**Figure S1: (A)** HCs subtypes and markers in the zebrafish retina.

B-C) Typical examples of HCprs labelled with **(B)** Connexin55.5:rasGFP (Cx55.5) and **(C)** Trβ2:tdTomato. Red arrowheads point to HCs within the HC layer. Dashed line represents the OPL. Scale bar 10μm. Insets of the red-boxed region shows magnified HC. Scale bar 5μm. **(D)** Schematic showing measurement of HC distance from the OPL at rounding (left) (analyzed data can be found in Figure 1E and divided by marker the same data is seen in Supplementary Figure 1E) and HC angle of division (right) (analyzed data can be found in Figure 1G). Scale Bar, 5 μm. **(E)** Position of mitotic HCprs for different HC marker populations. (The pooled version of this analysis can be found in Figure 1E this is the same data divided by marker) **(F)** Schematic showing depth of migration measurements. Relative thickness of the INL was quantified by drawing a line (D) from the OPL to the IPL using Fiji Line tool. To quantify HC position, a line (d') was drawn from the center of HC to the OPL. Relative percentage of HC depth was defined by dividing d'/D. For earlier developmental stages before the IPL formation, thickness of the INL was measured once the IPL was formed within the same region (analyzed data can be found in Figure 3 F and G).
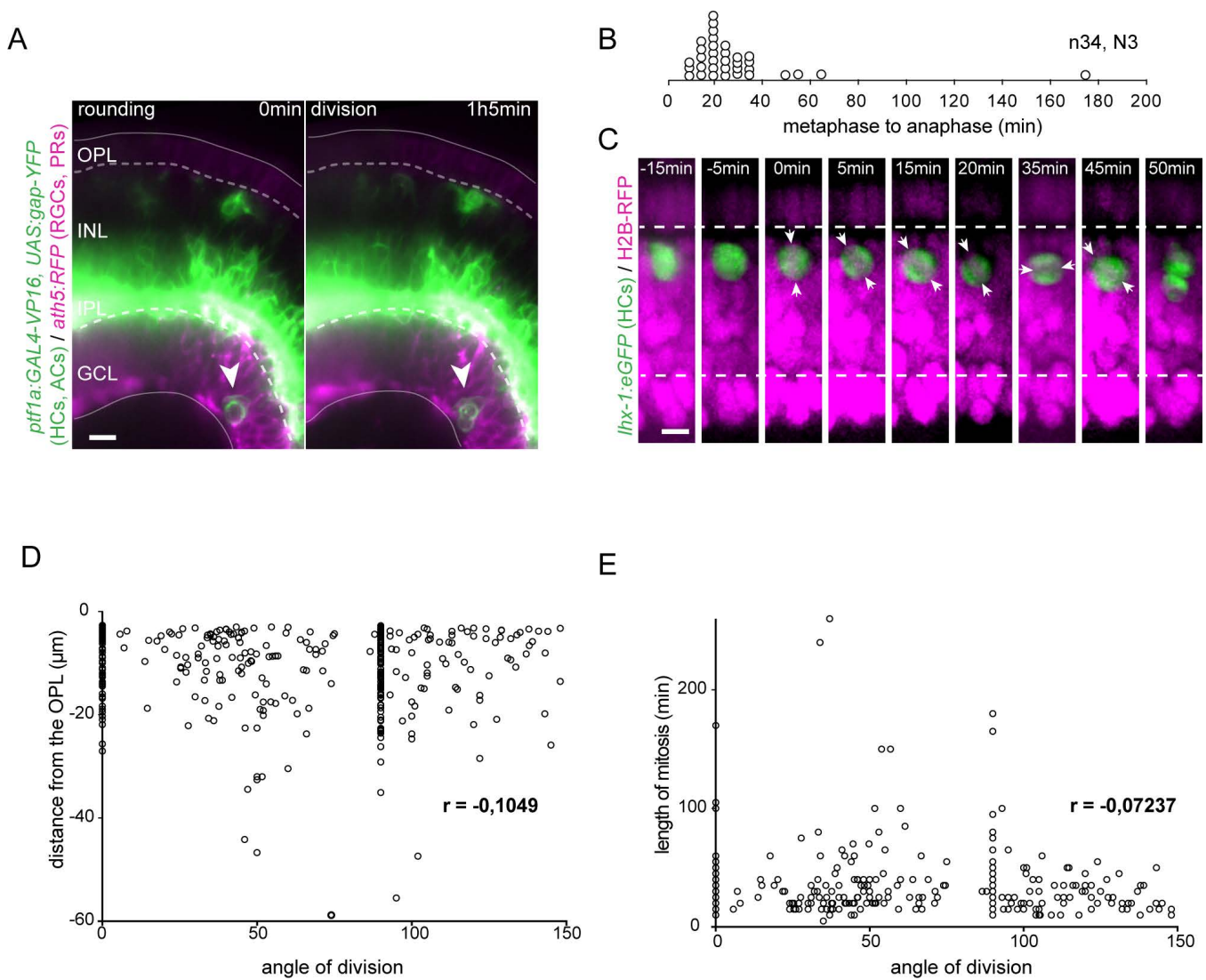
Figure S2: Heterogeneity in mitotic position and behaviour of committed HCpr subtypes.

