Supplementary methods

Microarray data analysis

Raw images were analyzed using ImaGene v5.6 software (BioDiscovery Inc., El Segundo, CA). Data filtering, normalization and statistical analysis was carried out by using MATLAB (Natick, MA).

miRNA expression criteria based on signal and background intensities

Expression judgment was based on three steps: (1) Determine detected probes in a hybridization; (2) Determine expressed miRNAs in a biological sample; (3) Determine whether miRNA was expressed at a developmental stage in all biological samples. At the first step, the Hy5 intensity was recovered after Lowess normalization. For each array a threshold was set based on background intensity distribution. The threshold was the mean background value plus $3.29 \times \sigma_{bg}$ (σ_{bg}: standard deviation of background intensities), corresponding to $p<5 \times 10^{-4}$ (one-tail). Probes with signal intensity greater than the threshold were treated as detected. At the second step, if more than 2/3 of probes from replicate measurements corresponding to a miRNA were beyond the intensity threshold, the miRNA was classified expressed in the biological sample. At the third step, only those miRNAs were considered as expressed at a development stage, that were classified expressed in all biological replicates.

Hierarchical cluster analysis

For hierarchical cluster analysis and visualization the software Cluster 3.0 and Java TreeView was used (http://bonsai.ims.u-tokyo.ac.jp/~mdehoon/software/cluster/software.htm). Data included in the analysis contained expressed miRNAs in brain and/or retina. Relative expression values for each miRNA, in all conditions were median centered. The complete linkage clustering method was applied with centered correlation similarity metric.

Self organizing map algorithm

Analysis was carried using GeneCluster 2.0 software (http://www.broadinstitute.org/cancer/software/genecluster2/gc2.html) on the pre-filtered microarray dataset containing only expressed miRNAs in retina. Relative expression values for each miRNA, in all conditions were median centered. Default parameters were applied with 500,000 iterations, random vector initialization and bubble neighborhood.

Predicted miRNA target genes

We collected mouse miRNA target genes predicted by TargetScan and miRanda (released Sep 2008), respectively. There is approximately 35% overlap between the two databases. Only targets predicted by both methods are adopted in our study.

Quantitative real time PCR

miRNA quantification from total retina with DNA primers

1ug of total RNA (containing the miRNA fraction) was tailed and reverse transcribed with the NCode miRNA First-Strand cDNA synthesis kit (Invitrogen). Forward primers corresponded to the mature miRNA sequences (Table S1). Geometric mean of expression of 5S ribosomal RNA, beta actin (*Actb*) and glyceraldehyde-3-phosphate dehydrogenase (*Gapdh*) were used for normalization. For significance analysis Student`s two tail t test was applied.

miRNA quantification from LCM samples and total retina with LNA primers

LNA primers for *miR-136*, *miR-143*, *miR-184*, *miR-204*, *miR-210* and *miR-211* were purchased from Exiqon. cDNA was generated from 4 independents sets of LCM samples (ONL, INL, GCL) and from C57BL/6 total retina from E18, P1, P5, P12 and adult stages using miRNA specific reverse transcription primers provided in the miRCURY LNA™ microRNA PCR system (Exiqon). Cycling and melting curve analysis was carried out as described previously.

Host gene mRNA quantification

cDNA generated with the NCode system was used as template for qPCR. Intron spanning primer pairs of the corresponding genes were designed with the Universal ProbeLibrary Assay Design Center (Table S2).