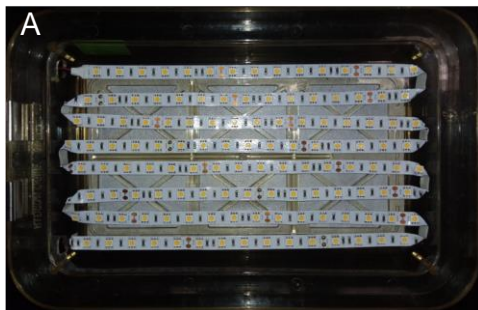
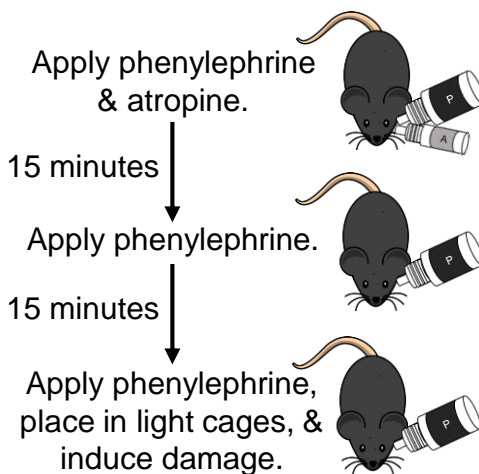


Supplemental Figure 1



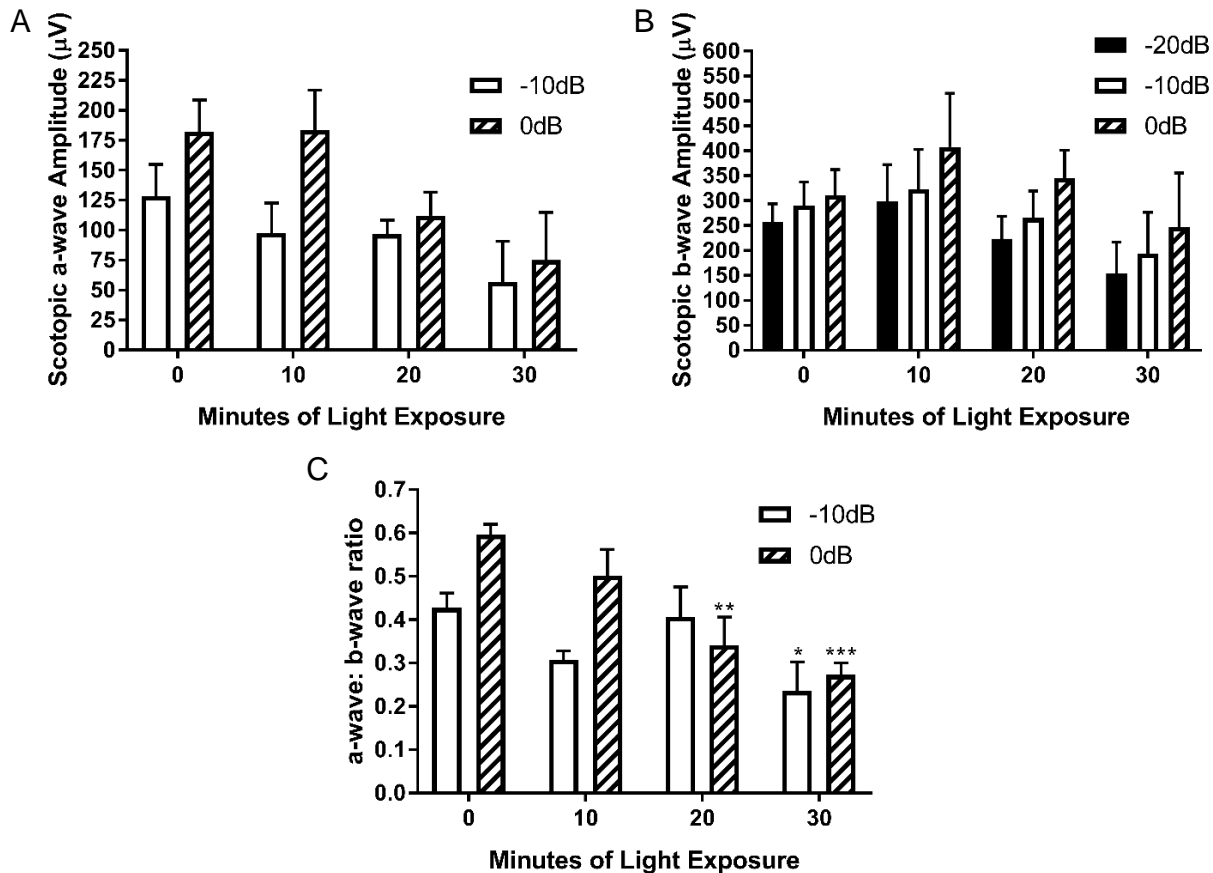
↑ 10, 20, or 30 minutes.

