

Fig. S1. Lineage tracing to examine maintenance of photoreceptor cell fate and position in the retinal epithelium.

(A) A more representative example of the 75% p.d. wildtype apical network than that in Fig. 1D.

(B) Genetic elements of the G-TRACE lineage trace system result in coexpression of GFP and RFP in photoreceptor nuclei: *chp-Gal4* drives photoreceptor-specific expression of RFP and FLP. FLP removes the stop in the actin-stop-GFP cassette, establishing a permanent lineage mark (GFP).

(C) Lineage-traced photoreceptor nuclei in a WT 100% p.d. retina reside apically and are never detected beneath the fenestrated membrane.

(D,E) Lineage-traced photoreceptor nuclei in an *ablnull* 100% p.d. retina show that even though cell position changes, with some nuclei found beneath the fenestrated membrane, cell fate is not changed. Schematic shows the apical (red) and basal (blue) planes imaged.

Scale bars = 10 μ m.

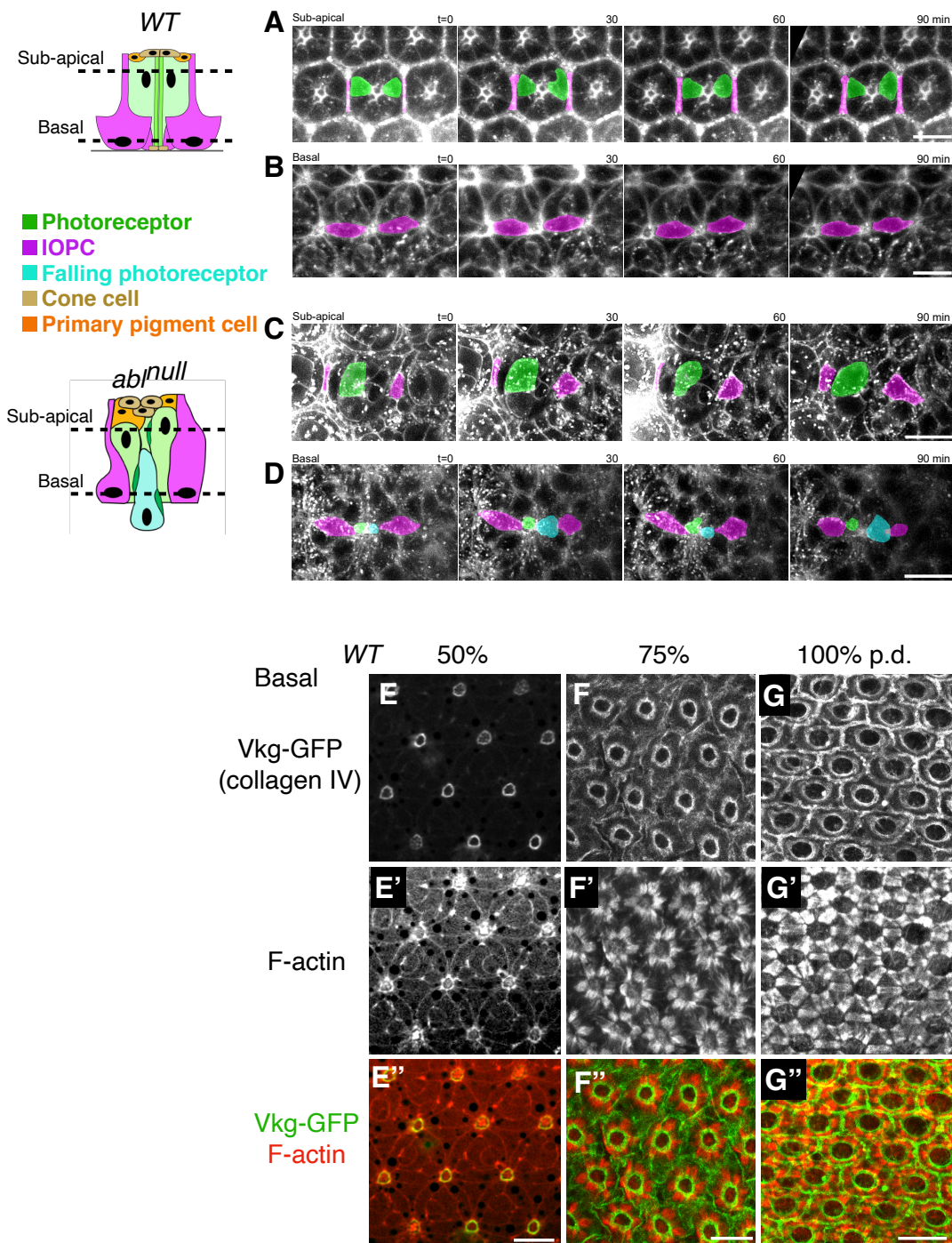


Fig. S2. Time-lapse images comparing apical and basal network pattern in *WT* and *abl* mutant retinas.