(A) Rare example of HCpr dividing in the RGC layer. ptf1a:GAL4-VP16, UAS:gap-YFP (green, HCpr and AC); ath5:RFP (PR and RGC). White arrow: mitotic HCpr. White dashed line: OPL (top), IPL (bottom). Scale Bar, 10 μm. **(B)** Spread of metaphase to anaphase duration in mitotic HCprs. **(C)** Example of HCpr spindle rocking before mitosis. lhx-1:eGFP (HCpr, green); H2B-mcherry (nuclei, magenta), white arrows: position of metaphase plate. White dashed line: the OPL (top), the IPL (bottom). Scale Bar, 5 μm. **(D)** Distribution of HCpr mitotic angle versus position does not show a strong correlation. **(E)** Distribution of HCpr angle versus length of mitosis does not show a strong correlation.
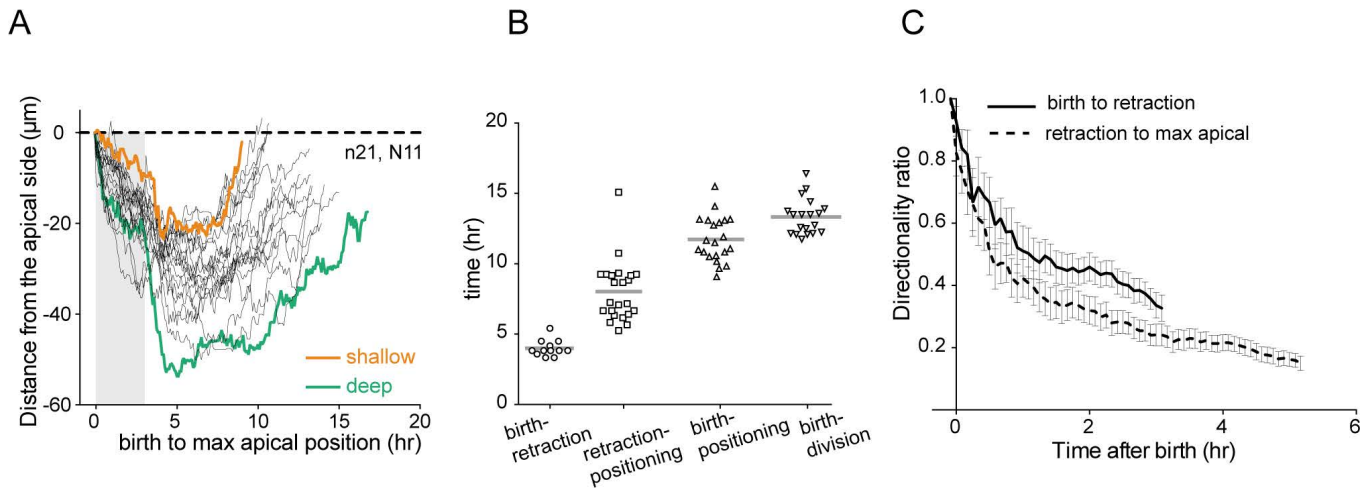
Figure S3: HCpr migration is heterogenous