B Dark adapt for >4 hrs.
Protocol initiated between 10:00 P.M. & 12:00A.M.

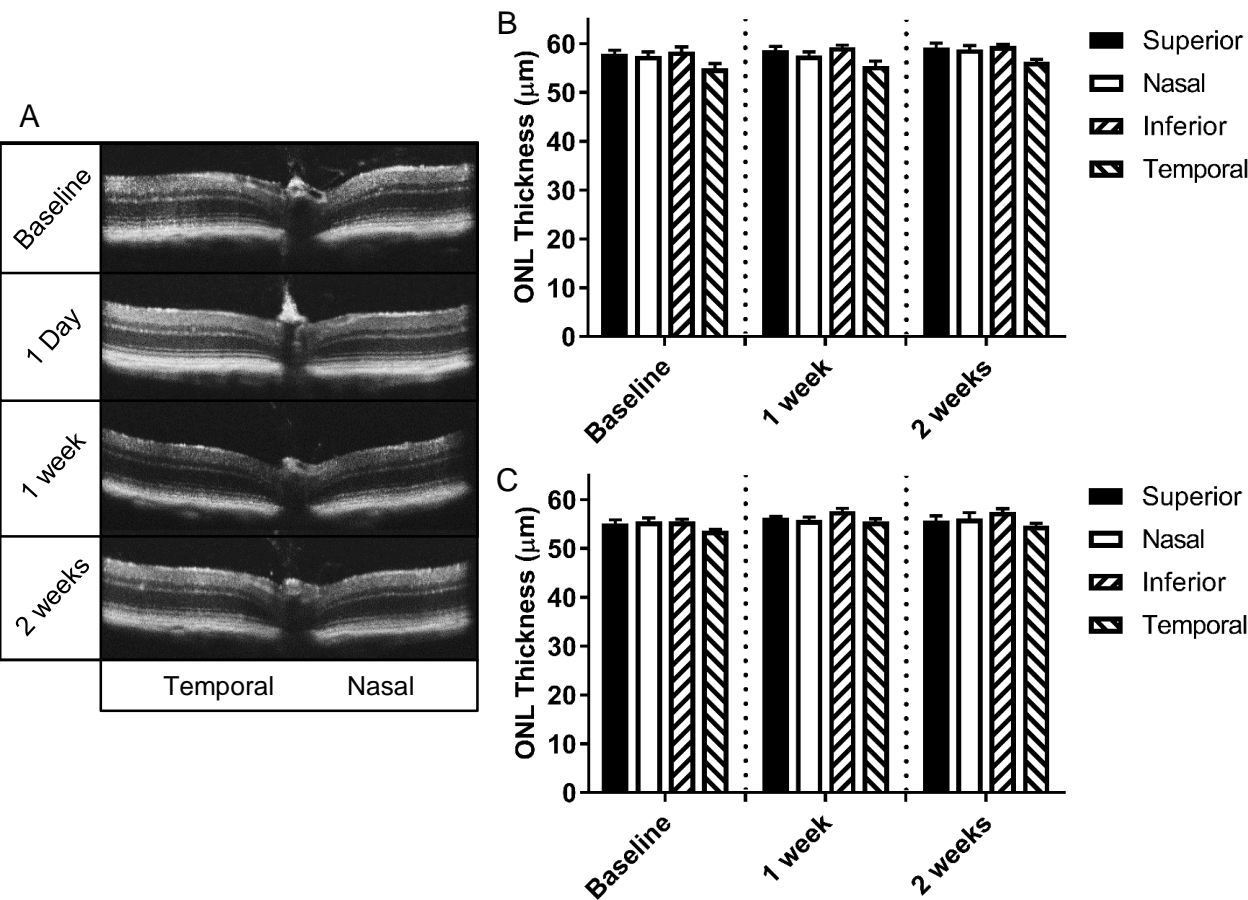


Supplemental Figure 1. The Light Damage Protocol. **A.** LED lights were fastened in rows to the lid of the cages. This ensured even distribution of light to the cage floor. **B.** I307N Rho or WT mice between eight- and twelve-weeks of age were dark adapted for greater than four hours prior to onset of exposure to light, which was initiated during a consistent period of time between 10:00P.M. and 12:00A.M. Pupils of the mice were dilated via successive applications of phenylephrine and/or atropine eye drops separated in time. Mice were then placed in cages equipped with lights modulated to an intensity of 20,000 lux for 10, 20, or 30 minutes.

Supplemental Figure 2



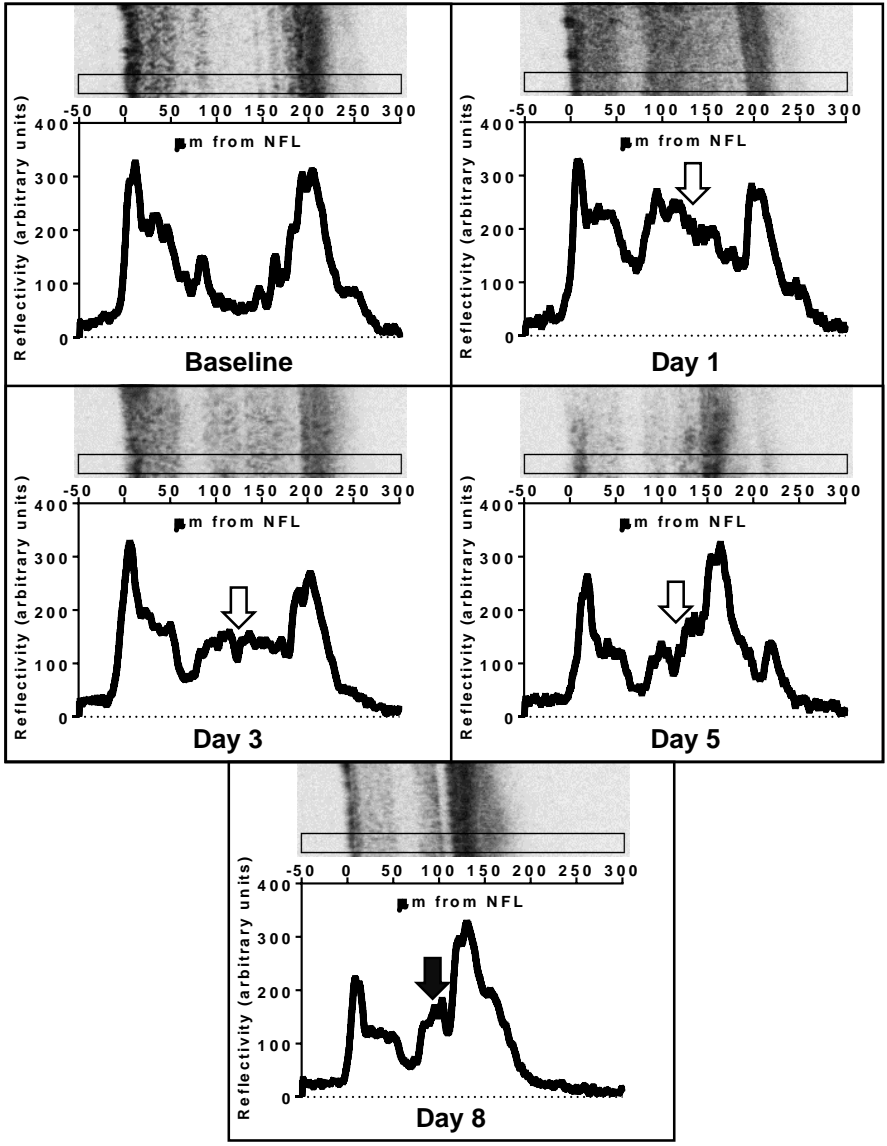
Supplemental Figure 2. The Extent of Functional Retinal Degeneration Increases With Exposure Time to Light. I307N *Rho* mice were subjected to either no light or 10, 20, or 30 minutes of 20,000 lux of light ($n=5$ per group). Scotopic ERG was performed two-weeks after light damage with ascending flash intensities that included -20dB ($2.5 \text{ cd}\cdot\text{sec}/\text{m}^2$), -10dB ($0.25 \text{ cd}\cdot\text{sec}/\text{m}^2$), and 0dB ($0.025 \text{ cd}\cdot\text{sec}/\text{m}^2$). Increased exposure times were associated with larger diminutions of the **A**. a-wave and **B**. b-wave amplitudes. The a-wave corresponding with the -20dB flash intensities failed to increase over background electrical noise. **C**. The b-wave to a-wave ratio was measured in response to flashes with an intensity of $2.7 \text{ cd}\cdot\text{sec}/\text{m}^2$ in a separate cohort of mice that had been exposed to 30 minutes of 20,000 lux of light two-weeks prior. The bar graphs represent the mean ERG amplitudes in microvolts (μV) or the b-wave to a-wave ratio \pm SEM for a sample of 5 mice per group. **A-B**. One-way ANOVA for separate intensities followed by multiple comparisons tests employing the baseline condition as the reference was utilized to determine statistical significance. **C**. A two-tailed unpaired student's t-test was employed to establish statistical significance with a p-value of 0.05.



