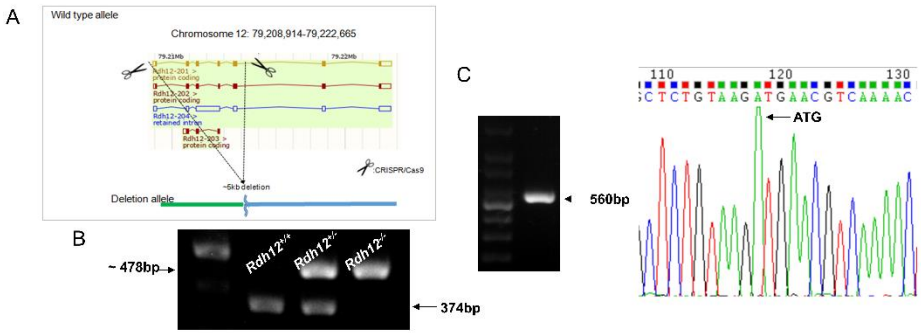
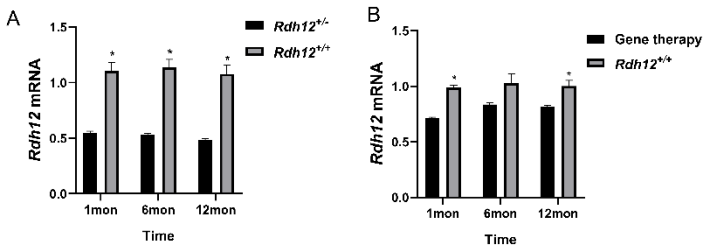
**Supplementary Material: *Rdh12*$^{-/-}$ mice construction and AAV2/8-*mRdh12* expression**

PCR amplification and DNA sequencing were used for *Rdh12*$^{-/-}$ genotyping. Tails of each mouse was collected to isolate genomic DNA. The targeting region was amplified by PCR (Forward 5'-CCTGGGGATTGGACACATACCAACA-3', Reverse 5'-CAGACGGATGGCTCCATTTGCTGTA-3') and (Forward 5'-CCTGGGGATTGGACACATACCAACA-3', Reverse 5'-ACTGGAGTTCCTCCTCAACACGCTA -3'). Studies[1] have shown that different variants of *Rpe65* (Leu450Met or Met450Leu) have different kinetics and resistance to light-induced retinal degeneration. To determine the sequence of *Rpe65* by PCR using the following primer pair: (Forward, 5′-CGGACTTGGGTTGAATCACTTT-3′; Reverse, 5′-TAGTTTGCTGAAGAAAGGCTGGTTA-3′).




Supplementary figure 1: Genotyping of *Rdh12*$^{-/-}$ mouse**.** *Rdh12*$^{-/-}$ mice verification and AAV2/8-*mRDH12* expression. A: Schematic diagram of *mRdh12* knockout strategy. B: Genotyping from *Rdh12*$^{+/+}$, *Rdh12*$^{+/-}$, and *Rdh12*$^{-/-}$ mice. One band length in 478bp represented *Rdh12*$^{-/-}$ genotype; One band length in 375bp represented *Rdh12*$^{+/+}$ genotype; and *Rdh12*$^{+/-}$ showed two bands in electrophoresis gel, they are 375bp and 478bp, respectively. C: PCR product of *Rpe65* is 560bp in length and the Sanger sequencing results of *Rpe65* AA450 position is Atg (Met), not Ctg (Leu).

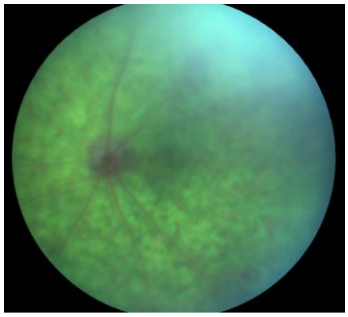
Total RNA was isolated from *Rdh12*$^{+/+}$, *Rdh12*$^{+/-}$, and *Rdh12*$^{-/-}$ adult mice neural retina after 1 months, 6 months and 12 months from birth used RNAiso Plus (9109, TaKaRa, Japan); 1000 ng RNA was reverse-transcribed with a cDNA synthesis kit (RR047A, TaKaRa, Japan). qPCR was performed with synthesized cDNA as a template and TB Green probe for m*Rdh12* (RR820A, TaKaRa, Japan). Gapdh and β-actin were used as reference genes for normalization. qPCR analysis was performed in biological triplicates for each genotype. Total RNA was isolated and detected from the adult mice neural retina after 1 months, 6 months and 12 months of AAV2/8-*mRdh12* subretinal injections.