(A) Single cell trajectories of HCs from birth to final positioning. This graph depicts distance of migrating HCs shown in Figure 3C in micron from apical side. **(B)** Scatter plot showing length of different phases of HC migration. **(C)** Directionality plots of HCs for different phases of migration; from birth to retraction of the apical attachment (black line, first 1hr33min) and from retraction to final positioning (black dotted line, first 2hr40min).
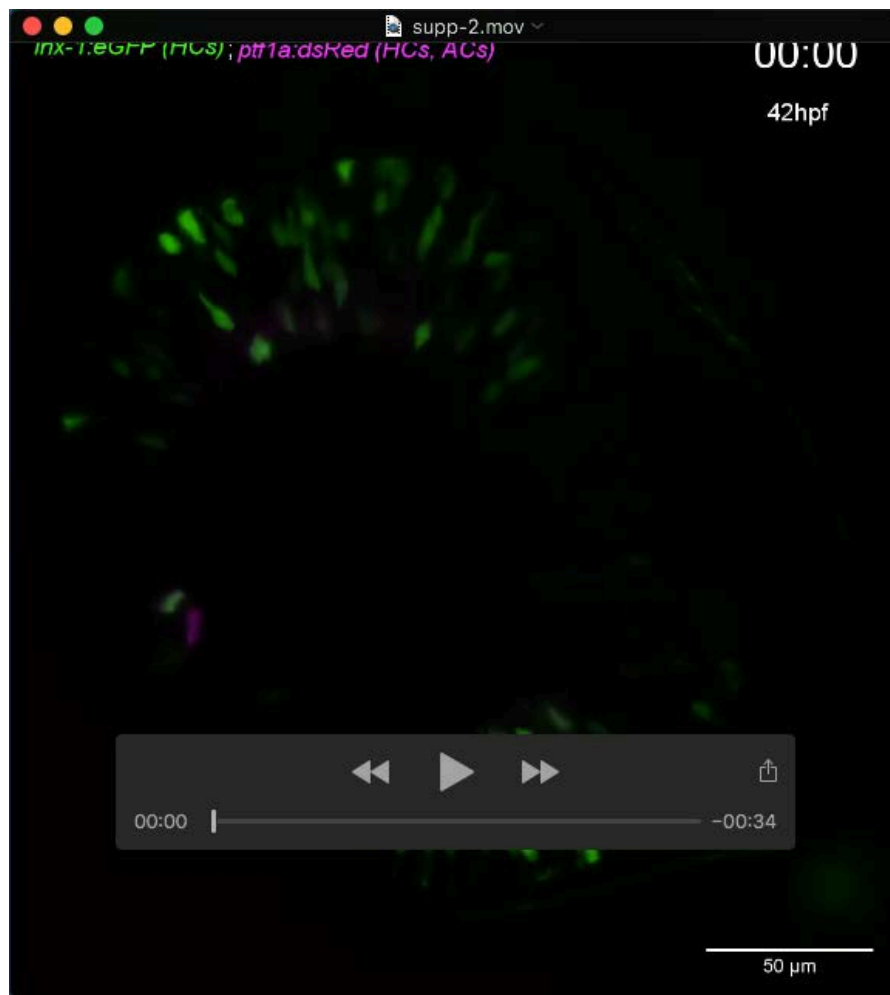
Movie 1: HC lamination during zebrafish retina development. Light-sheet time-lapse imaging of HC layer formation