

SUPPLEMENTARY FIGURES

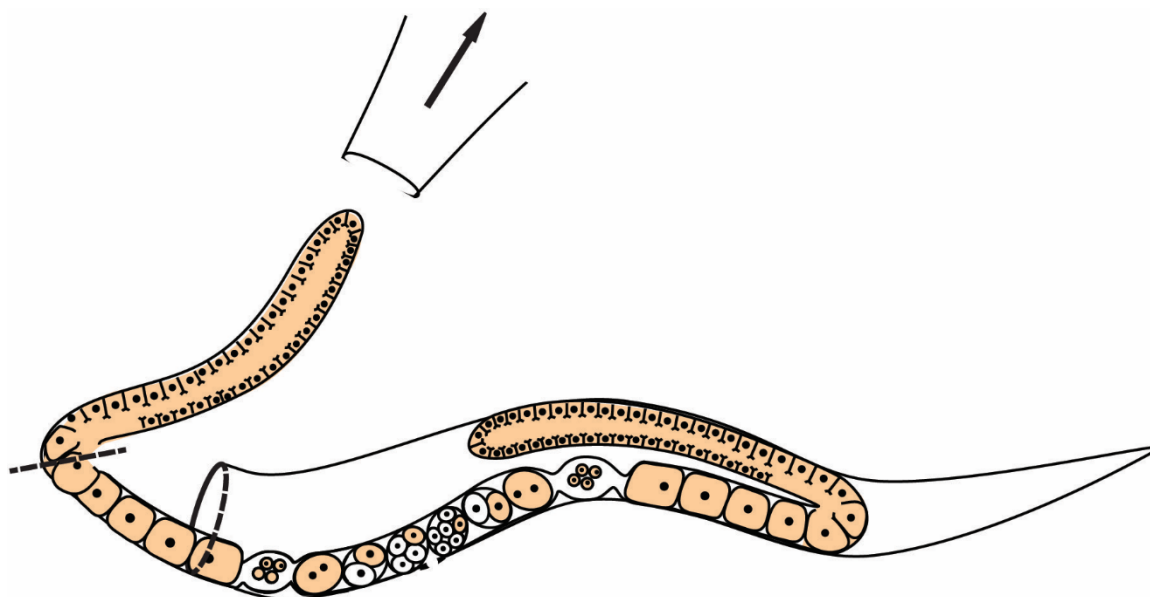


Figure S1. Germline dissection method. Heads were removed with 27.5 gauge needles, allowing the distal portion of the germline to pop out of the worm. Glass capillaries pulled with an opening just large enough to fit the end of the germline were used to rapidly detach them at the ventral to distal bend, eliminating the need to first dissect germlines away from the worm, while at the same time excluding more mature oocytes and the spermatheca from samples. Detached germlines were suctioned using an aspirator tube assembly system (Sigma A5177). Using this method instead of first detaching germlines with a scalpel improved sample consistency and allowed us to harvest approximately five times as many germlines in the same duration.