(A-D). Schematics and stills from time-lapse movies (Videos S1 and S2) of 50% p.d. retinas injected with CellMask (white). False color shows photoreceptors (green), fallen photoreceptors (cyan) and secondary IOPCs (magenta) in a representative ommatidium. *Abl* loss results in irregular photoreceptor and secondary IOPC cell shapes and contacts that change over the time-course. Basally, photoreceptor “falling” is marked by the appearance of the cyan-marked cell between IOPC feet. If comparing the stills shown in **(D)** with Video S2, please note that the plane of sectioning for generating the lateral view video passes through the four labelled cells visible in the basal plane. Between ~75-85 min in the video, the most apical part (neck) of the fifth cell (the left-most cyan falling photoreceptor) is visible in the basal xy view, but is not within the plane of sectioning and therefore is not visible in the lateral view. Similarly, the majority of the cellular volume seen in the lateral view falls below the left-most magenta cell and so is not seen in the xy basal view.

(E-G). Progression of basal ECM deposition at 50, 75 and 100% p.d., visualized with Viking-GFP (collagen IV), occurs in sync with the contraction of the IOPC feet and expansion of the central rings in a *WT* retina. Scale bars = 10 μm .

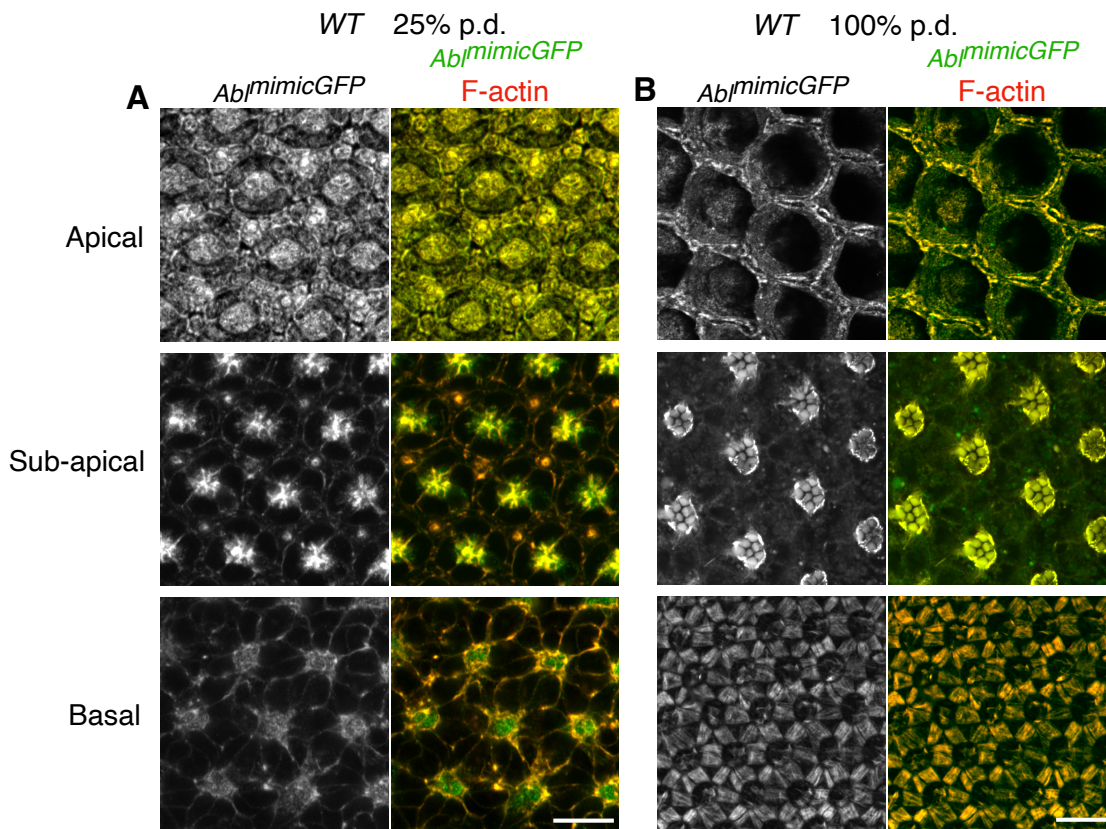


Fig. S3. *AbI* is enriched in photoreceptor and IOPC F-actin structures at 25% and 100% p.d.. (A,B) Apical, subapical and basal planes showing the enrichment of *AbI^{mimicGFP}* and F- actin in photoreceptors and IOPCs. Scale bar = 10 μ m.

