# SUPPLEMENTARY MATERIAL 

corresponding to:

# Mutation of frizzled8a delays neural retinal cell differentiation and results in microphthalmia in zebrafish 

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Supp. Fig. S1. Validation of the fz8a mutant genotype. RT-PCR and sequence analyses of fz8a mutant transcripts. (A) Genotyping of wild-type embryos, heterozygous and homozygous fz8a mutants, using indicated primer pairs. (B) RT-PCR analysis of fz8a mutant transcripts in wild-type, normal siblings and microphthalmic MZfz8a embryos. (C) Sequence analysis of the PCR products from wild-type and microphthalmic embryos. The deleted region is underlined in the wild-type sequence and the TALENs targeting sequences are shadowed.


Supp. Fig. S2. Phenotype analysis of MZfz8a mutants from three independent fish pairs. Numbers on the top of each stacked column indicate total embryos analyzed at 72 hpf.


Supp. Fig. S3. Specification of the brain region and the eye field is not affected in MZfz8a mutants. In situ hybridization of indicated marker genes at bud stage. ( $\mathbf{A}, \mathbf{A}^{\prime}$ ) otx2, anterior neural plate. (B, $\mathbf{B}^{\prime}$ ) six3b, forebrain. ( $\mathbf{C}, \mathbf{C}^{\prime}$ ) rx3, eye field. ( $\mathbf{D}, \mathbf{D}^{\prime}$ ) pax2a, midbrain/hindbrain boundary. ( $\mathbf{E}, \mathbf{E}^{\prime}$ ) pax6a, eye field. All embryos are anterior view with dorsal region on the top. Scale bar, $250 \mu \mathrm{~m}$.


Supp. Fig. S4. Abnormal lamination of retinal layers in microphthalmic embryos. (A-B") Wild-type embryos and normal siblings show similar organization of retinal layers. Microphthalmic embryos display abnormal organization of retinal layers. (C) Statistics of the relative width of different retinal layers between wild-type and microphthalmic embryos. Bars represent the mean $\pm$ s.d. from three independent experiments with a total of 20 sections at similar position for each condition ( ${ }^{*}, P<0.05 ;{ }^{* * *}, P<0.001$ ). Scale bar, $50 \mu \mathrm{~m}$.


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