during retina development. lhx-1:eGFP (green) labels HCs and ptf1a:dsRed labels both HCs and ACs (magenta). HCs are born at the apical side of the retina and then migrate to the basal INL. After stop-over within the AC layer (magenta), HCs undergo apical migration to form the thin HC layer below the OPL at the apical side of the retina. Imaging started around 42hpf and ended at 70hpf. Time interval 5min. Mitotic HCprs are labelled with an arrow. Time in h:min and scale bars = 50 μm. Related to Figure 1 and 3.
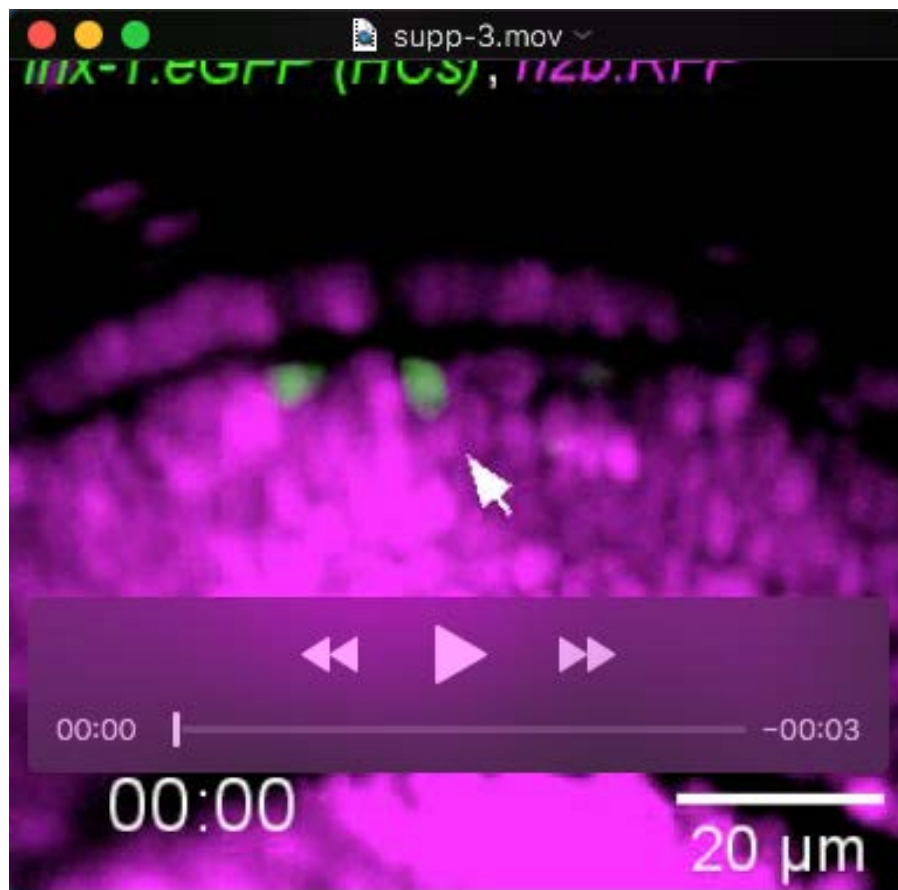
