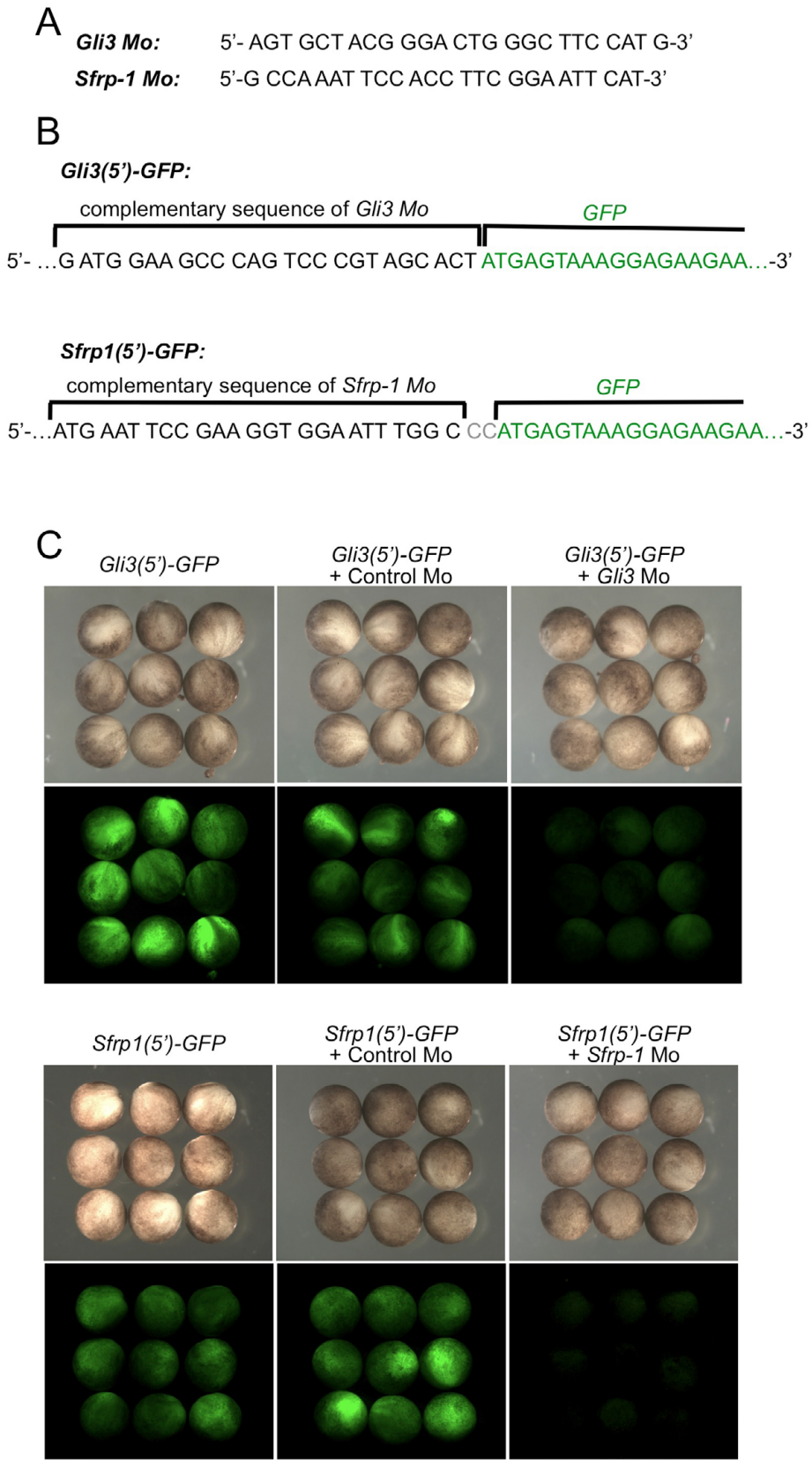

Fig. S1. In vivo validation of *Gli3* and *Sfrp-1* morpholino efficiency. (**A**) *Gli3* and *Sfrp-1* morpholinos (Mo) have been designed to target the ATG region of the corresponding transcripts. (**B**) Schematic representation of the chimeric constructs containing *GFP* fused downstream of *Gli3* or *Sfrp-1* Mo-complementary sequence [*Gli3(5')-GFP* or *Sfrp-1(5')-GFP*]. (**C**) In vivo GFP fluorescence was analysed at stage 19 following co-injection of the indicated Mo and GFP mRNA constructs. *Gli3* and *Sfrp-1* Mo inhibited GFP translation from *Gli3(5')-GFP* and *Sfrp-1(5')-GFP* constructs, respectively, whereas control Mo did not.