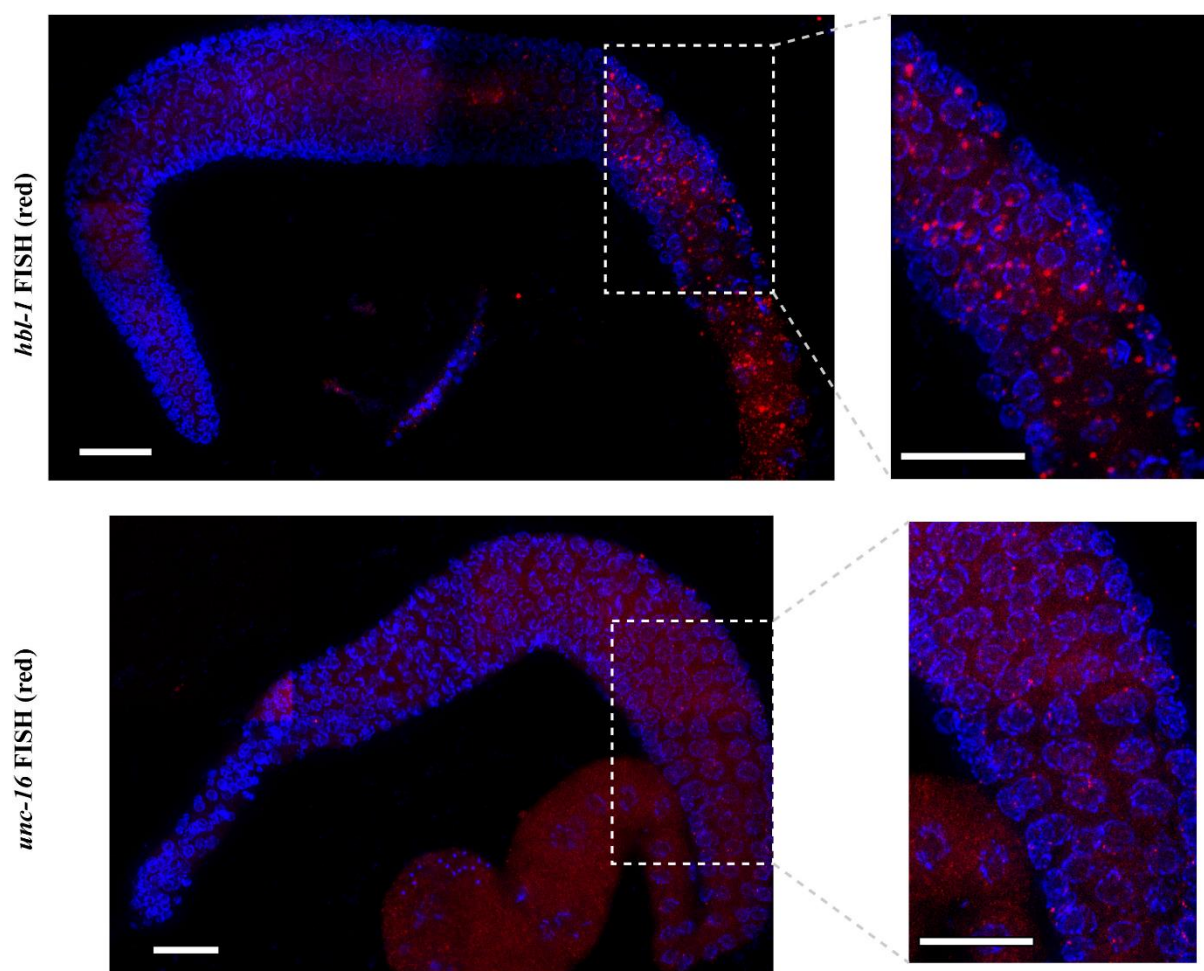
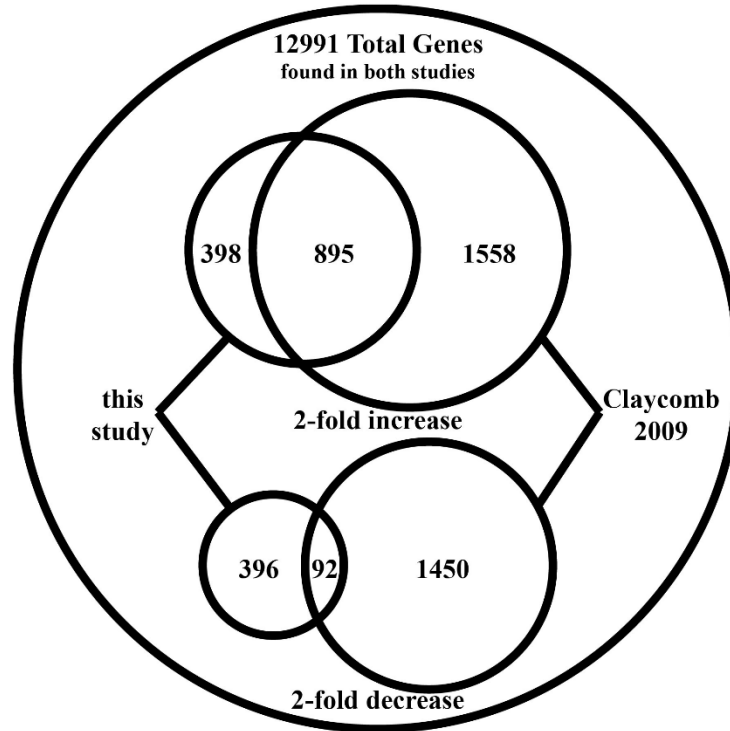