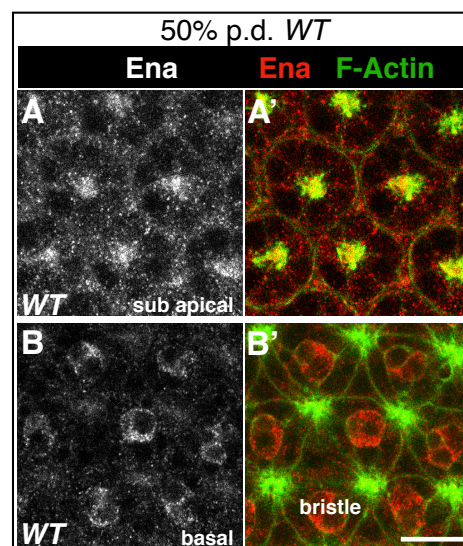


Fig. S4. *Ena* protein expression in a wildtype 50% p.d. retina. (A,B) *Ena* protein expression at subapical and basal planes. Scale bar = 10 μ m.

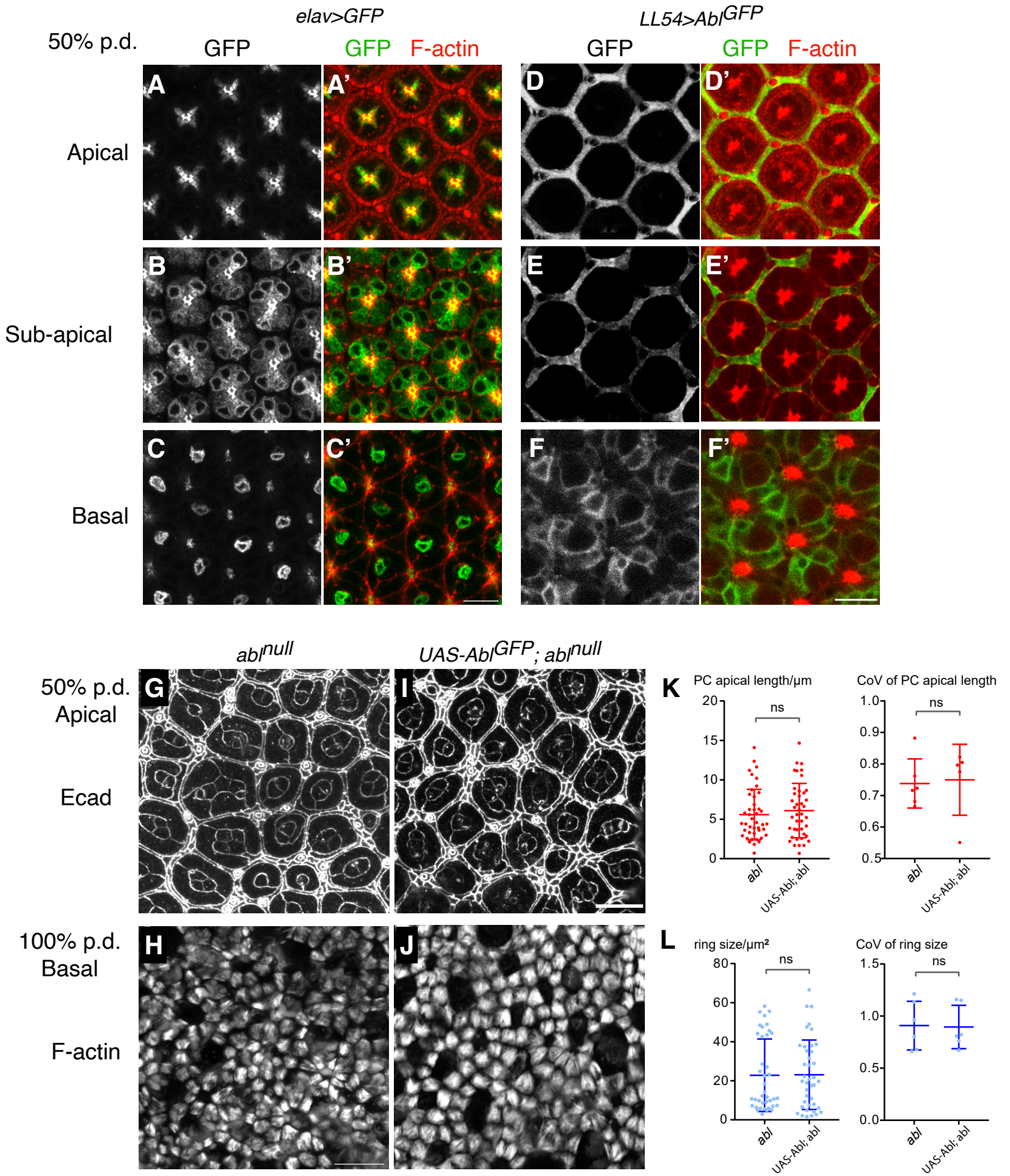


Fig. S5. Specificity of Elav- and LL54-Gal4 expression and lack of rescue by UAS-Abi^{GFP} in the absence of a Gal4 driver.

(A-C') Elav-Gal4 drives expression specifically in photoreceptors and bristle neurons, with no leaky expression detected in IOPCs, cone or primary pigment cells. Because pan-neuronal expression of UAS-Abi^{GFP} is lethal, we expressed GFP (UAS-Cd8-GFP, Bloomington Stock 32187).

(D-F') LL54-Gal4 drives expression specifically in IOPCs, with no leaky expression detected in photoreceptors or cone cells.

(G-L) Without a Gal4 driver, the UAS-Abi^{GFP} transgene does not rescue apical or basal network pattern. **(G)** shows the Fig. 1F *ab^{null}* disc, cropped and oriented with the two ommatidia in the top right of 1F at the bottom center of **G**. Scale bars = 10µm. **(K,L)** Measurement and CoV of secondary IOPC apical length and basal ring size. Plots of secondary IOPC show measurements made in at least 30 ommatidia in a single disc for each genotype. In the CoV plots, for each genotype, and for each data point, measurements were made in at least 30 ommatidia/retina, n= minimum of 4 retinas. The *ab^{null}* data sets in **K** were also used for Fig. 1I and Fig. 4K. The *ab^{null}* data sets in **L** were also used for Fig. 1O and Fig. 6G,H.

