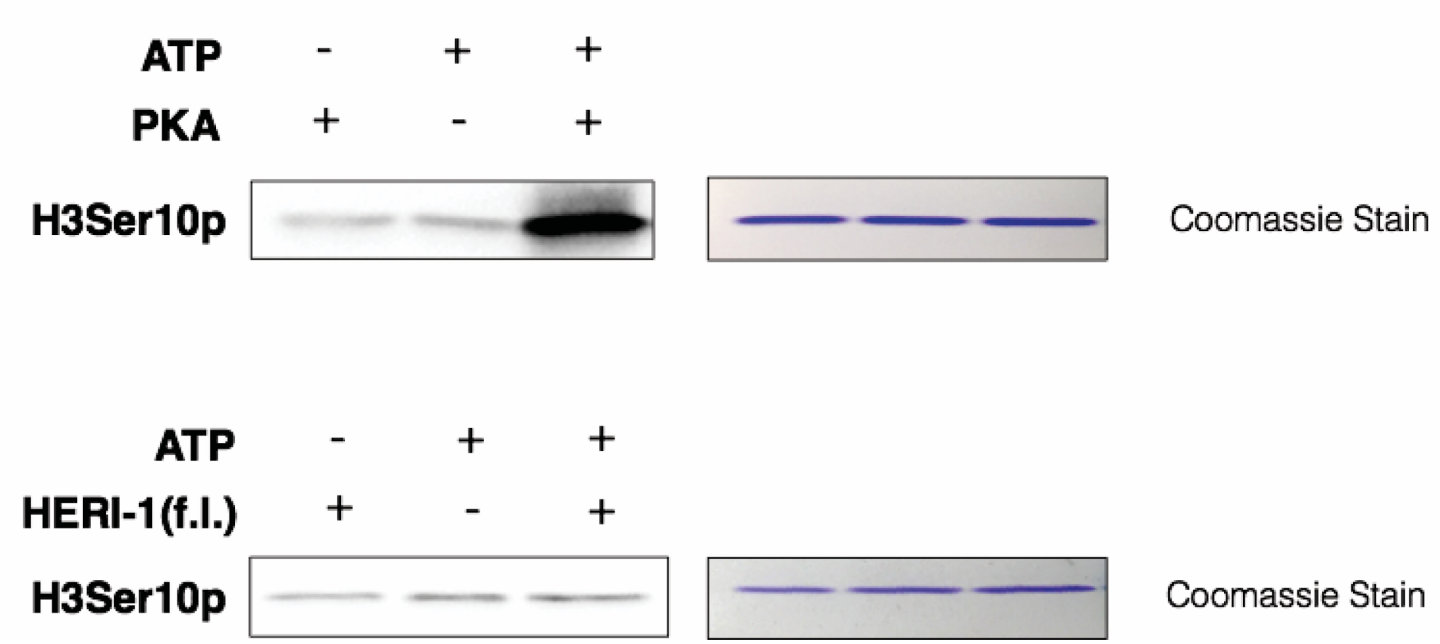


Figure S9. No kinase activity detected with recombinant HERI-1 protein *in vitro*. 1ug of purified Histone H3 was incubated with +/- ATP and +/- cAMP-dependent protein kinase (PKA) (NEB, cat. no. P6000S) (top panel), or with full length HERI-1 recombinant protein (bottom panel). Kinase reactions were performed in kinase assay buffer (200mM final ATP and 1x Kinase Buffer (NEB) for 1hr at 37º in a final volume of 15uL). After incubation, 15uL of 2x Loading buffer were added to each reaction. Incubated at 95º for 5 min, and resolved in a 7.5% Polyacrylamide gel. Detection of H3Ser10p was determined by using the anti-H3Ser10p antibody from Millipore (clone CMA312) at a dilution of 1:1000. Purified Histone H3 samples stained with coomassie blue are shown as loading controls. Full length (f.l).