Figure S2. Germline expression of soma-specific transcripts. *hbl-1* and *unc-16* transcripts in dissected germlines of wild-type adult worms. Red – FISH probe. Blue – DAPI/DNA. Insets zoom in on pachytene germ cells. Scale bar: 20 μ m.

A Genes Up and Down >2-Fold in *csr-1*
 (dissected germlines, *csr-1* RNAi, mRNA-seq, this study) to
 (whole worms, *csr-1* mutant, tiling array, Claycomb 2009)



B mRNA-seq Expression Change in *csr-1* RNAi Germlines
 Compared to *csr-1* Gene Sets in Whole Worms

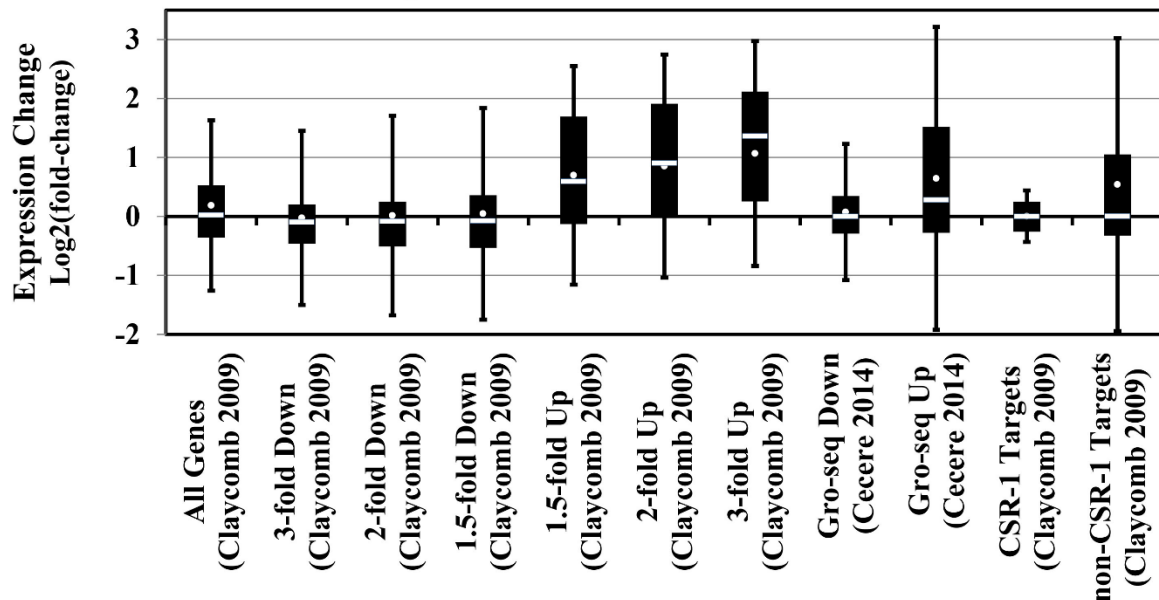


Figure S3. Comparison of *csr-1* RNAi germline expression to *csr-1* gene sets in whole worms. A) Genes up and down-regulated >twofold in dissected *csr-1* RNAi germlines (mRNA-seq, this study) compared to *csr-1* whole worms (tiling microarray, Claycomb 2009). B) Comparison of expression changes in *csr-1* RNAi germlines compared to *csr-1* gene sets in whole worms. Box, upper and lower quartiles; line, median; dot, mean; bars, standard deviation from mean.

