

S1 Fig. pH3 expression in the optic placode.

We analyzed the number of mitotically active cells in the optic placode by staining against pH3 (green). We also used antibodies against Eya (red) to identify the optic placode (yellow outline), and we counterstained with DAPI (grey). At stage 10 virtually all Eya-positive cells in the placode co-express pH3 (A), but very few express it later, during stages 11 (B) and 12 (C). Scale bars represent 20  $\mu\text{m}$ .

S2 Fig. The PR marker Hazy is expressed in all PR precursors in *tll49l*, *ptc9*, and *N55e11* mutants.

We stained against Kruppel (green) to label the larval eye in embryos around stage 14, and co-stained against Hazy (magenta). Hazy is a transcription factor that regulates the development of all types of *Drosophila* PRs in wildtype conditions [17, 18, 65]. Similar to wildtype, all PR precursors express Hazy in *tll* (A), *ptc* (B) and *Notch* (C) mutant embryos. Scale bars represent 20  $\mu\text{m}$ .

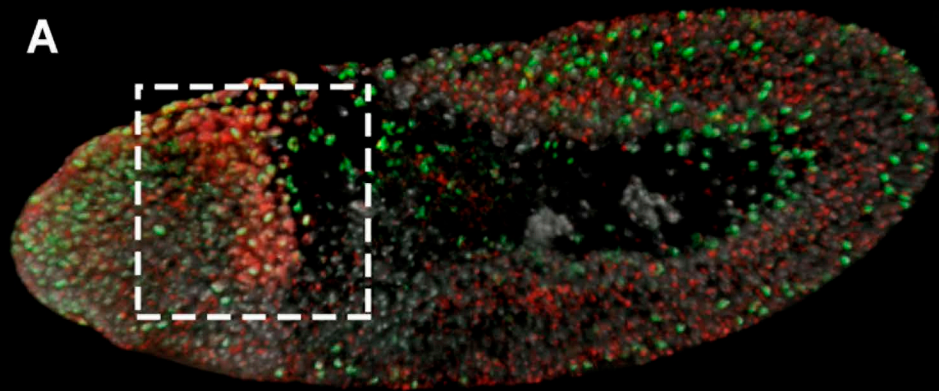
S3 Fig. Quantification of optic placode cell numbers.

The optic placode contains the same number of cells in *N55e11* mutants and *so>tll* embryos compared to wildtype embryos (counted at stage 11). The number of cells in the optic placode is increased in *tll49l* mutants and *ptc9* mutants compared to wildtype embryos (counted at stage 11). Number of all optic placode cells: Anova:  $p < 0.001$   $F(4,43) = 15.05$ ; wildtype vs *tll49l*  $p < 0.001$ ,  $t = -5.627$ ; wildtype vs *so>tll*  $p =$

pH3 Eya DAPI

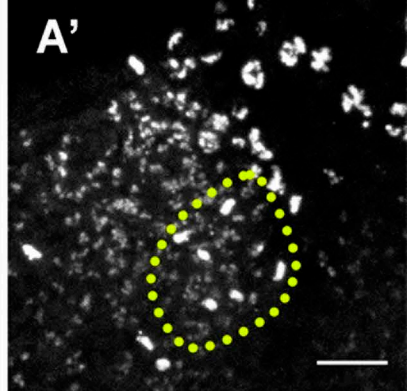
Stage 10

A



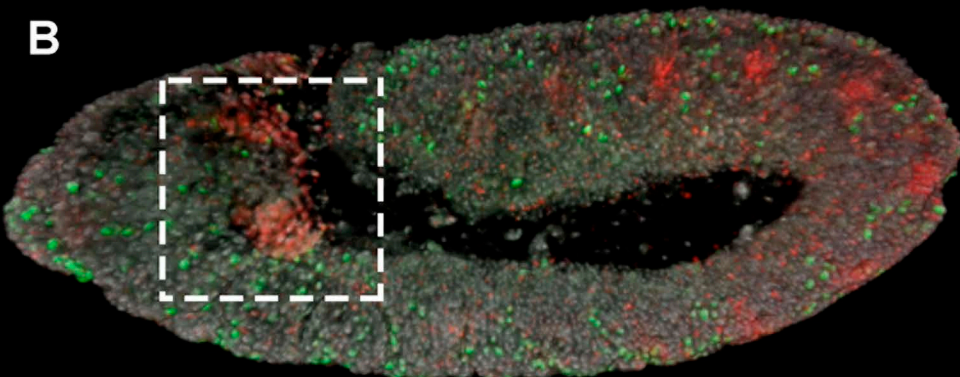
pH3

A'