Movie 2: HC exhibit spindle rocking during division. Time-lapse imaging of HC division dynamics using Tg(lhx-1:eGFP); Tg(hsp70:h2b:RFP). lhx-1:eGFP (green) labels HCs and H2B-RFP labels nuclei (magenta). Arrow points to the tracked HC. Time interval 5min. Time in h:min and scale bars = 20 μm. Related to Figure 1 and Supplementary Figure 2.
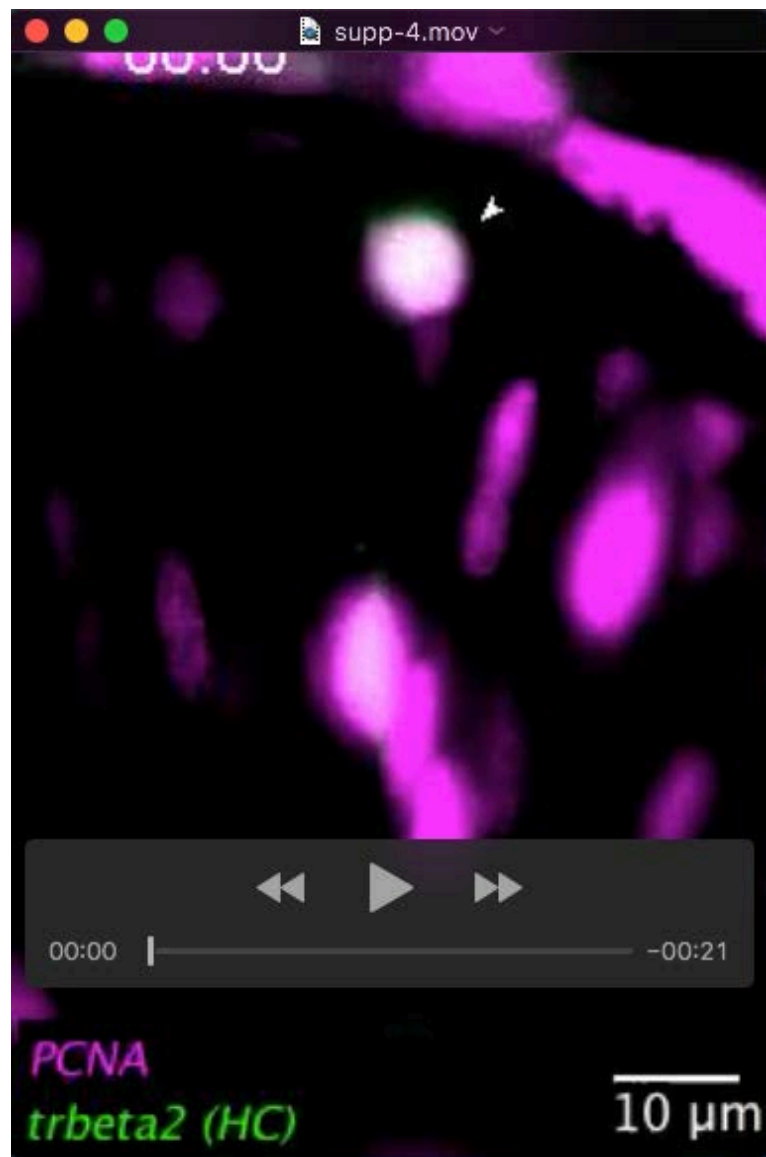
Movie 3: HC cell cycle in a developing retina. Light-sheet time-lapse imaging of PCNA dynamics from HC birth to its final cell cycle. Mosaic expression of hsp70:GFP-PCNA (magenta, nuclei) and Trβ2:tdTomato (green, HCs and PRs). White arrowhead points to the tracked HC. Cyan and red