

B

## Percentage of Sip1+ cells out of

 Pou4f2+ cells

F Photoreceptor layer width


Figure S1: Measurements of the percentage of ACs and RGCs expressing Sip1 and the width of the different retinal layers in control and Sip $1^{\text {loxp/loxp }} ; a C r e$ retinae.

The different repeats of the measurements of the percentage of Sip1+ cells out of Glyt1+ and Gad67+ cells (A) and Pou4f2+ cells (B) are presented in scatter plots (N=3). The different repeats of the measurement of the width of the OPL (C), IPL (D), INL (E), photoreceptors layer (F), GCL $(\mathrm{G})$ and the total retina $(\mathrm{H})$ in control and Sipl ${ }^{\text {loxp/loxp }}$;aCre retinae are presented in scatter plots $(\mathrm{N}=4)$. The median in the scatter plots is represented by a line passing through the data points
A Calbindin+ cells number in the retina

B Pax6+ cells number in the


Pkca+BP cells number in the retina
$\left.\begin{array}{c}F \\ 60 \\ 50 \\ 40 \\ 30 \\ 20 \\ 10 \\ 10 \\ 0\end{array}\right]$


G GS+P27+ cells number in the $\left.\begin{array}{c}60 \\ 50 \\ 40 \\ 30 \\ 20 \\ 10 \\ 0\end{array}\right] \stackrel{\square}{\square} \quad \begin{gathered} \\ \\ \end{gathered}$

Figure S2: Measurements of the number of the different INL cells types in control and Sip $1{ }^{\text {loxp/loxp }} ;$ aCre retinae.

The different repeats of the quantification of the number of Calbindin+ HCs (A), Pax6+ ACs (B), Gad67+ GABAergic ACs (C), Glyt1+ glycinergic ACs (D), Is11+ BPs (E), Pkca+ BPs (F) and GS+P27+ Muller glia (G) in control and Sip1 $1^{\text {loxp/loxp }} ;$ aCre retinae are presented in scatter plots ( $\mathrm{N}=3$ for $\mathrm{Pkc} \alpha, \mathrm{N}=4$ for the rest). The median in the scatter plots is represented by a line passing through the data points.


## Figure S3: ERG traces.

Representative tracings of a control (black) and a mutant (gray) animal to the 5 stimulus conditions presented in Fig. 2K, L.


Figure S4: Pkca and cleaved Caspase3 positive cells and the mRNA levels of Sip1, Foxn4, Neurod4 and Neurod1 in control and Sip1 ${ }^{\text {loxp/loxp }}$;aCre eyes

IIF was used for the detection of Pkca+ cells in P14 control (A) and
 (B,F), E18.5(C,G) and P0 (D,H) control and Sip1 ${ }^{\text {loxp/loxp }} ;$ aCre retinae. The number of cCasp3+ cells in a normalized field of $180 \mu \mathrm{M}$ was quantified in each of the stages (I). qPCR analysis was used in order to quantify the relative mRNA levels of Sip1, Foxn4, NeuroD4 and NeuroD1 in whole eyes of E15.5 control (N=3) and Sip1 ${ }^{\text {loxploxp }} ;$ aCre $(\mathrm{N}=5)$ embryos (J). Significant differences ( $\mathrm{P}<0.05$ ) are marked with an asterisk. Scalebars: 50 $\mu \mathrm{m}$ for A and E and $100 \mu \mathrm{~m}$ for B-D and F-H.


Number of IsI1+ BP number in
E


Figure S5: The number of Ptf1a+ and Pou4f2+ and Isl1+ cells in control and Sip $1^{\text {loxp/loxp }} ; a C r e$ retinae.

The different repeats of the quantification of the number of Ptf1a+ cells at E14.5 (A) and E18.5 (B), Pou4f2+ in the NBL (C) and GCL (D) at E16.5 and Is11+ cells at P7 (E) are presented in scatter plots ( $\mathrm{N}=3$ for all counts). The median in the scatter plots is represented by a line passing through the data points. The mean percentage of Isl1+ cells in Sipl ${ }^{\text {loxp/loxp }}$;aCre retinae at P7 and P14 in comparison to their respective control is also presented ( $\mathrm{N}=3$ for P 7 and $\mathrm{N}=4$ for P 14 ) $(\mathrm{F})$. Significant differences $(\mathrm{P}<0.05)$ are marked with an asterisk.


Figure S6: Birthdating of Isl1+ cells in control and Sip $1^{\text {loxp/loxp }}$;aCre retinae.

The different repeats of the quantification of the number of $\operatorname{BrdU}+A p 2 \beta+$ cells in P0 retinae that were injected with BrdU at E13.5 (A), E14.5 (B) and E16.5 (C) are presented in scatter plots ( $\mathrm{N}=3$ for E 13.5 and $\mathrm{E} 16.5, \mathrm{~N}=4$ for E 14.5 ). The median in the scatter plots is represented by a line passing through the data points


Figure S7: The birth dates of RGCs and cone photoreceptors are not delayed in the Sip $1^{\text {loxp/loxp }} ; a C r e$ retina.