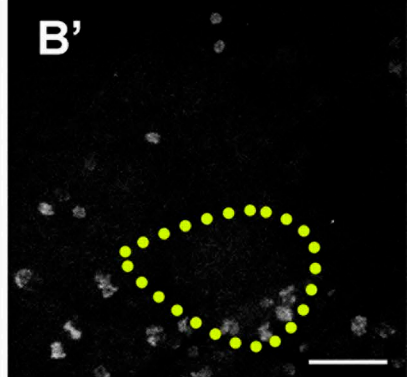


Stage 11

B

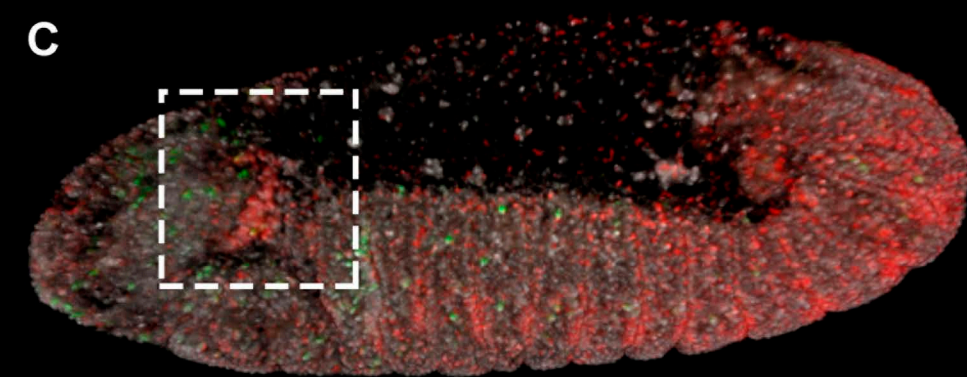


B'

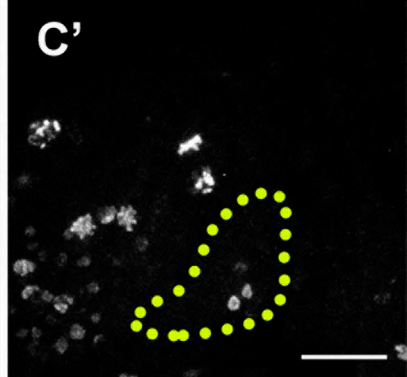


Stage 12

C



C'



*tll<sup>49l</sup>*

*ptc<sup>9</sup>*

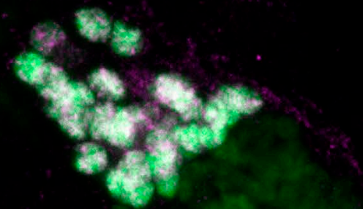
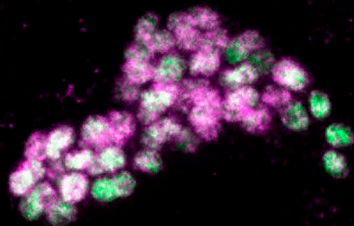
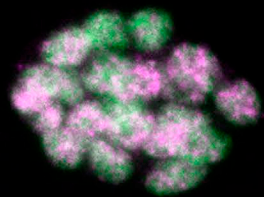
*N<sup>55e11</sup>*

A

B

C

Kr  
Hazy

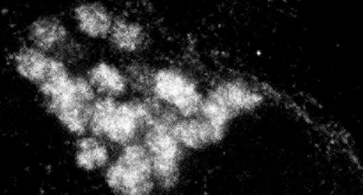
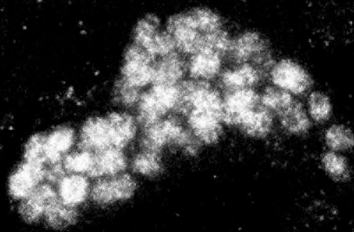
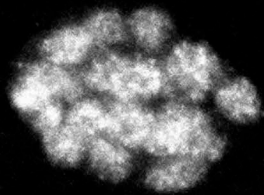