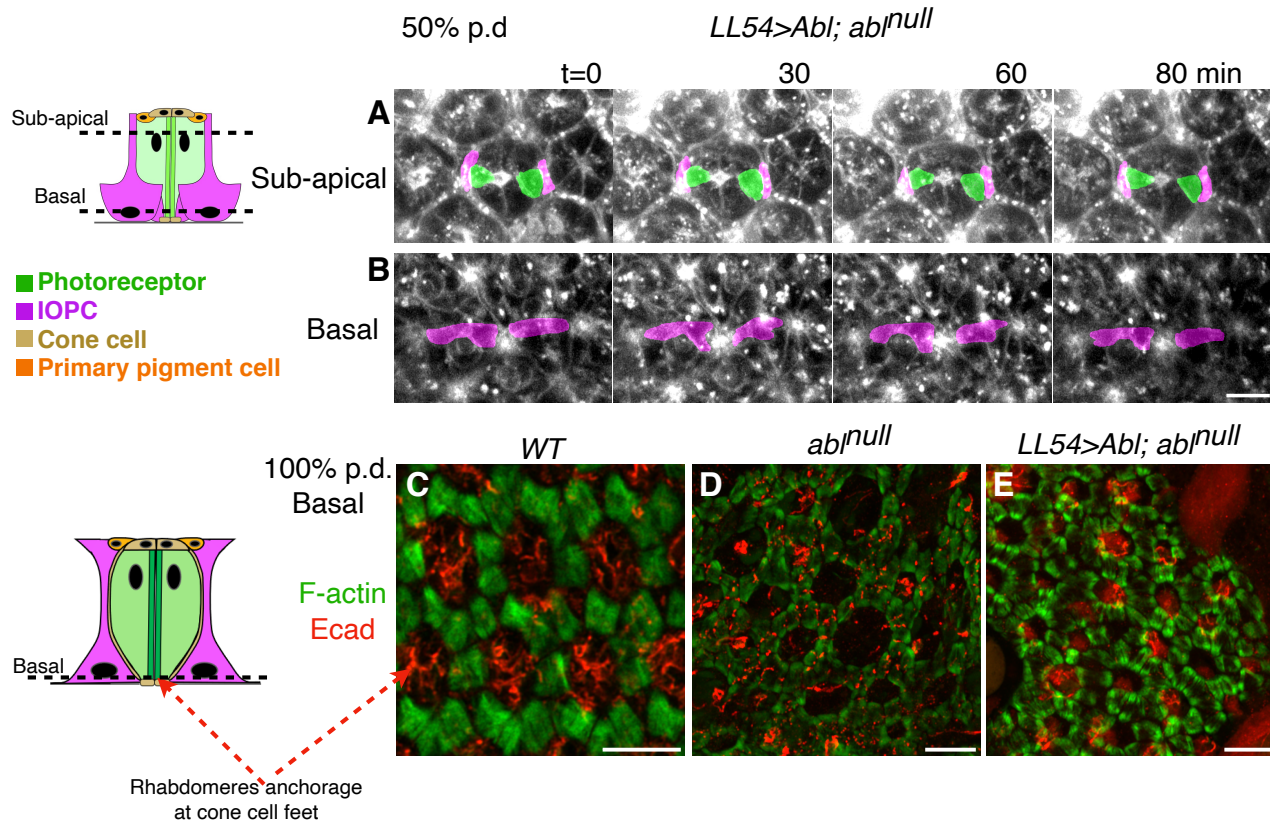
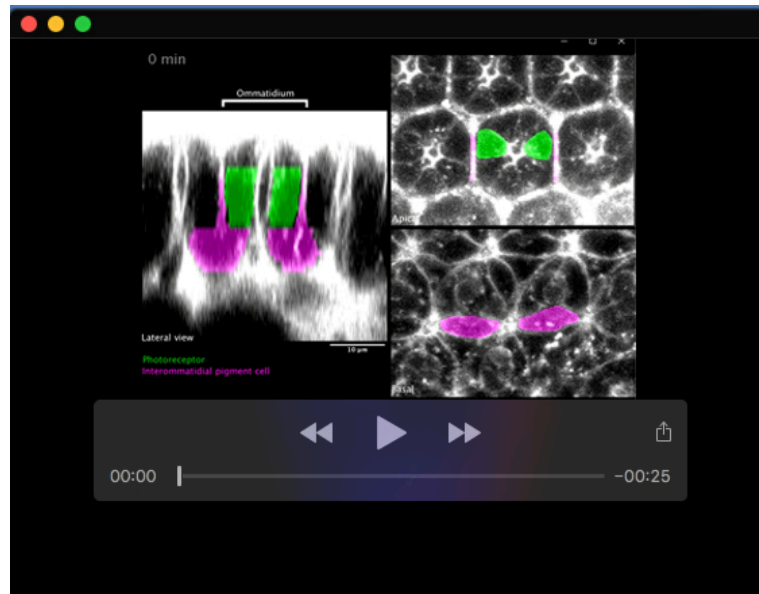


Fig. S6. IOPC-specific expression of Abl in an otherwise *ablnull* retina restores correct cell-cell contacts within the scaffold.

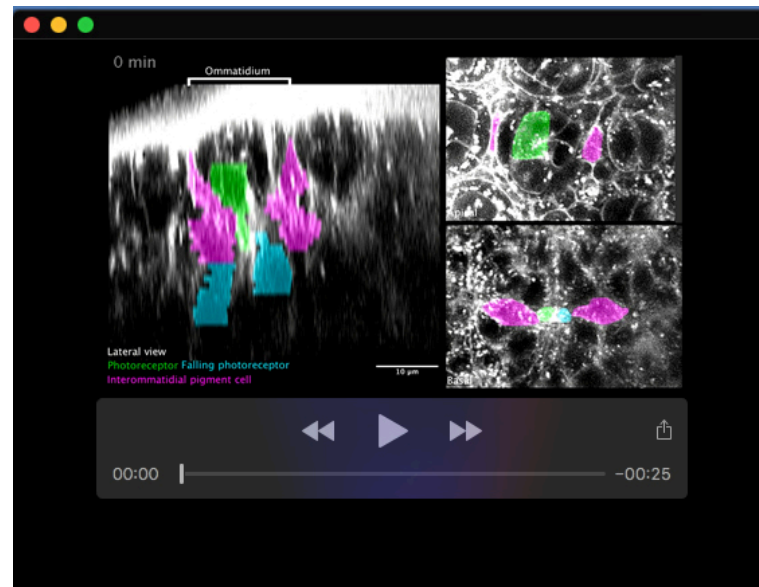
(A,B) Schematics and stills from a time-lapse movie (Videos S3) of a 50% p.d. *LL54>Abl; ablnull* retina injected with CellMask (white) showing restoration of apical and basal pattern. False color shows photoreceptors (green) and secondary IOPCs (magenta) in a representative ommatidium. Compare to Figure S2A-D.