Supplementary figure 2: A: qPCR of *Rdh12*$^{-/-}$ mouse. Relative mRNA expression of RDH12 were compared between *Rdh12*$^{+/-}$, and *Rdh12*$^{+/+}$ mice in 1, 6 and 12 months, and it showed significant

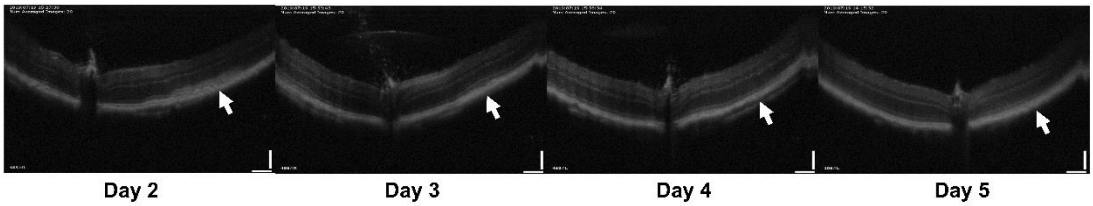
expression differences between *Rdh12*$^{+/-}$, and *Rdh12*$^{+/+}$ mice in all tested months. B: Q-PCR analysis of RDH12 mRNA from *Rdh12*$^{+/+}$ and AAV2/8-m*Rdh12* treatment *Rdh12*$^{-/-}$ mice that *Rdh12*$^{-/-}$ mice received a subretinal injection of AAV2/8-*mRdh12* ($4.5x10^{10}$vg). Averages and SEM were plotted, n=6-8 per group. *$P$ <0.05.

GFP was detected by Micron IV fundus camera after 1 month of AAV2/8-GFP injection.



Supplementary figure 3: Fundus imaging system showed the fluorescence of GFP.

After 5 days, OCT imaging detected the disappearance of bullous retinal detachment and the restoration of retinal structure, which can be considered as a successful injection.




Supplementary figure 4: OCT detected the recovery of retinal structure after subretinal injection.

**Supplementary Methods**

## Virus preparation and titre

Recombinant AAV8 vector was produced through a triple transient transfection method. The plasmid construct, AAV serotype-specific packaging plasmid and helper plasmid, in a ratio of 1:1:3 at 20 μg total DNA per ml of DMEM, were mixed with Polyethylenimine (Polysciences Inc.) to a final concentration of 50 μg ml$^{-1}$ and incubated for 10 min at room temperature to form transfection complexes that were added to 293 T cells at 50 μg DNA per 15-cm plate and left for 72 h. The cells were collected, concentrated and lysed by freeze–thaw (3x) in PBS to release the vector. AAV8 was bound to an AVB Sepharose column (GE Healthcare), and eluted with 50 mM Glycine pH2.7 into 1 M Tris pH 8.8. Vectors were washed in 1 × PBS and concentrated to a volume of 100–150 μl using Vivaspin 4 (10 kDa) concentrators. Viral particle titres were determined by quantitative real-time PCR (qPCR) using an ITR binding assay as previously described.[2]

## References

1. Wenzel A, Reme CE, Williams TP, Hafezi F, Grimm C. The Rpe65 Leu450Met variation

increases retinal resistance against light-induced degeneration by slowing rhodopsin regeneration. *The Journal of neuroscience : the official journal of the Society for Neuroscience*. 2001;21(1):53-58. doi:10.1523/jneurosci.21-01-00053.2001

2. Aurnhammer C, Haase M, Muether N, et al. Universal real-time PCR for the detection and quantification of adeno-associated virus serotype 2-derived inverted terminal repeat sequences. *Hum Gene Ther Methods*. Feb 2012;23(1):18-28. doi:10.1089/hgtb.2011.034