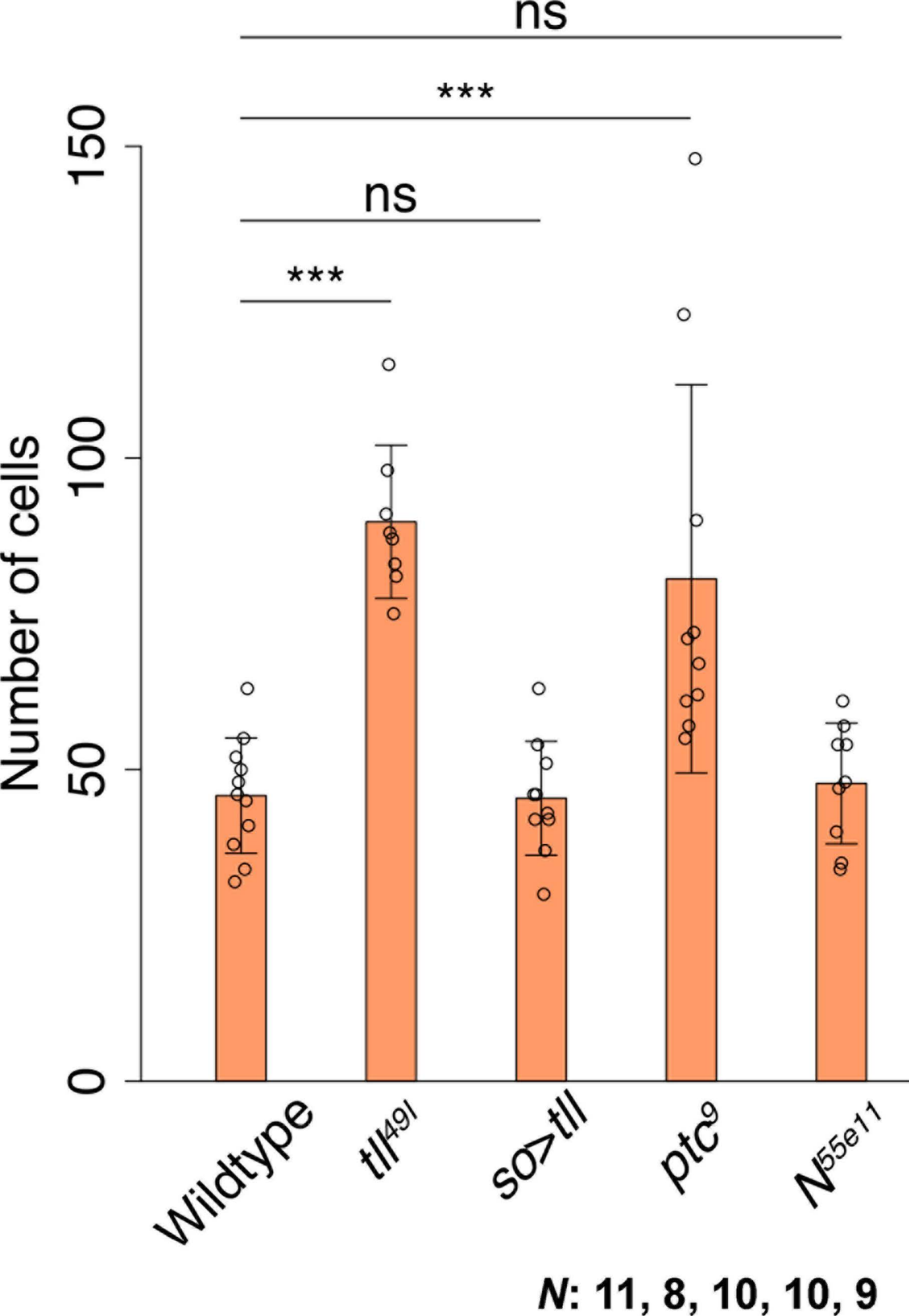
A'

