**Supplemental Figure 1.** Detected PC, PE and PS in *Elov4* KO retina. (A) Relative percent of the total phosphatidylcholine (PC) molecular species containing LC-PUFAs (14 ≤ C ≤ 24). (B) VLC-PUFAs were not detected in phosphatidylethanolamine (PE) in WT and KO retina at 8 weeks and 12 months of age. Fatty acids provided in parenthesis are not absolute but rather most probable combinations. (C) VLC-PUFAs were not detected in phosphatidylserine (PS) in WT and KO retina at 8 weeks and 12 months of age. *p ≤ 0.05; **p ≤ 0.01; ***p ≤ 0.005; ****p ≤ 0.001. Quantitative data are expressed as mean ± SEM (n = 4 independent retinal samples per genotype).

![Graph A](image1.png)

![Graph B](image2.png)

**Supplemental Figure 2.** Rod function was similar in 5-week-old mice. (A) Rod a- and b-wave responses to increasing light intensities were not different between the mice. (B) Rmp$^3$ and V$_{max}$ were not different between the mice (n = 11 WT, 4 Het, 8 KO). Quantitative data are expressed as mean ± SEM.
Supplemental Figure 3. VLC-PUFA-deficient mice have a delayed rod response time. Rod b-wave implicit time was increased in KO mice compared to WT and Het mice (n = 8 WT, 6 Het, 6 KO). Quantitative data are expressed as mean ± SEM.

Supplemental Figure 4. Retinas were similar at 5 weeks between WT, Het, and KO mice. Representative H&E-stained retina from WT, Het, and KO mice. Images were acquired at 240 nm superior to the optic nerve head. RPE, retinal pigmented epithelium; OS, outer segments; IS, inner segments; ONL, outer nuclear layer; OPL, outer plexiform layer; INL, inner nuclear layer; IPL, inner plexiform layer; GCL, ganglion cell layer. Scale bar = 50 μm.
Supplemental Figure 5. KO mice had retinal atrophy at 12 months. Representative H&E-stained retina from WT, Het, and KO mice showed that the outer nuclear layer (ONL) was thinner in the KO retina compared to control mice. Images were acquired at 240 nm superior to the optic nerve head. RPE, retinal pigmented epithelium; OS, outer segments; IS, inner segments; ONL, outer nuclear layer; OPL, outer plexiform layer; INL, inner nuclear layer; IPL, inner plexiform layer; GCL, ganglion cell layer. Scale bar = 50 μm.