

Supplemental Figure 1: Chx10-Cre is selectively active in the retina and eliminates Lhx2 expression in Chx10-Cre; Lhx $2^{\text {lox/lox }}$ retinas. (A - D) Immunostaining for dsRed (red) to detect tdTomato expression in E10.5-E13.5 Chx10-Cre;Ai9 eye sections. (E - L) Immunostaining of Chx10-Cre;Lhx2 ${ }^{\text {lox/+ }}(\mathrm{E}-\mathrm{H})$ and Chx10-Cre;Lhx2 ${ }^{\text {lox/lox }}$ eye sections ( $\mathrm{I}-\mathrm{L}$ ) for Lhx2 (red) and Pax6 (green). Nuclei are counter-stained with DAPI (blue). (Scale Bars: $100 \mu \mathrm{~m}$ )

A



C

|  | No. of eyes <br> with lens | No. of eyes <br> without lens | Total |
| :---: | :---: | :---: | :---: |
| Chx10-Cre;Lh $\times 2^{\text {Iox/lox }}$ | 5 | 6 | 11 |
| Chx10-Cre; Lh $\mathbf{2}^{\text {lox/lox }}$; <br> pMes-Fgf10 | 15 | 0 | 15 |
| Total | 20 | 6 | 26 |
| Fisher's Exact Test: P $=0.002$ |  |  |  |



Supplemental Figure 2: Overexpression of Fgf10 led to significant increase in lens, but not retinal, size. (A) Real-time quantitative PCR analysis shows induction of Fgfl 10 mRNA expressions in pMes-Fgfl0 retinas. Data represent mean normalized to Gapdh values $\pm$ SEM. (Unpaired two-tailed t-test; $\mathrm{n}=3 ; * \mathrm{P}<0.05 ; * * \mathrm{P}<0.01$ ) (B) Graph indicating the average lens size of Chx10-Cre;Lhx2 lox/+ , Chx10-Cre;Lhx2 $2^{\text {lox/ }+} ;$ pMes-Fgf10, Chx10-Cre;Lhx $2^{\text {lox/lox }}$ and Chx10Cre;Lhx2 ${ }^{\text {lox/lox }}$;pMes-Fgf10 animals at E11.5, E12.5, E13.5 and P0.5. (C) Contingency table depicting the number of eyes with or without detectable lenses at P0.5 for Chx10-Cre;Lhx2 ${ }^{\text {lox/lox }}$ and Chx10-Cre;Lhx2 ${ }^{\text {lox/lox }}$;pMes-Fgfl0 animals. Eye sections immunostained for Prox 1 and $\beta$ Crystallin were used in this analysis. (D) Graph indicating average retinal area of Chx10Cre;Lhx $2^{\text {lox/+ }}$, Chx10-Cre;Lhx2 $2^{\text {lox/ }}$;pMes-Fgfl0, Chx10-Cre;Lhx2 ${ }^{\text {lox/lox }}$ and Chx10Cre;Lhx2 ${ }^{\text {lox/lox }} ;$ pMes-Fgfl0 animals at E11.5, E12.5, E13.5 and P0.5. (One-way ANOVA followed by post hoc Tukey's test; $\mathrm{n} \geq 4$ for E11.5; $\mathrm{n} \geq 9$ for E12.5; $\mathrm{n} \geq 6$ for E13.5; $\mathrm{n} \geq 7$ for P0.5; *P<0.05; **P $<0.01 ; * * * * \mathrm{P}<0.0001$; Error bars indicate SEM)


Supplemental Figure 3: Cre-mediated induction of Fgf10 expression in Chx10Cre;Lhx2 ${ }^{\text {lox/lox }} ;$ pMes-Fgf10 retinas at later developmental stages. In situ hybridization analysis of Fgf10 mRNA expression levels at E17 (A - E) and P0 (F - J). Sections from nonpigmented eyes were included in C and H to show the expression of Fgf 10 in peripheral neuroretina in Chx10-Cre;Lhx2 ${ }^{\text {lox// }}$; pMes-Fgfl0 eyes (red arrows). Dotted red circles mark the lenses (D, E and J). (Scale Bars: $100 \mu \mathrm{~m}$ )


Supplemental Figure 4: Fgf10 overexpression in control background led to tethering of lens to the cornea. ( $\mathrm{A}-\mathrm{H}$ ) Eye sections of Chx10-Cre;Lhx2 ${ }^{\text {lox/+ }}$ and Chx10-Cre;Lhx2 ${ }^{\text {lox/+ }} ;$ pMesFgfl0 animals immunostained with E-cadherin (red) and N-cadherin (green). Nuclei are counter-stained with DAPI (blue). White asterisks indicate persistent lens stalks seen in Chx10Cre;Lhx2 ${ }^{l o x /+}$; pMes-Fgf10 eyes (B, D, F and H). (I and J) External eye photos showing cornea opacification observed in some Chx10-Cre;Lhx2 ${ }^{\text {lox/+ }} ;$ pMes-Fgf10 animals (J). The anchor point of the lens stalk in the Chx10-Cre;Lhx2 ${ }^{\text {lox/+ }} ; p M e s-F g f 10$ animal could be detected in the image (J; black notched arrowhead).


Supplemental Figure 5: Chx10-Cre;Lhx2 ${ }^{\text {lox/lox }} ;$ pMes-Fgf10 rescue animals expressed lens epithelial cell marker E-cadherin and the lens fiber cell marker $\mathbf{N}$-cadherin. Developmental time-course of immunohistochemistry for N-cadherin (green) and E-cadherin (red) expression in lenses at E11.5 (A - D), E12.5 (E - H), E13.5 (I - L), E15.5 (M - P), E18.5 (Q - T) and P0.5 (U - X) of Chx10-Cre;Lhx2 ${ }^{\text {lox/+ }}$ (A, E, I, M, Q, U), Chx10-Cre;Lhx2 ${ }^{l o x /+} ;$ pMes-Fgf10 (B, F, J, N, R, V), Chx10-Cre;Lhx $2^{\text {lox/lox }}$ (C, G, K, O, S, W) and Chx10-Cre;Lhx $2^{\text {lox/lox }}$;pMes-Fgf10 animals (D, H, L, P, T, X). Nuclei are counter-stained with DAPI (blue). (Scale Bars: $100 \mu \mathrm{~m}$ )