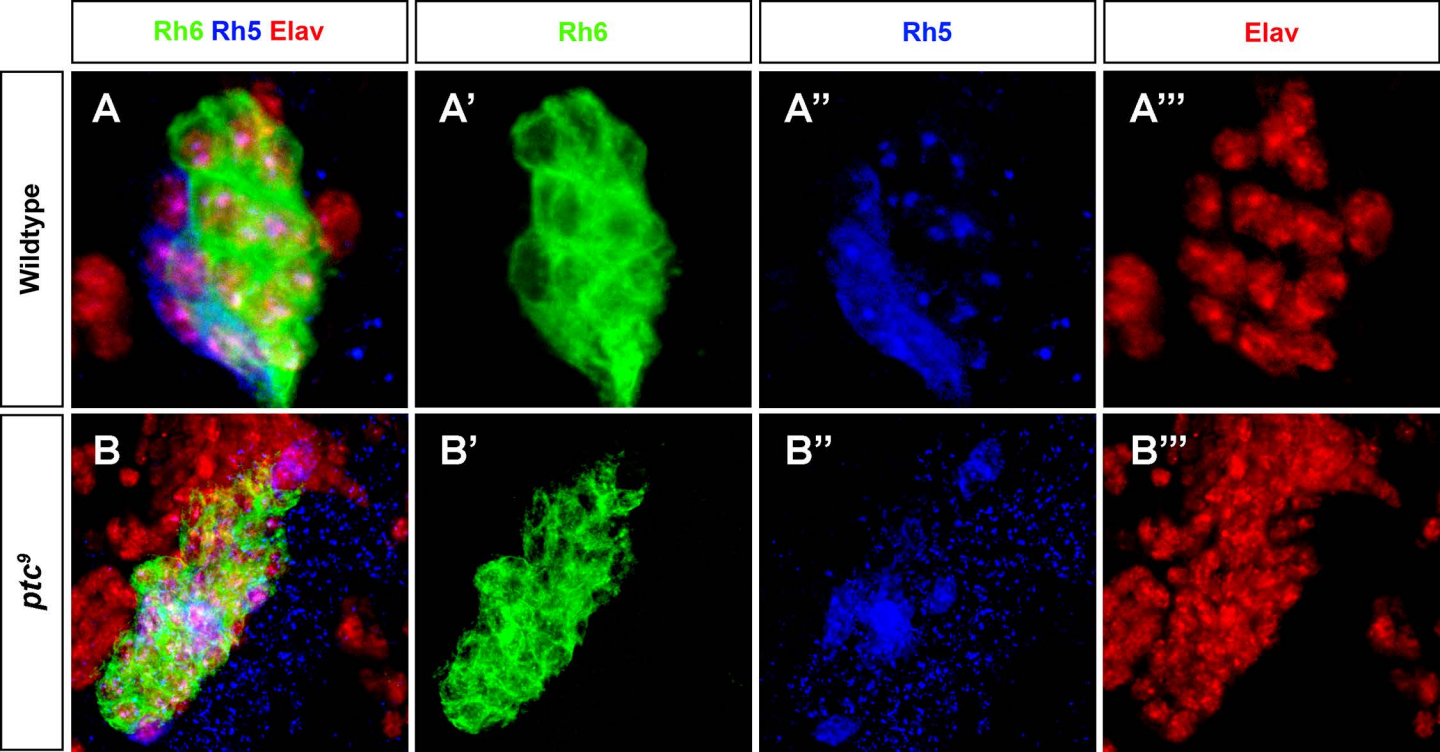
B'

C'

Hazy



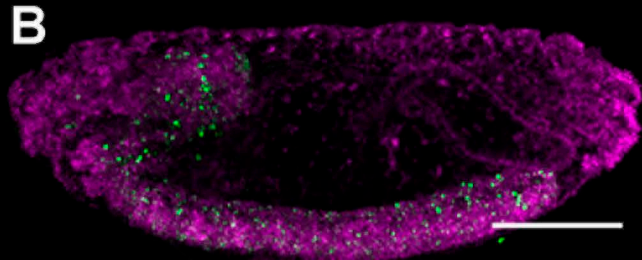
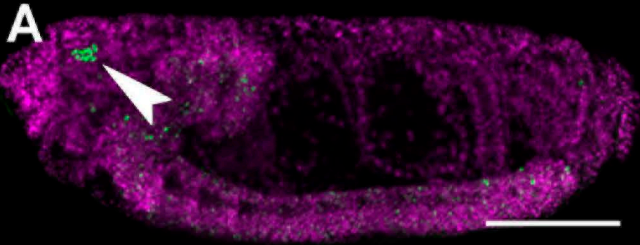




**Control**

***so>kuzDN, TII***

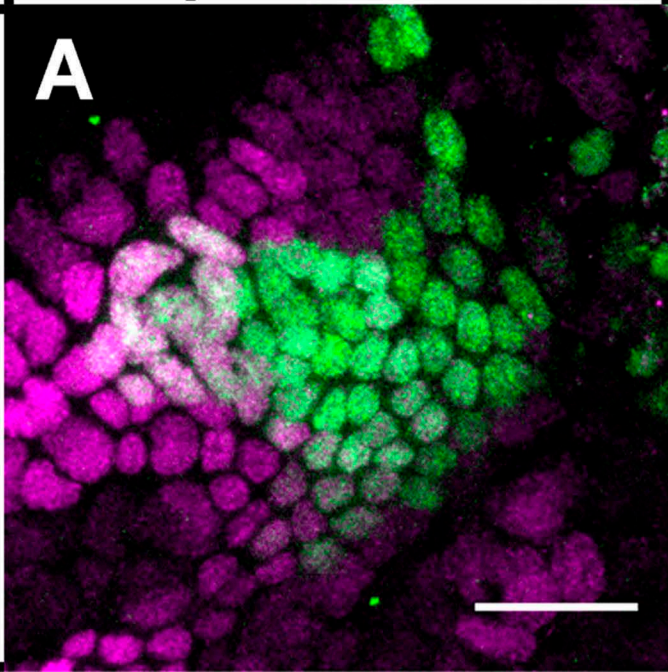
**Eya** DAPI



*ptc-lacZ*

A

Eya  $\beta$ Gal



$N^{55e11}; ptc-lacZ$

B