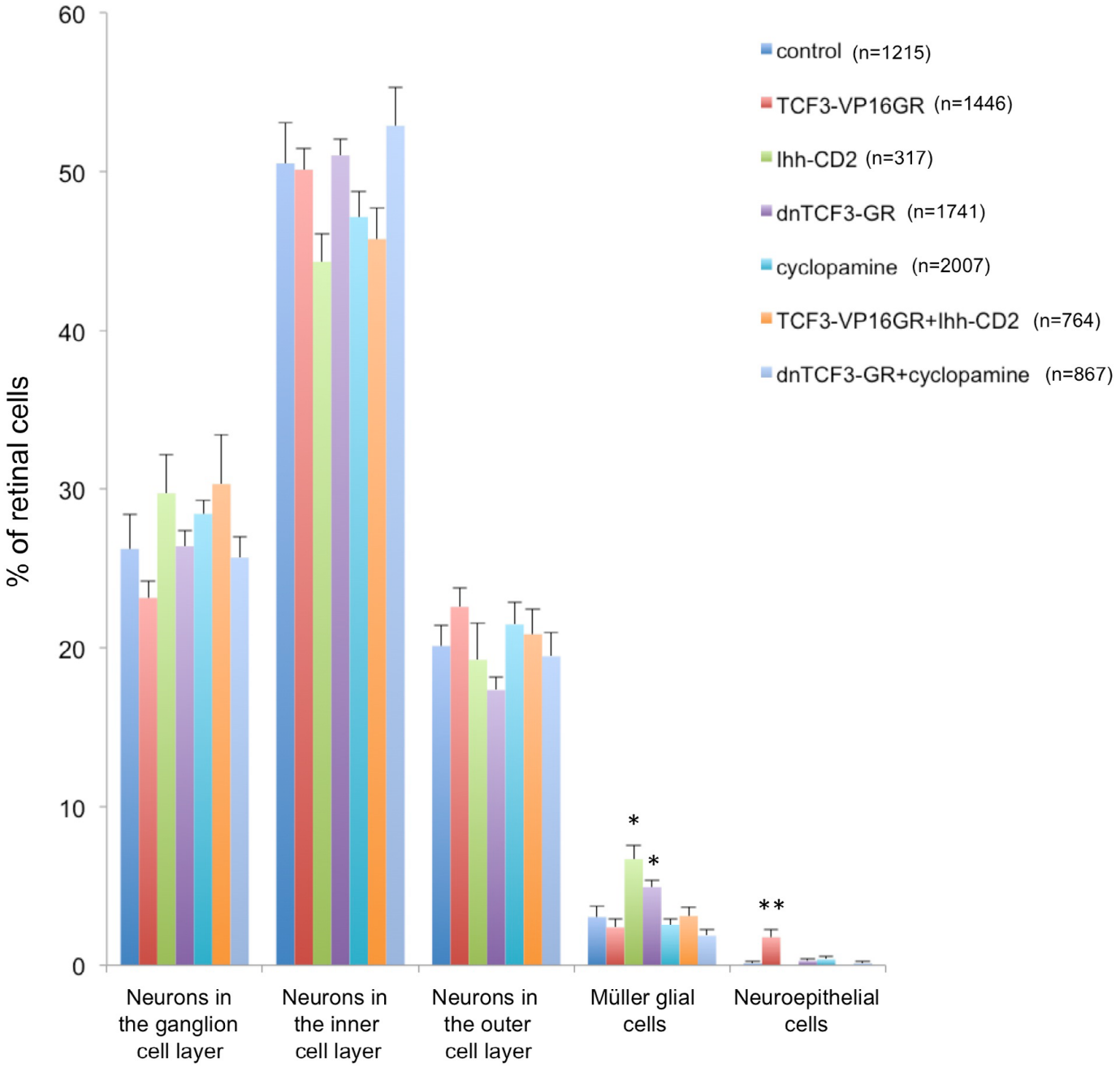

Fig. S2. Clonal analysis of cell type distribution in the retina following interference with Wnt and Hedgehog signalling pathways. Percentage of retinal cells observed in stage 41 retinas following in vivo lipofection with the indicated constructs. Cyclopamine treatment was performed from stage 18 onwards on embryos lipofected with *GFP*. Hedgehog or Wnt signalling perturbation significantly affects the percentage of Müller or neuroepithelial cells (see also Fig. 1). However, as these cells represent very minor cell subpopulations, their variations do not significantly impact on the percentage of neurons (which represent around 95% of total cells). Total number of counted cells is indicated for each condition. *P<0.05, **P<0.01 (Student's t-test).