Supplemental Figure 3. WT Mice are Resistant to the Light Damage Protocol.

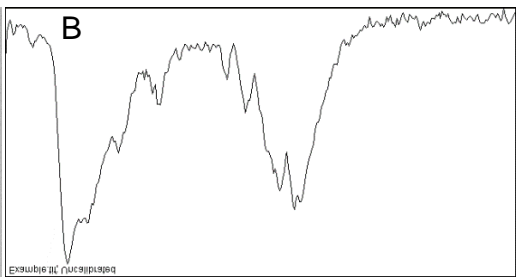
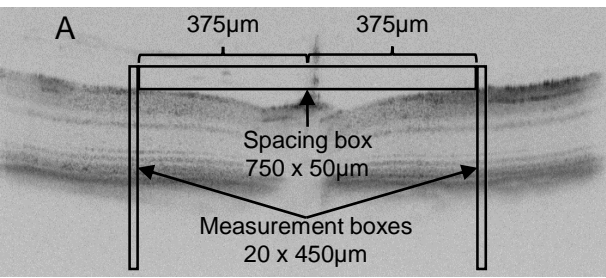
A-B. Four WT littermates of I307N *Rho* mice, aged eight- to twelve-weeks, were subjected to the light damage protocol for a 30 minute duration. SD-OCT scans were performed 2 weeks prior to light challenge, as well as 1 day, 1 week, and 2 weeks after. **A.** Representative SD-OCT B-scans centered on the ONH showing grossly normal retinal architecture. **B.** ONL thickness was not significantly different in any of the measured retinal quadrants after exposing WT mice to bright light. **C.** Five I307N *Rho* mice, aged eight- to twelve-weeks, did not display significant differences in ONL thickness in any of the retinal quadrants without light challenge at the baseline, 1 week and 2 week time points. Bar graphs represent the mean ONL thickness of a retinal quadrant \pm SEM. One-way ANOVA for separate quadrants followed by multiple comparisons tests employing the baseline condition as the reference were utilized to determine statistical significance.