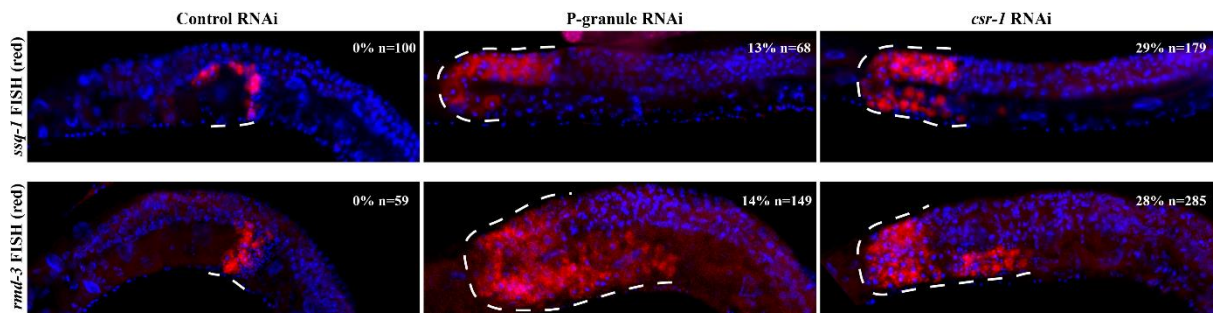


Figure S4. Distal expansion of *ssq-1* and *rmd-3* following P-granule and *csr-1* RNAi. Images show the left gonad arm in fixed, young adult worms. Expansion of expression is observed following either P-granule or *csr-1* RNAi. Red, FISH probe; blue, DAPI/DNA. Dashed line indicates domain of expression. The percentage of worms showing *ssq-1* and *rmd-3* expansion and the total number of worms examined are shown for each condition.