arrowheads point to HC sister cells after HC final division. Time interval 5min. Time in h:min and scale bars = 10 μm. Related to Figure 2.
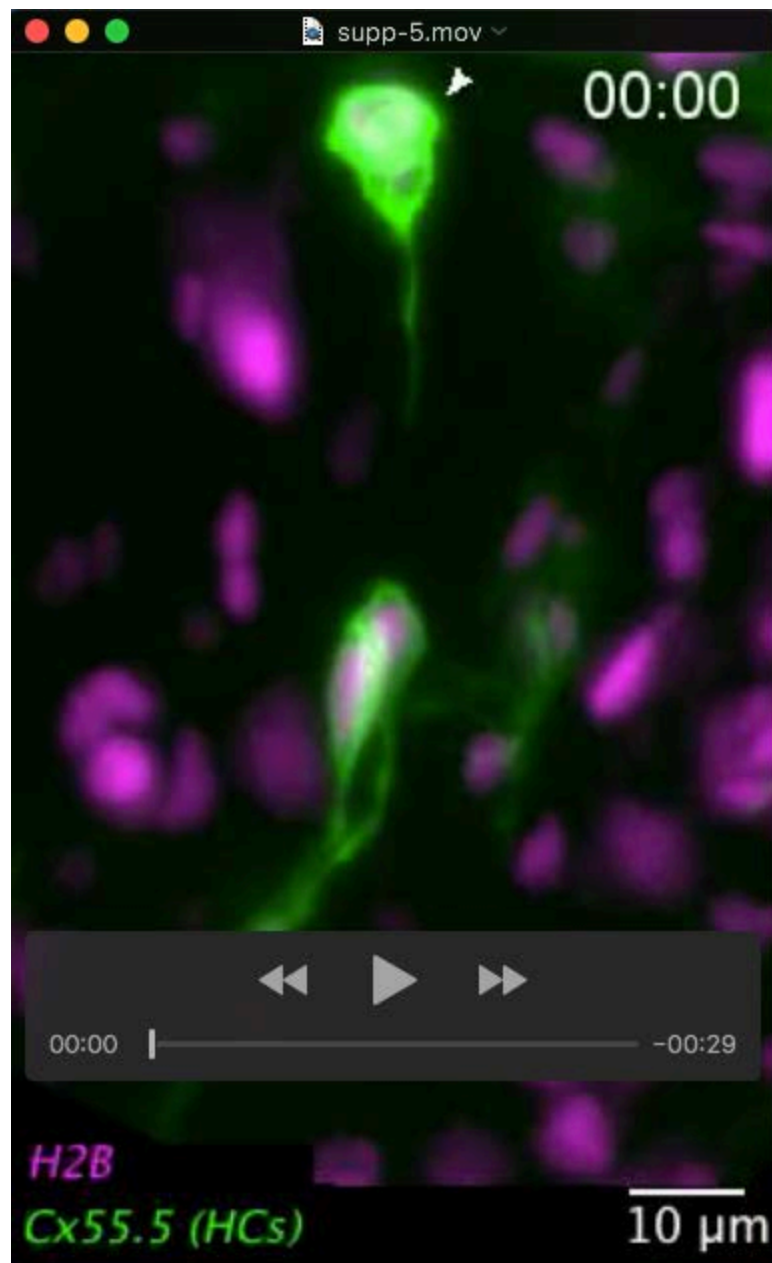
Movie 4: HCs undergo a bidirectional migration. Light-sheet time-lapse imaging of HC migration dynamics from birth to final positioning. Connexin55.5:rasGFP (green, HCs) and hsp70:H2B-RFP (magenta, nuclei). White arrowhead points to the tracked HC. Cyan and red arrowheads follow HC sister cells from division to final positioning. Time interval 5min. Time in h:min and scale bars = 10 μm. Related to Figure 3 and Supplementary Figure 3.

