



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:

The number of samples used in the study was not pre-determined by power analysis but is in line with the typical sample size used in this model organism. The total sample size for each experiment, given as cells or animals (N), is provided in the text, Figures, Figure Legends and Supplementary File 2,3.

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:

In behavioural assays, technical replicates were acquired as each light stimulus type (with specific wavelength, irradiance, duration, pulse frequency) was presented a minimum of 3 times and a maximum of 9 times per animal. Biological replicates were obtained from individual animals sampled from multiple clutches (e.g. non-siblings) on different days. Numbers of biological replicates (i.e. number of animals or cells 'N') are reported in the text, Figures, Figure Legends and Supplementary File 2,3. No outlier exclusion was performed.

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:

Statistical tests, p-values, N-values, and additional information are reported in Supplementary File 2,3. All tests were two-tailed and were chosen after data were tested for normality and homoscedasticity. Summary data are presented as mean \pm SEM unless otherwise stated.

(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:

Allocation of animals to experimental groups was based on genotype and opsin expression. In behavioural assays, opsin-negative siblings were used as control and were subjected to the same stimulation protocol as opsin-expressing animals. In electrophysiological recordings, GFP-expressing opsin-negative primary motor neurons were used as control and were targeted in *Tg(mnx1:Gal4;UAS:EGFP)* or *Tg(parga-GFP)* larvae. Similar numbers of biological replicates (i.e. animals or cells) were used in each group.

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)
- Avoid stating that data files are "available upon request"

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



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Videos showing example trials for each behavioural assay are provided as Video 1–5.
Source data files are provided for graphs displayed in Figure 2–9.