

## Life Sciences Reporting Summary

Nature Research wishes to improve the reproducibility of the work that we publish. This form is intended for publication with all accepted life science papers and provides structure for consistency and transparency in reporting. Every life science submission will use this form; some list items might not apply to an individual manuscript, but all fields must be completed for clarity.

For further information on the points included in this form, see [Reporting Life Sciences Research](#). For further information on Nature Research policies, including our [data availability policy](#), see [Authors & Referees](#) and the [Editorial Policy Checklist](#).

### ▶ Experimental design

#### 1. Sample size

Describe how sample size was determined.

No sample size calculations were performed. Sample size was uniformly determined by analyzing 6-20 animals for each condition for each replicate, with experiments having a minimum of 2 independent replicates (now indicated in Methods). In some instances, like experiments with more inherent variability (behavioral assays) more animals were analyzed per replicate (20-40 animals; this is now indicated in Methods). These sample sizes are sufficient to detect significance by the two-sided Wilcoxon rank-sum tests used in this study.

#### 2. Data exclusions

Describe any data exclusions.

No data was excluded

#### 3. Replication

Describe whether the experimental findings were reliably reproduced.

All experiments were independently repeated at least twice with similar results (now indicated in Statistics and Reproducibility section of Methods); any case where replication failed is indicated in the corresponding figure legend.

#### 4. Randomization

Describe how samples/organisms/participants were allocated into experimental groups.

There was no method of randomization; experiments were performed based on condition and genotype, and animals were not further allocated into experimental sub-groups necessitating randomization.

#### 5. Blinding

Describe whether the investigators were blinded to group allocation during data collection and/or analysis.

Investigators were blinded to genotype (control vs mutant or rescue) or experimental conditions (control vs experiment) for all experiments.

Note: all studies involving animals and/or human research participants must disclose whether blinding and randomization were used.

## 6. Statistical parameters

For all figures and tables that use statistical methods, confirm that the following items are present in relevant figure legends (or in the Methods section if additional space is needed).

n/a Confirmed

- The exact sample size ( $n$ ) for each experimental group/condition, given as a discrete number and unit of measurement (animals, litters, cultures, etc.)
- A description of how samples were collected, noting whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
- A statement indicating how many times each experiment was replicated
- The statistical test(s) used and whether they are one- or two-sided (note: only common tests should be described solely by name; more complex techniques should be described in the Methods section)
- A description of any assumptions or corrections, such as an adjustment for multiple comparisons
- The test results (e.g.  $P$  values) given as exact values whenever possible and with confidence intervals noted
- A clear description of statistics including central tendency (e.g. median, mean) and variation (e.g. standard deviation, interquartile range)
- Clearly defined error bars

See the web collection on [statistics for biologists](#) for further resources and guidance.

## ► Software

Policy information about [availability of computer code](#)

### 7. Software

Describe the software used to analyze the data in this study.

R and R studio statistical software for statistical tests. Zeiss Zen Black software for image acquisition and processing.

For manuscripts utilizing custom algorithms or software that are central to the paper but not yet described in the published literature, software must be made available to editors and reviewers upon request. We strongly encourage code deposition in a community repository (e.g. GitHub). [Nature Methods guidance for providing algorithms and software for publication](#) provides further information on this topic.

## ► Materials and reagents

Policy information about [availability of materials](#)

### 8. Materials availability

Indicate whether there are restrictions on availability of unique materials or if these materials are only available for distribution by a for-profit company.

No restrictions

### 9. Antibodies

Describe the antibodies used and how they were validated for use in the system under study (i.e. assay and species).

No antibodies were used in this study

### 10. Eukaryotic cell lines

a. State the source of each eukaryotic cell line used.

No eukaryotic cell lines

b. Describe the method of cell line authentication used.

No eukaryotic cell lines

c. Report whether the cell lines were tested for mycoplasma contamination.

No eukaryotic cell lines

d. If any of the cell lines used are listed in the database of commonly misidentified cell lines maintained by [ICLAC](#), provide a scientific rationale for their use.

No eukaryotic cell lines

## ► Animals and human research participants

Policy information about [studies involving animals](#); when reporting animal research, follow the [ARRIVE guidelines](#)

### 11. Description of research animals

Provide details on animals and/or animal-derived materials used in the study.

no vertebrate or higher invertebrate animals

## 12. Description of human research participants

Describe the covariate-relevant population characteristics of the human research participants.

No human research subjects