Supplemental Fig. 4



Supplemental Figure 4. Hyper-Reflectivity on SD-OCT Characterizes the Acute Phase Following Light Damage. Longitudinal reflectivity profiles were generated utilizing ImageJ from a longitudinal 20 μ m section of the SD-OCT B-scan positioned 375 μ m from the ONH. Representative profiles corresponding to the baseline, as well as day 1, 3, 5, and 8 post-light damage time points, are displayed. The baseline reflectivity profile displayed a hypo-reflective area that overlapped with the ONL. With progression to days one and three after injury, the area of the plot encompassing the ONL evolved a positive deflection signifying the hyper-reflective signal that seems to originate from the OPL and subretinal space. The positive deflection largely disappeared by day five, and was absent by day eight. Interruption of the positive deflection of the reflectivity signal (white arrows) may indicate the area where the hyper-reflectivity associated with the OPL and subretinal space converge in the ONL. A new hyper-reflective signal emerged at day eight (black arrow) that likely represented the unnatural juxtaposition of the OPL and OLM.

Supplemental Figure 5



C

A	B
Reference	Match?
1	=MATCH(A1,C:C,0)
1	...
2	...
2	...
3	...
3	...
4	...
4	...
...	...
...	...
650	...
650	=MATCH(A1,C:C,0)

D

A	B	C
Reference	Match?	X-value
1	2	1
1	2	1
2	#N/A	3
2	#N/A	3
3	4	4
3	4	4
4	6	6
4	6	6
5	#N/A	7
5	#N/A	7
6	8	8
6	8	8

E

2	#N/A
2	#N/A
5	#N/A
5	#N/A
6	8
6	8
4	6
4	6
3	4
3	4
1	2
1	2

Supplemental Figure 5. Generation of Longitudinal Reflectivity Profiles with ImageJ.

A. A spacing box was drawn to the dimensions of 750 x 50 μ m and centered over the ONH with the lower corners contacting the NFL. Reflectivity was detected with the analyze gel function in measurement boxes with dimensions of 20 x 450 μ m placed at the same height of, but just lateral to, the spacing box. **B.** The analyze gel function generated the reflectivity curves, which had to be vertically transformed before being saved as an X-Y coordinate system in a .txt file. The X-Y coordinates generated by ImageJ contain X-values that are integers, each appearing twice (e.g. 1, 1, 2, 2...). The minimum and maximum X-values in the X-Y coordinate sets are 1 and 650 (e.g. 1, 1, 2, 2, ... 649, 649, 650, 650). However, ImageJ skips many X-values (e.g. 1, 1, 2, 2, 4, 4), therefore missing X-values need to be populated with **C.** a formula in Excel. All possible X-values were pasted into "column A" as a reference (1, 1, 2, 2, ... 649, 649, 650, 650). The excel formula (=MATCH(A2,C:C,0)) was input into each cell of "column B." **D.** The X-values for a given X-Y coordinate set were pasted into "column C". "Column B" quickly populates with either #N/A or a random number. #N/A means that the X-value in column C of that row is missing in the X-Y coordinate set. The random number means that the X-value is not missing. For the example in the figure, X-values 2 and 5 are missing. **E.** Copy and special paste the values from "column C" and "column D" into adjacent columns of a new spreadsheet. Missing X-values can be quickly grouped by sorting "column C" in descending alphabetical order, making sure to *expand the selection*. X-values adjacent to #N/A in the now sorted "column D" can then be copy and pasted into the X-Y coordinate set. This technique was modified from: (https://www.youtube.com/watch?v=zktCVGfH1_k).