(C-E). Basal plane of 100% p.d. retinas, with schematic, shows how restoration of Abl to the IOPCs rescues the anchorage of the rhabdomeres to the cone cell feet.

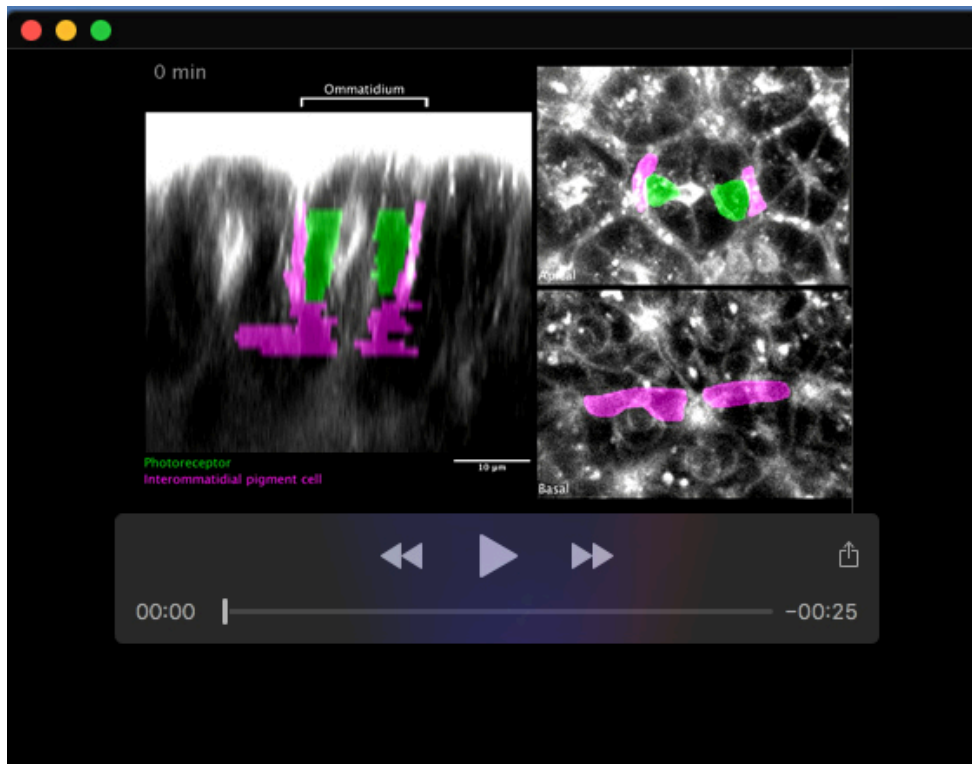
Scale bars = 10µm.



Movie 1. Time lapse movie of a 50% p.d. WT retina injected with CellMask to show cell outlines and rhabdomeres and with a representative tracked ommatidium false-colored to identify photoreceptors and IOPCs. Frames are re-aligned to keep the center of the tracked ommatidium constant. Framer rate: 5 min/frame; Duration: 90 min. Scale bar: 10 μ m. Related to Figure 1 and S2.



Movie 2. Time lapse movie of a 50% p.d. *AbI*^{null} retina injected with CellMask to show cell outlines and rhabdomeres and with a representative tracked ommatidium false-colored to identify photoreceptors and IOPCs. Frames are re-aligned to keep the center of the tracked ommatidium constant. Framer rate: 5 min/frame; Duration: 90 min. Scale bar: 10 μ m. Related to Figure 1 and S2.



Movie 3. Time lapse movie of a 50% p.d. *LL54>Abi^{GFP};abi^{null}* retina injected with CellMask to show cell outlines and rhabdomeres and with a representative tracked ommatidium false-colored to identify photoreceptors and IOPCs. Frames are re-aligned to keep the center of the tracked ommatidium constant. Framer rate: 5 min/frame; Duration: 90 min. Scale bar: 10μm. Related to Figure 5 and S6.