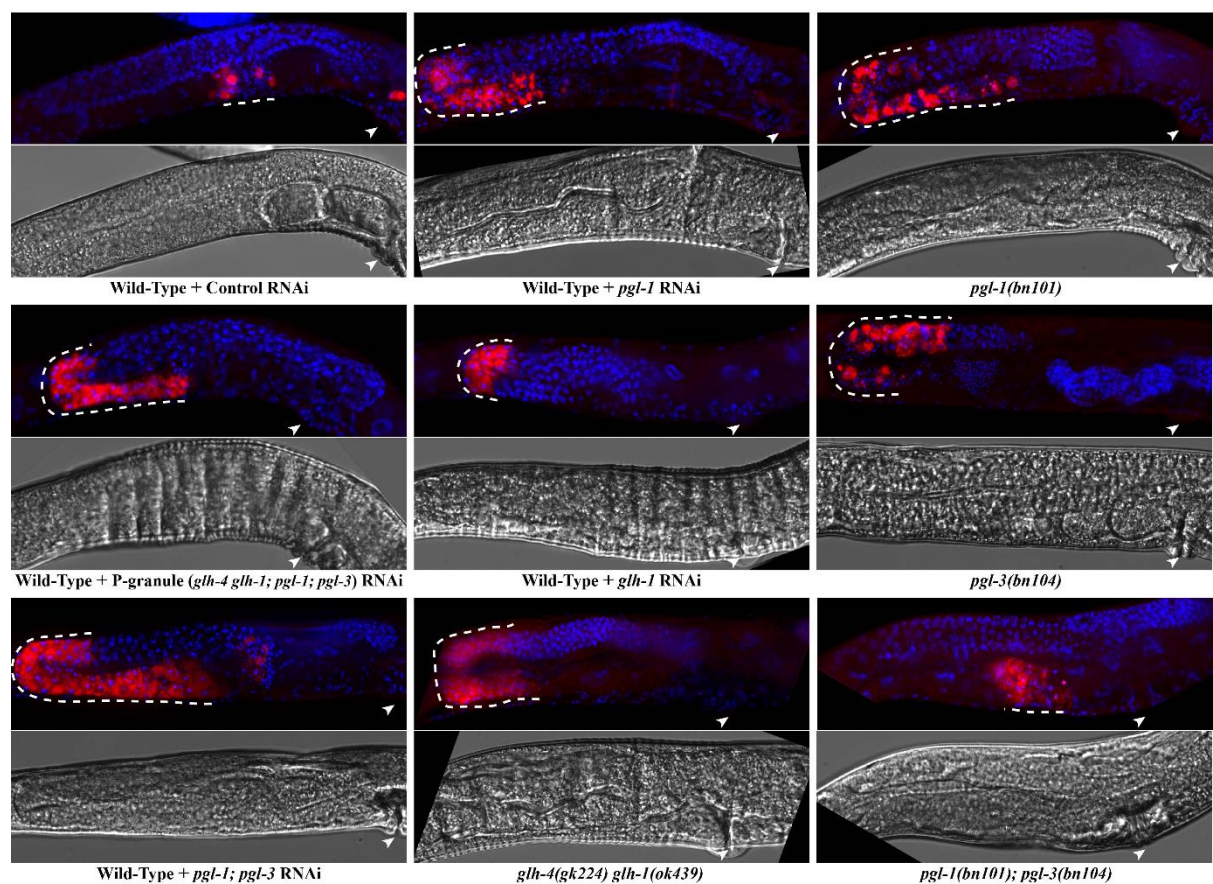


Figure S5. Distal expansion of *msp-3* transcripts when P granules are compromised. Images show the left gonad arm in fixed whole worms of day 1 adults. Ventral side down. Red, *msp-3* FISH probe; blue, DAPI/DNA. Dotted line indicates domain of *msp-3* expression. Arrowheads point to vulva.