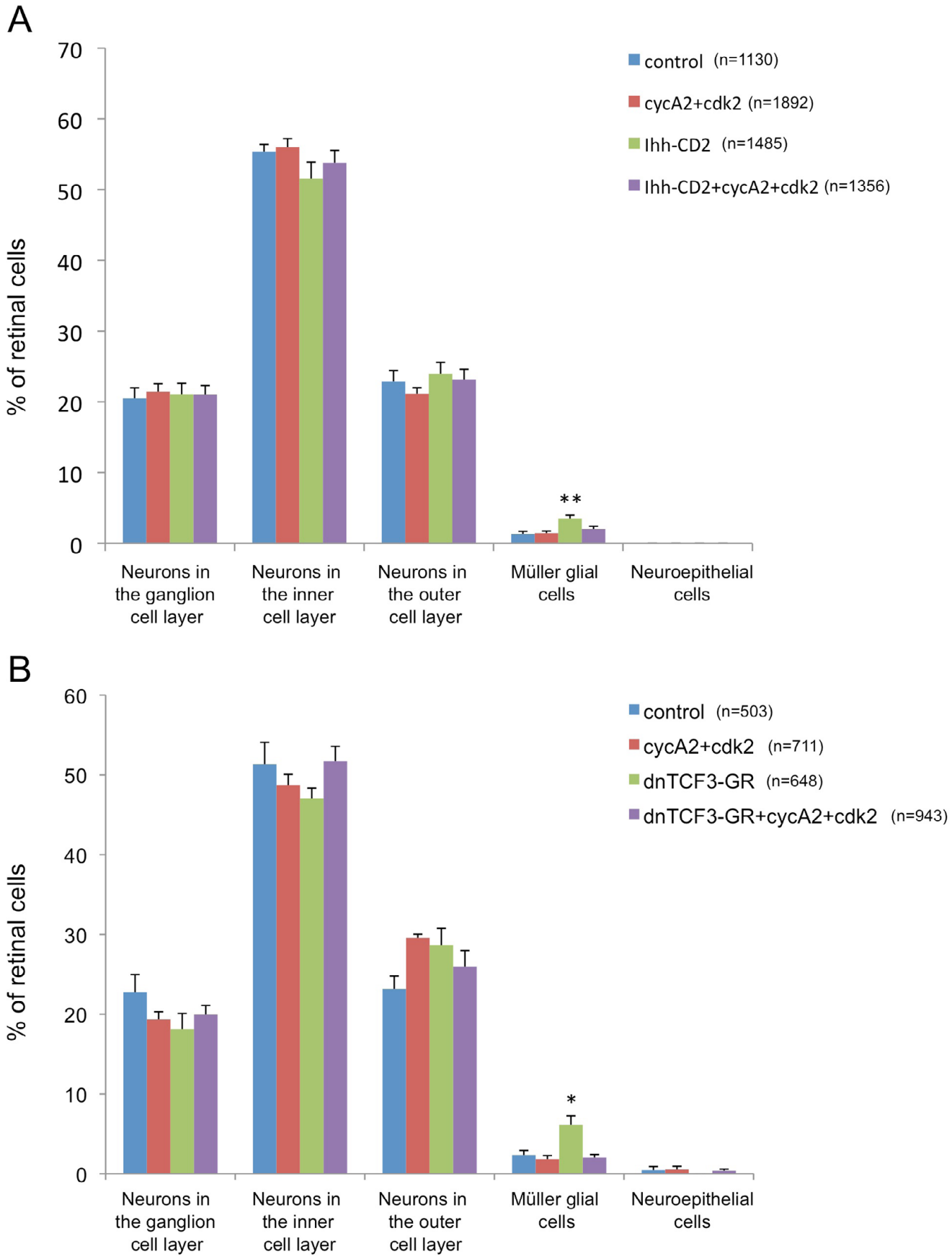

Fig. S3. Co-lipofection of *cyclinA2*/*cdk2* with either *Ihh-CD2* or *dnTCF3-GR* leads to a cell distribution indistinguishable from that observed in a control situation. (**A**,**B**) Percentage of retinal cells observed in stage 41 retinas following in vivo lipofection with the indicated constructs. Total number of counted cells is indicated for each condition. **P*<0.05, ***P*<0.01 (Student's *t*-test).

