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eLife's transparent reporting form

We encourage authors to provide detailed information within their submission to facilitate the interpretation and replication of experiments. Authors can upload supporting documentation to indicate the use of appropriate reporting guidelines for health-related research (see EQUATOR Network), life science research (see the BioSharing Information Resource), or the ARRIVE guidelines for reporting work involving animal research. Where applicable, authors should refer to any relevant reporting standards documents in this form.

If you have any questions, please consult our Journal Policies and/or contact us: editorial@elifesciences.org.

Sample-size estimation

- You should state whether an appropriate sample size was computed when the study was being designed
- You should state the statistical method of sample size computation and any required assumptions
- If no explicit power analysis was used, you should describe how you decided what sample (replicate) size (number) to use

Please outline where this information can be found within the submission (e.g., sections or figure legends), or explain why this information doesn't apply to your submission:

We relied on the ability of the *Drosophila melanogaster* system to provide large numbers of animals for experimentation. Power analysis was not computed in advance. Replicates and sample sizes are reported in the legends of each figure.

Replicates

- You should report how often each experiment was performed
- You should include a definition of biological versus technical replication
- The data obtained should be provided and sufficient information should be provided to indicate the number of independent biological and/or technical replicates
- If you encountered any outliers, you should describe how these were handled
- Criteria for exclusion/inclusion of data should be clearly stated
- High-throughput sequence data should be uploaded before submission, with a private link for reviewers provided (these are available from both GEO and ArrayExpress)

Please outline where this information can be found within the submission (e.g., sections or figure legends), or explain why this information doesn't apply to your submission:

For all figures and conditions at least 2 biological replicates were performed. Information about biological replicates for each figure panel can be found in the respective figure legends. Only Figure 3N contained technical replicates and had one data point excluded – both as detailed in the methods section for qRT-PCR.



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Statistical reporting

- Statistical analysis methods should be described and justified
- Raw data should be presented in figures whenever informative to do so (typically when N per group is less than 10)
- For each experiment, you should identify the statistical tests used, exact values of N, definitions of center, methods of multiple test correction, and dispersion and precision measures (e.g., mean, median, SD, SEM, confidence intervals; and, for the major substantive results, a measure of effect size (e.g., Pearson's r, Cohen's d)
- Report exact p-values wherever possible alongside the summary statistics and 95% confidence intervals. These should be reported for all key questions and not only when the p-value is less than 0.05.

Please outline where this information can be found within the submission (e.g., sections or figure legends), or explain why this information doesn't apply to your submission:

For **Fig3M** the Bonferroni-Šídák multiple comparison test was used to analyze differences between mGFP100Dv1 and GFP0D levels within individual tissues, as this is the key biological question. For **Fig3N** Dunnett's multiple comparison of mRNA levels in each tissue to testis was performed to answer the key biological question of whether the testis has more reporter transcript than other tissues. For **Fig5E** an unpaired T-test was performed for comparison between only two groups. For **Fig5F** Tukey's HSD test for multiple comparisons was performed to analyze differences in fertility between sexes within the same genotype, and between the same sex across the two genotypes. For most graphs, raw data is displayed as individual points with mean +/- SEM indicated. Figure legend for each panel lists N, statistical analysis, and p-value (p>0.05,n.s.);(p<0.05,*);(p<0.01,**);(p<0.001,***);(p<0.001,***). A list of exact p-values for all tests is reported in the attached Excel file "reported_statistics".

(For large datasets, or papers with a very large number of statistical tests, you may upload a single table file with tests, Ns, etc., with reference to sections in the manuscript.)

Group allocation

- Indicate how samples were allocated into experimental groups (in the case of clinical studies, please specify allocation to treatment method); if randomization was used, please also state if restricted randomization was applied
- Indicate if masking was used during group allocation, data collection and/or data analysis

Please outline where this information can be found within the submission (e.g., sections or figure legends), or explain why this information doesn't apply to your submission:

Animals were allocated to groups by genotype or experimental condition as indicated in the figure legends. No masking was used.

Additional data files ("source data")

- We encourage you to upload relevant additional data files, such as numerical data that are represented as a graph in a figure, or as a summary table
- Where provided, these should be in the most useful format, and they can be uploaded as "Source data" files linked to a main figure or table
- Include model definition files including the full list of parameters used
- Include code used for data analysis (e.g., R, MatLab)



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Avoid stating that data files are "available upon request"

Please indicate the figures or tables for which source data files have been provided:

Source data has been provided for the following figure panels: Figure 1 panel R, Figure 3 panel M, Figure 5 panel E.