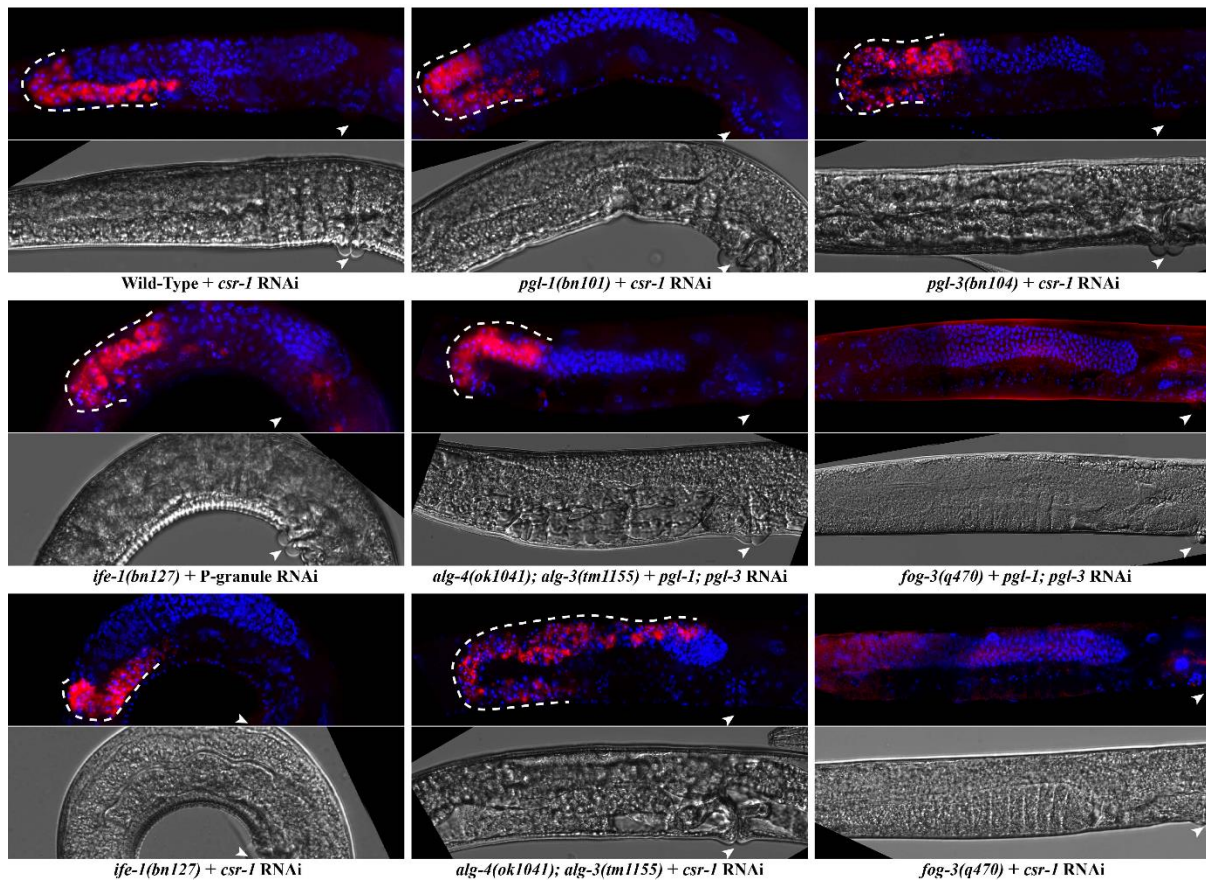


Figure S6. Distal expansion and suppression of *msp-3* expression. Images show the left gonad arm in fixed whole worms. Ventral side down. Red, *msp-3* FISH probe; blue, DAPI/DNA. Dotted line indicates domain of *msp-3* expression. Arrowheads point to vulva.

Table S1. Interactive table used to compare P-granule and *csr-1* RNAi mRNA-seq germline expression changes. Column legend:

- A) ID
- B) Chromosome
- C) Gene ID
- D) Control RNAi – mean sequence counts (average of four replicates, normalized using DESeq)
- E) P-granule RNAi – mean sequence counts (average of four replicates, normalized using DESeq)
- F) P-granule RNAi – Log2 fold change compared to control.
- G) p value – significance between the four control replicates to the four P-granule RNAi replicates
- H) adjusted p value - calculated using the Benjamini-Hochberg method for multiple testing correction (Anders and Huber, 2010).
- I) Set 1 genes – those genes differentially expressed following P-granule RNAi with a p value < 0.05
- J) Set 2 genes – those genes differentially expressed following P-granule RNAi with an adjusted p value < 0.05
- K) *csr-1* RNAi – mean sequence counts (average of four replicates, normalized using DESeq)
- L) *csr-1* RNAi – Log2 fold change compared to control.
- M) p value – significance between the four control replicates to the four *csr-1* RNAi replicates
- N) adjusted p value - calculated using the Benjamini-Hochberg method for multiple testing correction (Anders and Huber, 2010).
- O) Set 1 genes – those genes differentially expressed following *csr-1* RNAi with a p value < 0.05
- P) Set 2 genes – those genes differentially expressed following *csr-1* RNAi with an adjusted p value < 0.05
- Q) Germline enriched genes from (Reinke et al., 2004).
- R) Germline enriched gender neutral. Column Q genes with gametogenesis genes depleted from (Reinke et al., 2004).
- S) Soma enriched genes from (Reinke et al., 2004).
- T) Neuron enriched genes from (Watson et al., 2008).
- U) Spermatogenesis enriched genes from (Reinke et al., 2004).
- V) Spermatogenesis enriched genes from (Ortiz et al., 2014).
- W) List of genes encoding spermatogenesis proteins (Chu et al., 2006).

- X) All spermatogenesis genes (sum of columns U, V, W)
- Y) Oocyte enriched genes from (Reinke et al., 2004).
- Z) Oocyte enriched genes from (Ortiz et al., 2014).
- AA) Gene expression ratio in *csr-1* mutant tiling arrays from (Claycomb et al., 2009).
- AB) Genes significantly upregulated by GRO-seq in *csr-1* hypomorphic mutants from (Cecere et al., 2014).
- AC) Genes significantly down regulated by GRO-seq in *csr-1* hypomorphic mutants from (Cecere et al., 2014).
- AD) Genes targeted by CSR-1-bound 22Gs identified in (Claycomb et al., 2009).
- AE) Genes targeted by ALG-3/4-bound 26Gs identified in (Conine et al., 2013).
- AF) Genes targeted by CSR-1-bound 22Gs in males, identified in (Conine et al., 2013).

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