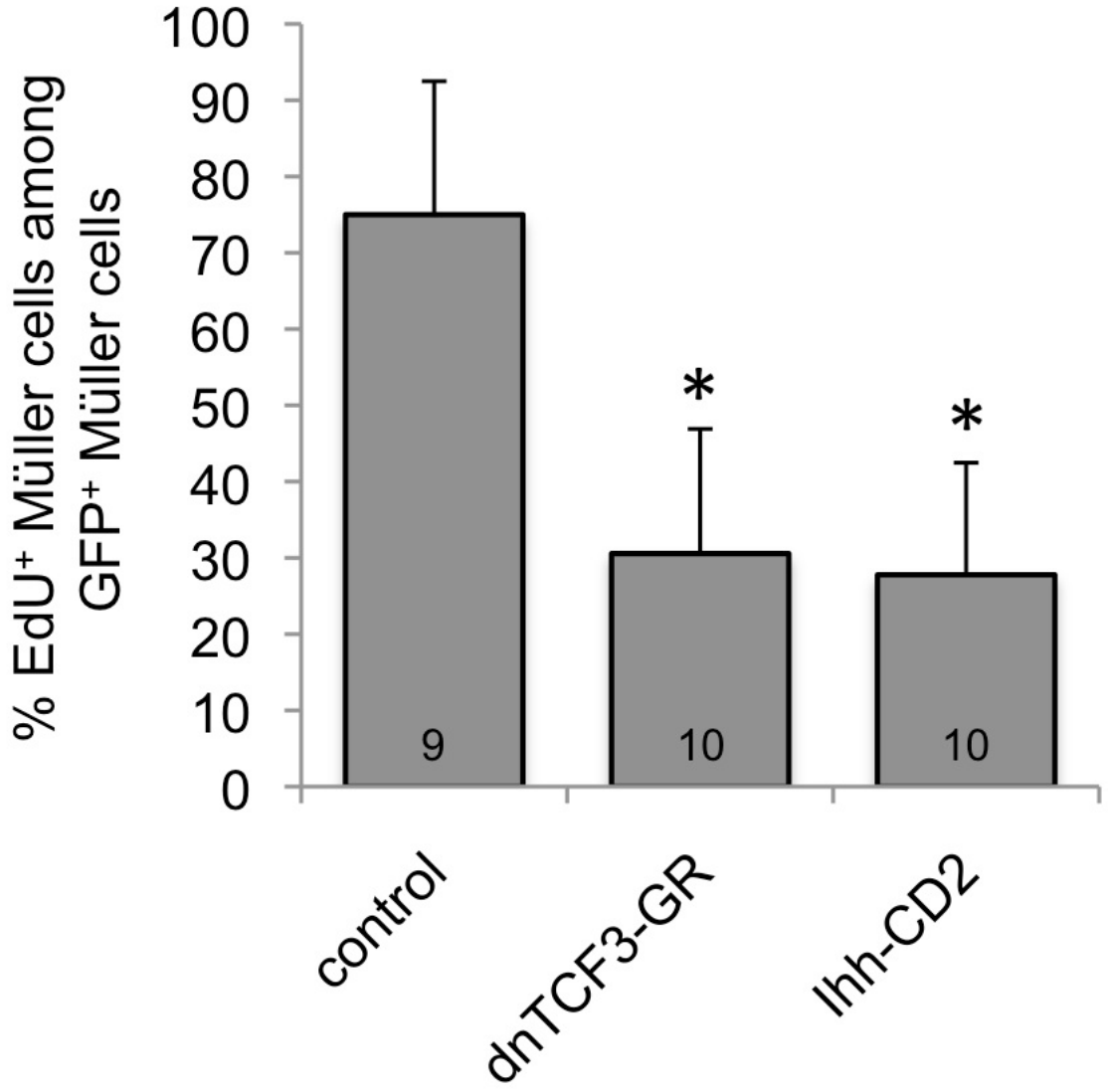

Fig. S4. Hedgehog and Wnt pathways have opposite impacts on cell cycle exit of Müller cells. Birthdating experiments (from stage 32 to stage 41) following in vivo lipofection with the indicated constructs. Transfected Müller cells that have exited the cell cycle before EdU exposure (stage 32) are EdU^- at stage 41, whereas Müller cells that have exited the cell cycle at any time during the EdU incorporation period are EdU^+. Graph represents the percentage of EdU^+ Müller cells among transfected Müller cells. Total number of analysed Müller cells per condition is indicated in each bar. *$P<0.05$ (Student's *t*-test).