For RGCs birthdating BrdU was injected to control and Sipl-mutant embryos at E13.5, E14.5 or E16.5. The retinas were harvested at P0 and IIF was used in order to detect BrdU+;Pou4f2+ cells. The mean number of these cells was then quantified in control and mutant retinae injected at the three time points ( $\mathrm{N}=3$ for E13.5 and E16.5, $\mathrm{N}=4$ for E14.5) (A). For photoreceptor birthdating BrdU was injected to control and Sip1mutant embryos at E14.5. The retinae were harvested at P0 and IIF was used in order to detect BrdU+ cells which are found in the presumptive location of photoreceptors $(\mathrm{N}=3)$ (B). The number of BrdU + Pou4f2+ cells in control and Sip1-mutant embryos in the different repeats at E13.5 (C), E14.5(D) and E16.5 (E) (N=3 for E13.5 and E16.5, N=4 for E14.5) and the number of BrdU+ photoreceptors in the different repeats at E14.5 $(\mathrm{N}=3)$ ( F$)$ are also presented as scatter plots. The median in the scatter plots is represented by a line passing through the data points.
Percentage of Brdu+ Ki67A out of all BrdU+ cells $\left.\begin{array}{r}30 \\ 25 \\ 20 \\ 15 \\ 10 \\ 5 \\ 0\end{array}\right] \quad \begin{aligned} & \text { Control }\end{aligned}$
Relative luciferase avtivity -
B $\left.\begin{array}{r}1.4 \\ 1.2 \\ 1 \\ 0.8 \\ 0.6 \\ 0.4 \\ 0.2 \\ 0\end{array}\right] \stackrel{\bullet}{ }$
D
$\left.\begin{array}{r}0.35 \\ 0.3 \\ 0.25 \\ 0.2 \\ 0.15 \\ 0.1 \\ - \\ 0.05 \\ 0\end{array}\right]$

E

Site 2

G


## Figure S8: Measurements of Cell cycle exit, Luciferase and chromatin IP analysis

The different repeats of the quantification of the percentage of BrdU+ Ki67- cells out of the BrdU+ cell population in control and Sip1 loxp/oxp $; a C r e$ retinae ( $\mathrm{N}=3$ ) (A). The different repeats of the quantification of the relative luciferase activity in the presence of Sip1 expression vector achieved by the 5 ' Ptf1a enhancer (B) and the 3 ' enhancer (C) $(\mathrm{N}=3)$ are presented in scatter plots. The repeats of the measurements of the percentage of input which was immune-precipitated by Sip1 AB or IgG in the vicinity of Sip1 binding site $1(\mathrm{D}), 2(\mathrm{E}), 3(\mathrm{~F})$ or $4(\mathrm{G})$ are also presented in scatter plots $(\mathrm{N}=4$ for sites 1,2,3 and $\mathrm{N}=3$ for site 4 ). The median in the scatter plots is represented by a line with the same color of the data points from which it is derived.

## Table S1

| Antigen | Source | Manufacturer | \# Catalog | Dilution |
| :--- | :--- | :--- | :--- | :--- |
| Ap2 $\alpha$ | Mouse | Santa Cruz | sc-12726 | $1: 50$ |
| Ap2 $\beta$ | Rabbit | Cell signaling | 2509 | $1: 50$ |
| BrdU | Mouse | Chemicon | MAB3222 | $1: 100$ |
| Calbindin | Mouse | Sigma | C9848 | $1: 5000$ |
| Cleaved <br> Caspase3 | Rabbit | Cell signaling | 9661 | $1: 300$ |
| Crx | Mouse | Abnova | \#H00001406- <br> M02 | $1: 400$ |
| Gad67 | Mouse | Millipore | MAB5406 | $1: 1000$ |
| Glutamin <br> Synthetase | Mouse | BD | 610517 | $1: 100$ |
| Glyt1 | Goat | Chemicon | AB1770 | $1: 4000$ |
| Igg | Rabbit | Santa Cruz | sc-2027 | $1: 100$ |
| Is11 | Mouse | DSHB | $40.2 D 6$ | $1: 100$ |
| Ki67 | Rat | Hako <br> bank | M7249 | $1: 10314897$ |
| Nf165 | Mouse | Therno <br> scientific | RB9019 | $1: 500$ |
| P27 | Rabbit | Convance | prb-278p | $1: 400$ |
| Pax6 | Rabbit | Santa Cruz | sc-208 | $1: 1200$ |
| Pkc $\alpha$ | Vector |  | $1: 200$ |  |
| PNA | Goat | Santa Cruz | sc-6026 | $1: 100$ |
| Pou4f2 | Rabbit | Acris | dp3501p | $1: 40$ |
| Prox1 | Rabbbit | Kind gift of Helena Edlund | $1: 500$ |  |
| Ptf1a | Santa Cruz | sc-48789 | $1: 200$ |  |
| Sip1 |  |  |  |  |

Table S2

| Sip1 forward | GGCAAGGCCTTCAAGTACAA |
| :--- | :--- |
| Sip1 reverse | AAGCGTTTCTTGCAGTTTGG |
| Foxn4 forward | AGCCACACCCCAAACACTAC |
| Foxn4 reverse | AAGCTGCCTGTTTTGCTGTT |
| Neurod4 forward | GCTCCAGTCAGATCACAGGAG |
| Neurod4 reverse | CAGCTCCACCATGTCCTTG |
| Neurod1 forward | AATTCGCCCACGCAGAAGGC |
| Neurod1 reverse | TGAGACACTCATCTGTCCAGC |
| Actb forward | CACAGCTGAGAGGGAAATCGTGC |
| Actb reverse | GATCTTGATCTTCATGGTGCTAGG |
| Ptf1a site 1 forward | TTCCCACGACAGGATGTGAG |
| Ptf1a site 1 reverse | TTTCTGGCAGGCCTGTCTCT |
| Ptf1a site 2 forward | ATCAATCCCTGTCTTGGTTCTCT |
| Ptf1a site 2 reverse | TTAGGCAGCTGCAGTCCA |
| Ptf1a site 3 forward | AGTGTTGGTCCCTAAAGACATTG |
| Ptf1a site 3 reverse | ACGTTCCCTTCTTGATAAATGG |
| Ptf1a site 4 forward | GCTCCATTTCAACCATTGTG |
| Ptf1a site 4 reverse | GAAGAGCCCTTGAGCTTGG |