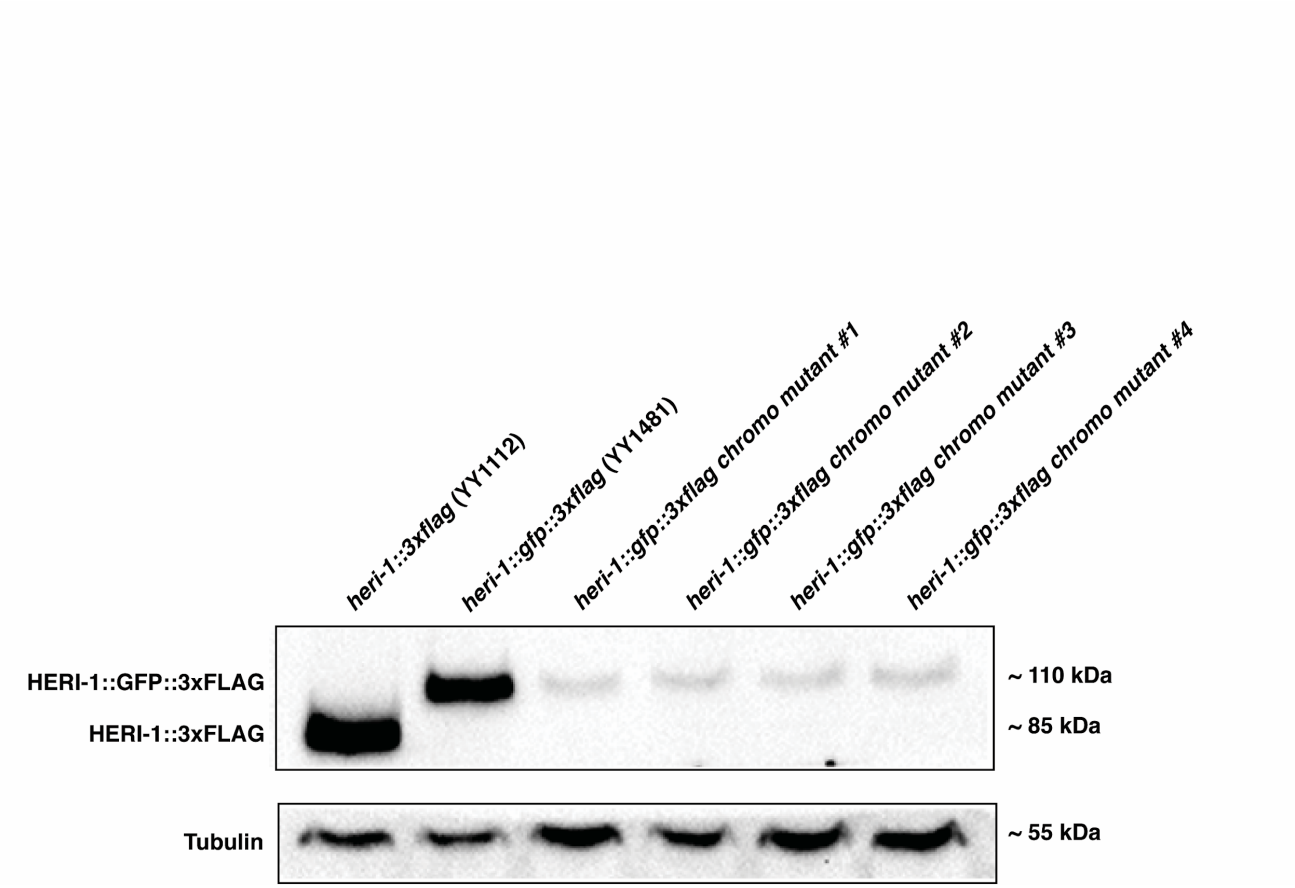


Figure S10. HERI-1(V60D) is unstable. *gg526* is a *heri-1* mutation identified by our screen that mutates the HERI-1 chromodomain (V60D, see Fig. 2). Western blot analysis comparing levels of HERI-1::3xFlag, HER-1::GFP::3xFlag, and HERI-1(V60D)::GFP::3xFLAG is shown. α-Flag antibody (Sigma, M2) was used to detect proteins and a α-tubulin antibody was used for loading control. Shown are 4 lines (mutant 1-4) of HERI-1(V60D)::GFP::3xFLAG animals produced by CRISPR- based *gfp::3xflag* tagging of *heri-1(gg526)*.