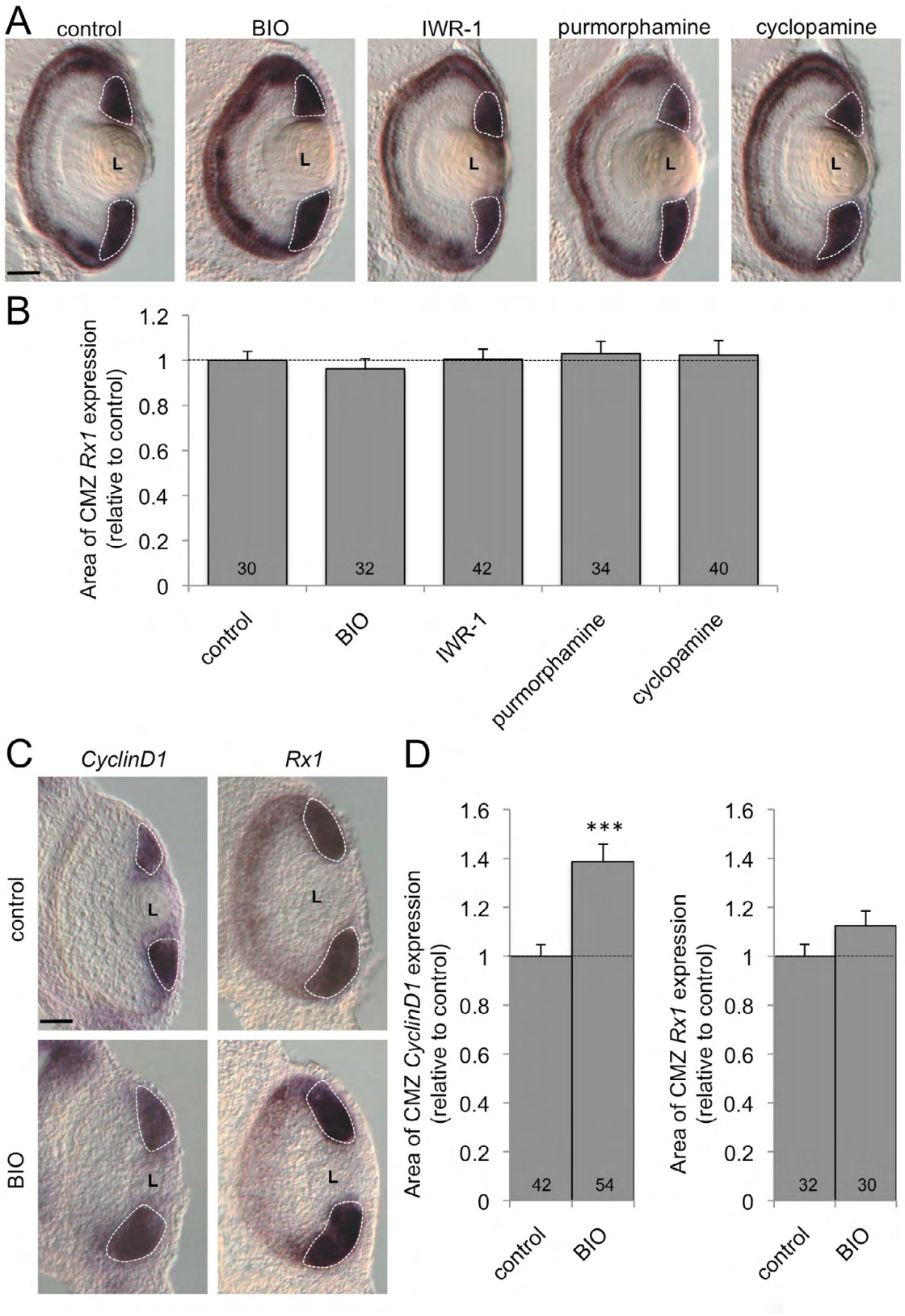

Fig. S5. The overall size of the CMZ is not affected upon pharmacological perturbation of the Wnt or Hedgehog signalling pathway. (**A**,**B**) In situ hybridisation analyses (A) of *Rx1* expression on stage 41 retinal sections 24 hours following BIO, IWR-1, purmorphamine or cyclopamine treatment as indicated. (B) Quantification of *Rx1* staining area per CMZ in each condition. (**C**,**D**) In situ hybridisation analyses (C) of *CyclinD1* or *Rx1* expression on stage 38 retinal sections 24 hours following treatment with BIO. (D) Quantifications of staining area for each transcript. Total number of analysed sections per condition is indicated in each bar. ***P<0.001 (Student's t-test). Labelling in the CMZ is delineated with dotted lines. L, lens. Scale bar: 40 µm.