Movie 5: HC sister cells undergo heterogenous dispersion. Light-sheet time-lapse imaging of HC sister cells from birth (HC final division) to final positioning. Transplanted lhx-1:eGFP (grey, HCs and PRs) into a control retina (left) shows that two sister cells end up at close vicinity to each other. The two sister cells final position is far from each other (grey, HCs). White arrowhead points to the tracked HC before its division. Cyan and red arrowheads follow HC daughter cells from division to final positioning. Time interval 5min. Time in h:min and scale bars = 10 µm. Related to Figure 4 and Supplementary Figure 3.

Table S1: Zebrafish lines corresponding to images and plots in figures

Line	Structures labeled	Figure	Reference
Tg(SoFa2)	Membranes of all retinal cells	1C, E-J Suppl. 1E Suppl. 2A, D-E	(Almeida et al., 2014)
Tg(ptf1a:Gal4-VP16, UAS:gap-YFP)	Membranes of ACs and HCs	1E-J Suppl. 1E Suppl. 2D-E	(Pisharath and Parsons, 2009) (Williams et al., 2010)
Tg(ptf1a:DsRed)	ACs and HCs	1B, D-J 3F-H	(Vitorino et al., 2009)
Tg(lhx-1:eGFP)	HCs and PRs	1B, D-J 3 4A, B, E, F Suppl. 1D-E Suppl. 2B-E Suppl. 3	(Swanhart et al., 2010) (Boije et al., 2015)
Tg(hsp70:H2B-RFP)	Chromatin	Suppl. 2B-C	(Dzafic et al., 2015)
Tg(βactin:maKate2-ras)	All membranes	1E-J 3B-D Suppl. 1E Suppl. 2D-E	(Matejčić et al., 2018)
Tg(ath5:gap-RFP)	Membranes of RGCs and PRs	1B Suppl. 1E Suppl. 2A	(Zolessi et al., 2006) (Icha et al., 2016)
Tg(actb2:mCherry-Hsa.UTRN)	F-Actin	1E-J 3A Suppl. 1D-E	(Compagnon et al., 2014)

Table S2: List of constructs corresponding to images and plots in figures

Construct	Structures labeled	Figure	Reference
trβ2:tdTomato	HCs and PRs	1E-J 2 3B-D Suppl. 1C, E Suppl. 2D-E	(Suzuki et al., 2013)
hsp70:PCNA-GFP	Cell cycle phase marker	2A-E Suppl. 2C	(Icha et al., 2016)
Connexin:55.5:rasGFP	Membrane of HCs	1E-J 3B-D 4C, E-F Suppl. 1B, E Suppl. 2D-E	(Weber et al., 2014)
hsp70:H2B-RFP	Chromatin	Suppl. 1B	(Strzyz et al., 2015)

Table S3: List of *in vivo* time lapses used in this study

Movie	Transgenes/ constructs injected	analyzed HC	Time-lapse started at (hpf)	Duration of time-lapse (hr)
1	*Tg(SoFa2)*	24	36	40
2	*Tg(SoFa2)*	18	42	34
3	*Tg(SoFa2)*	48	40	38
4	*Tg(ptf1a:Gal4-VP16, UAS:gap-YFP)*	32	35	42
5	*Tg(ptf1a:Gal4-VP16, UAS:gap-YFP)*	11	42	35
6	*Tg(ptf1a:Gal4-VP16, UAS:gap-YFP)*	3	48	10
7	*Tg(lhx-1:eGFP); Tg(ptf1a:DsRed)*	86	40	40
8	*Tg(lhx-1:eGFP) transplanted into WT*	12	42	40
9	*Tg(lhx-1:eGFP) transplanted into Tg(βactin:maKate2-ras)*	8	42	40
10	*Tg(lhx-1:eGFP); Tg(ath5:gap-RFP)*	37	42	35
11	*Tg(lhx-1:eGFP); Tg(ath5:gap-RFP)*	6	48	24
12	*Tg(lhx-1:eGFP); Tg(actb2:mCherry-Hsa.UTRN)*	31	48	30
13	*Tg(lhx-1:eGFP); Tg(actb2:mCherry-Hsa.UTRN)*	31	48	35
14	*Tg(lhx-1:eGFP); Tg(hsp70:H2B-RFP)*	33	42	30
15	*Tg(lhx-1:eGFP); Tg(hsp70:H2B-RFP)*	4	42	31
16	*trβ2:tdTomato* and *hsp70:PCNA-GFP* were co-injected.	1	42	11
17	*trβ2:tdTomato* and *hsp70:PCNA-GFP* were co-injected.	11	48	20
18	*trβ2:tdTomato* and *hsp70:PCNA-GFP* were co-injected.	4	42	20
19	*trβ2:tdTomato* and *hsp70:PCNA-GFP* were co-injected.	8	42	40
20	*trβ2:tdTomato* and *hsp70:PCNA-GFP* were co-injected.	5	42	35
21	*trβ2:tdTomato* and *hsp70:PCNA-GFP* were co-injected.	1	48	24
22	*trβ2:tdTomato* and *hsp70:PCNA-GFP* were co-injected.	1	42	28
23	*Connexin:55.5:rasGFP* and *hsp70:H2B-RFP* were co-injected.	8	42	31
24	*Connexin:55.5:rasGFP* and *hsp70:H2B-RFP* were co-injected.	9	42	30

REFERENCES

Almeida, A. D., Boije, H., Chow, R. W., He, J., Tham, J., Suzuki, S. C. and Harris, W. A. (2014). Spectrum of Fates: a new approach to the study of the developing zebrafish retina. *Development* **141**, 1971–1980.