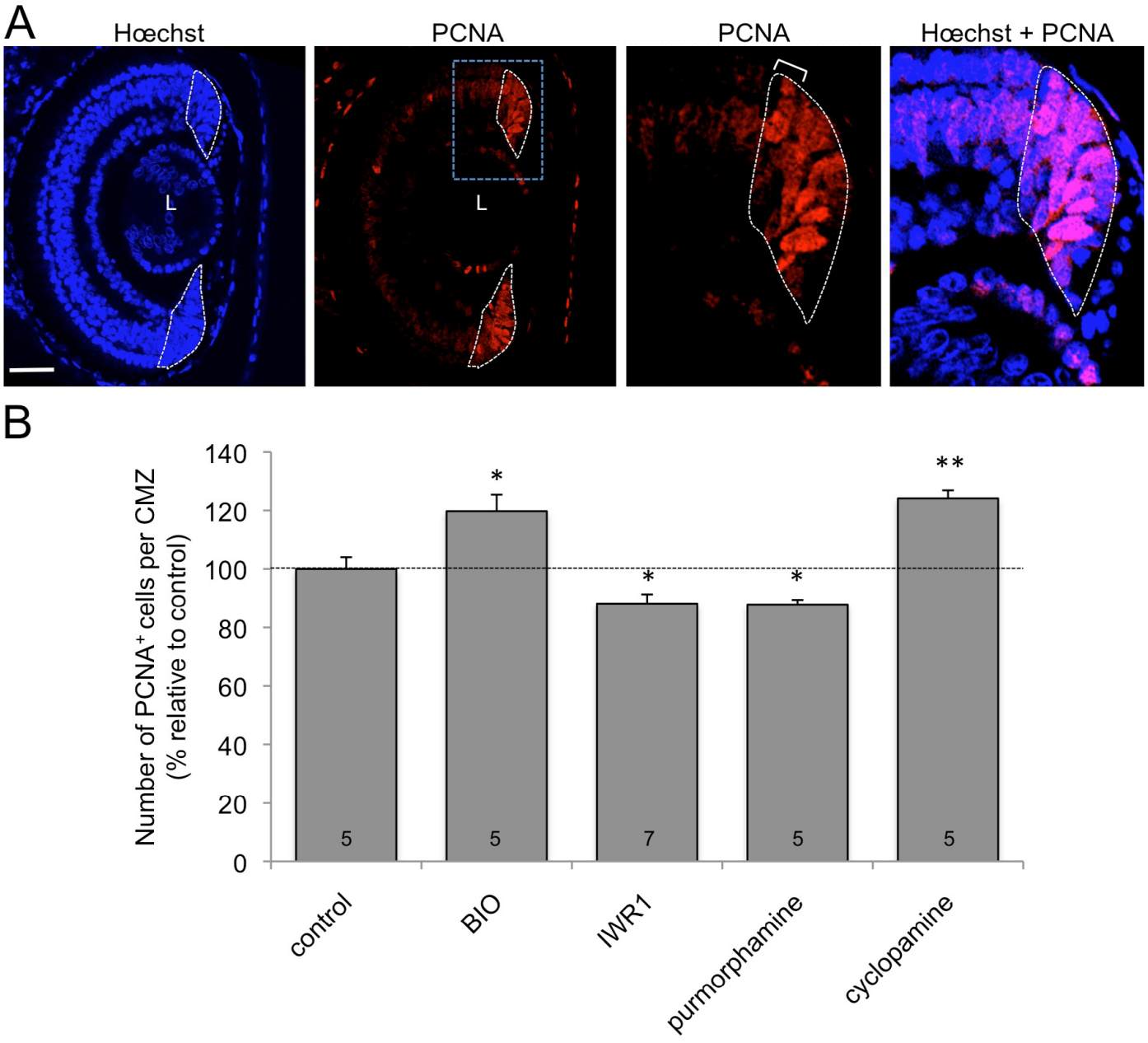

Fig. S6. PCNA immunostaining in the post-embryonic retina following pharmacological perturbation of the Wnt or Hedgehog signalling pathway. PCNA labelling analysis on stage 41 retinal sections 24 hours following treatment with the indicated drugs. (**A**) Typical control retinal section immunostained with anti-PCNA. Pictures on the right show high magnifications of the CMZ (delineated with dotted lines). Note that PCNA labels the whole CMZ, i.e. proliferative cells as expected, but also newly born postmitotic cells in the most central part of the CMZ (bracket). This is likely to be due to the long half-life of the antigen. (**B**) Quantification of PCNA$^+$ cells in the CMZ. Total number of analysed retinas per condition is indicated in each bar. *P<0.05, **P<0.01 (Student's t-test). L, lens. Scale bar: 40 µm.

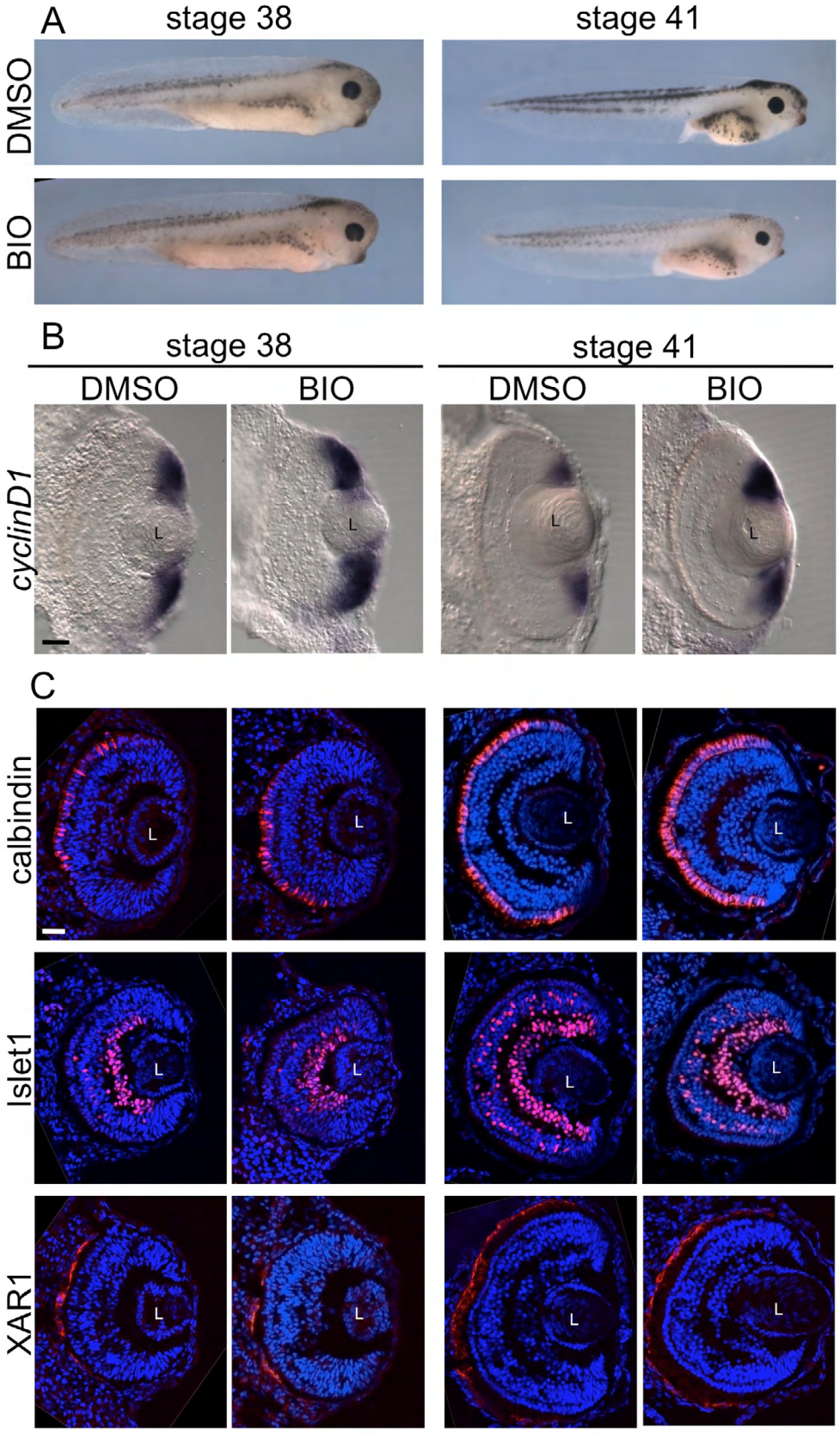

Fig. S7. Twenty-four hours of Wnt pathway activation does not delay development nor does it affect the central retina. (**A**) Tadpoles treated with BIO for 24 hours do not display any developmental delay, as judged by their overall morphology. (**B**) In situ hybridisation analysis of *CyclinD1* expression on retinal sections following a 24-hour BIO treatment, illustrating the effective activation of the Wnt pathway. (**C**) Immunofluorescence analysis of cell type-specific markers on sibling tadpoles. Calbindin is a marker of photoreceptor cells, Islet1 labels ganglion cells and a subtype of bipolar cells and XAR1 stains the RPE. L, lens. Scale bar: 40 µm.