Boije, H., Rulands, S., Dudczig, S., Simons, B. D. and Harris, W. A. (2015). The Independent Probabilistic Firing of Transcription Factors: A Paradigm for Clonal Variability in the Zebrafish Retina. *DEVCEL* **34**, 532–543.

Compagnon, J., Barone, V., Rajshekar, S., Kottmeier, R., Pranjic-Ferscha, K., Behrndt, M. and Heisenberg, C.-P. (2014). The Notochord Breaks Bilateral Symmetry by Controlling Cell Shapes in the Zebrafish Laterality Organ. *DEVCEL* **31**, 774–783.

Dzafic, E., Strzyz, P. J., Wilsch-Bräuninger, M. and Norden, C. (2015). Centriole Amplification in Zebrafish Affects Proliferation and Survival but Not Differentiation of Neural Progenitor Cells. *CellReports* **13**, 168–182.

Icha, J., Kunath, C., Rocha-Martins, M. and Norden, C. (2016). Independent modes of ganglion cell translocation ensure correct lamination of the zebrafish retina. *J Cell Biol* **215**, 259–275.

Matejčić, M., Salbreux, G. and Norden, C. (2018). A non-cell-autonomous actin redistribution enables isotropic retinal growth. *PLoS Biol* **16**, e2006018.

Pisharath, H. and Parsons, M. J. (2009). Nitroreductase-Mediated Cell Ablation in Transgenic Zebrafish Embryos. In *Zebrafish* (eds. Lieschke, G. J., Oates, A. C., and Kawakami, K., pp. 133–143. Totowa, NJ: Humana Press.

Strzyz, P. J., Lee, H. O., Sidhaye, J., Weber, I. P., Leung, L. C. and Norden, C. (2015). Interkinetic Nuclear Migration Is Centrosome Independent and Ensures Apical Cell Division to Maintain Tissue Integrity. *DEVCEL* **32**, 203–219.

Suzuki, S. C., Bleckert, A., Williams P.R. Takechi M, Kawamura S, and Wong R.O. (2013).Cone photoreceptor types in zebrafish are generated by symmetric terminal divisions of dedicated precursors.Proc Natl Acad Sci U S A Sep 10;110(37):15109-14. doi:10.1073/pnas

Swanhart, L. M., Takahashi, N., Jackson, R. L., Gibson, G. A., Watkins, S. C., Dawid, I. B. and Hukriede, N. A. (2010). Characterization of an lhx1a transgenic reporter in zebrafish. *Int J Dev Biol* **54**, 731–736.

Vitorino, M., Jusuf, P. R., Maurus, D., Kimura, Y., Higashijima, S.-I. and Harris, W. A. (2009). Vsx2 in the zebrafish retina: restricted lineages through derepression. *Neural Dev* **4**, 14.

Weber, I. P., Ramos, A. P., Strzyz, P. J., Leung, L. C., Young, S. and Norden, C. (2014). Mitotic position and morphology of committed precursor cells in the zebrafish retina adapt to architectural changes upon tissue maturation. *Cell Reports* **7**, 386–397.

Williams, P. R., Suzuki, S. C., Yoshimatsu, T., Lawrence, O. T., Waldron, S. J., Parsons, M. J., Nonet, M. L. and Wong, R. O. L. (2010). In vivo development of outer retinal synapses in the absence of glial contact. *Journal of Neuroscience* **30**, 11951–11961.

Zolessi, F. R., Poggi, L., Wilkinson, C. J., Chien, C.-B. and Harris, W. A. (2006). Polarization and orientation of retinal ganglion cells in vivo. *Neural Dev* **1**, 2.