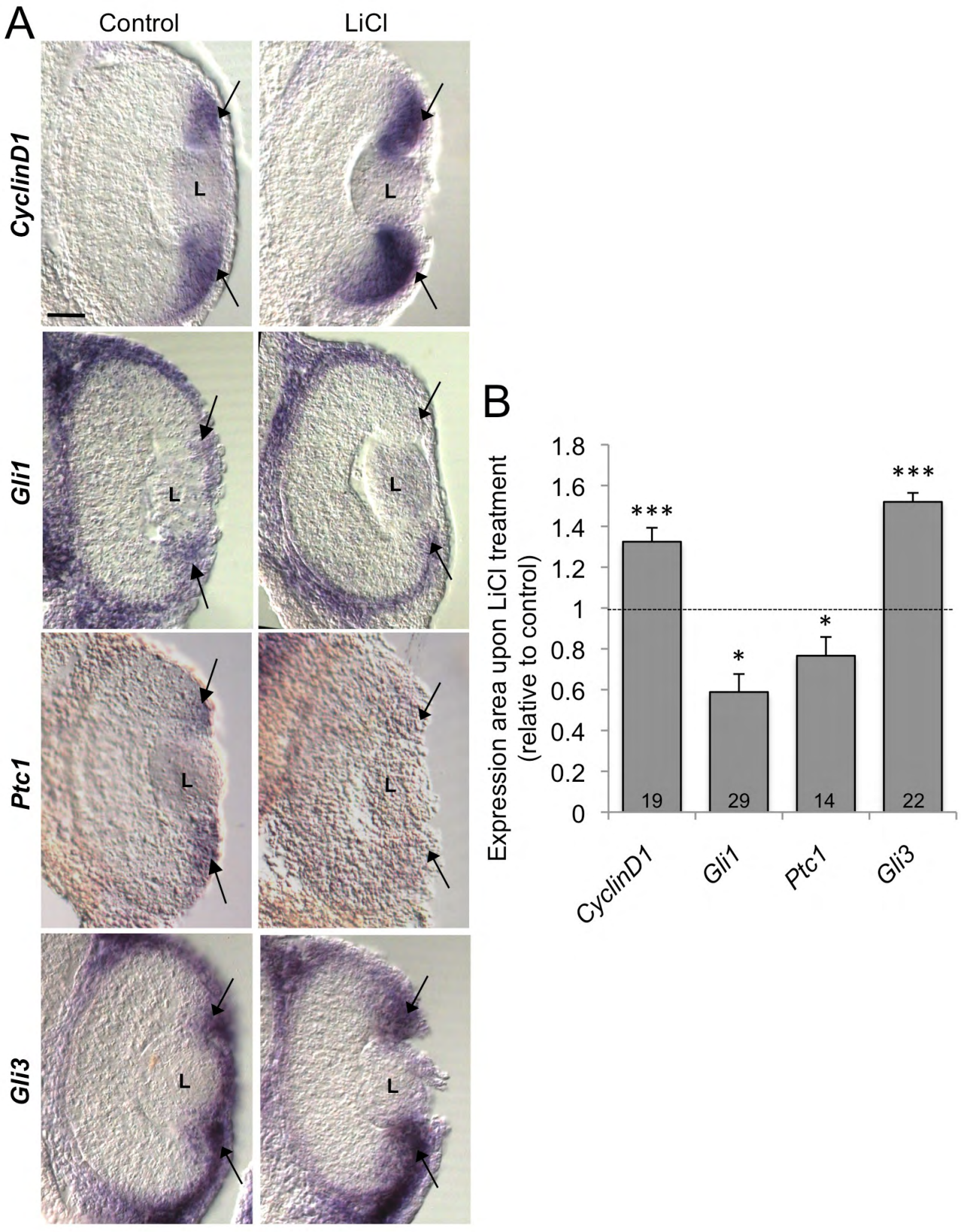

Fig. S8. Wnt pathway activation downregulates the Hedgehog target genes *Gli1* and *Ptc1* but increases *Gli3* expression. (**A**,**B**) In situ hybridisation analyses (A) of *CyclinD1*, *Gli1*, *Ptc1* or *Gli3* expression on stage 38 retinal sections 24 hours following LiCl treatment. Arrows indicate CMZ labelling. (B) Quantification of staining area for each gene. Compared with controls, LiCl-treated embryos exhibit decreased *Gli1* and *Ptc1* staining, whereas *Gli3* expression is enhanced. Total number of analysed sections per condition is indicated in each bar. **P*<0.05, ****P*<0.001 (Student's *t*-test). L, lens. Scale bar: 40 µm.

Table S1. PCR primer sequences

***Xenopus laevis* qPCR primers**		
Gene	**Forward primer**	**Reverse primer**
Gli3	GGCCCCCACCAACACCACTG	CCGTGGGACATTGACCGAAGGA
Patched1	CAGCTGCCCAGCCGAGGGTA	GGGCGAAATTGGCATCGCAGTA
ODC	GCTTCTGGAGCGGGCAAAGGA	CCAAGCTCAGCCCCCATGTCA
RPL8	CCACGTGTCCGTGGTGTGGCTA	GCGCAGACGACCAGTACGACGA
Sfrp1	CAGTGAGACAATGGCGGAGGTGAA	GGGGCGAAGAGAGAGCACAGGA
***Xenopus tropicalis* qPCR primers**		
Gene	**Forward primer**	**Reverse primer**
Patched1	CAGCTGCCCAGCCGAGGGTA	TCCTGGTCAGGCGGCGCTACTA
ODC	CATGGCATTCTCCCTGAAGTACCAGAA	GGACAGATGGTAGGGGCAAGCTCA
RPL8	CGCCACCGTTATCTCCCACAATC	CCACCAGCAACAACCCCAACA