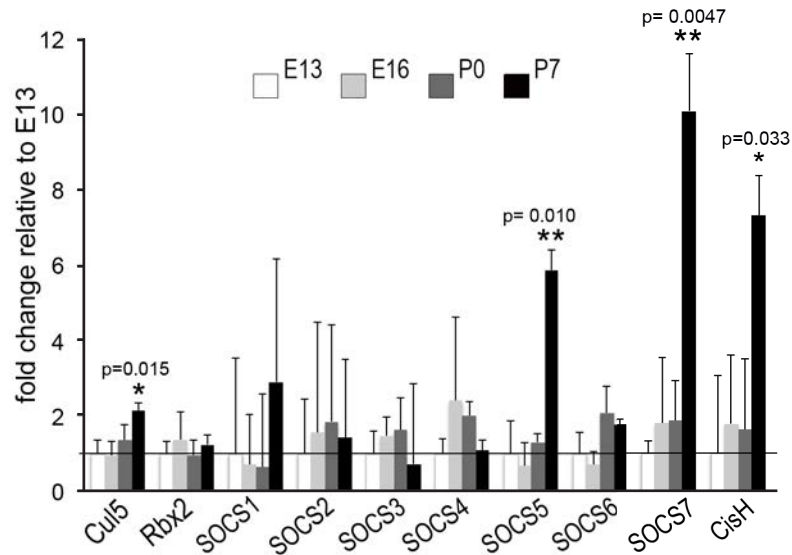
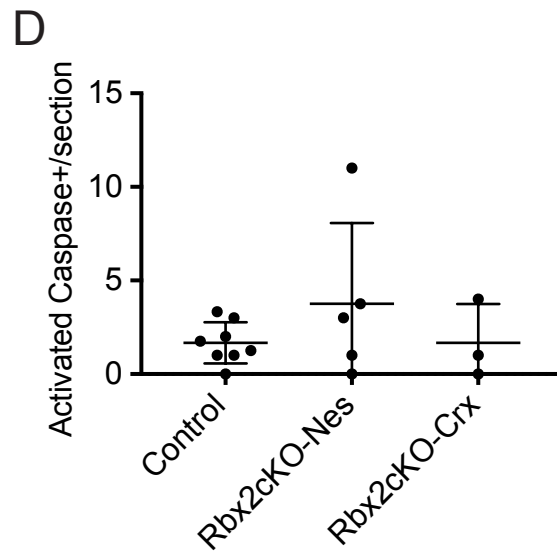
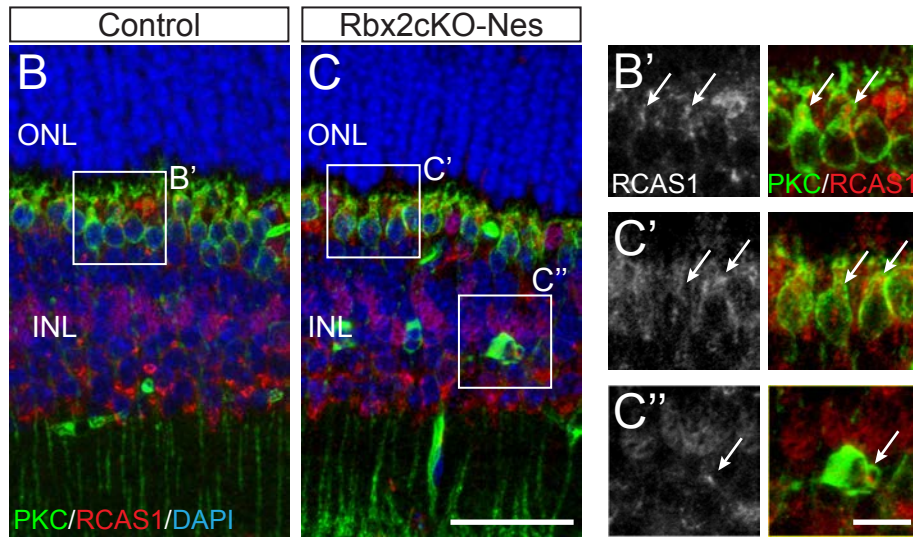
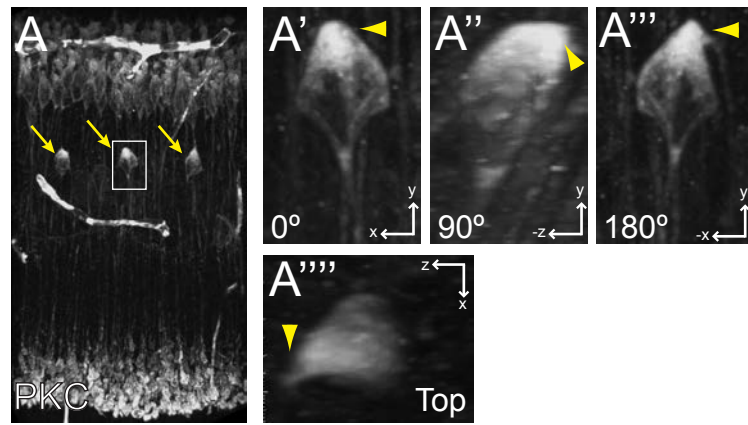


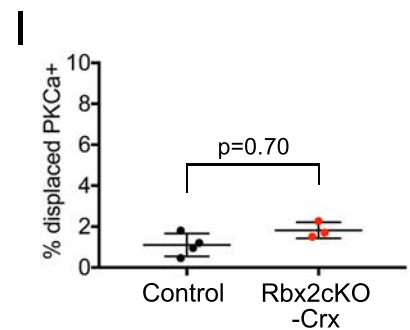
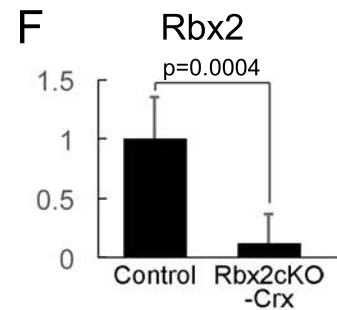
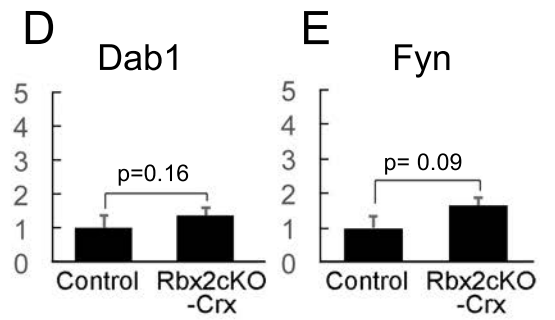
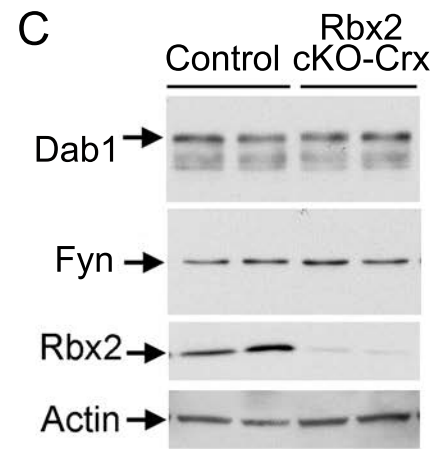
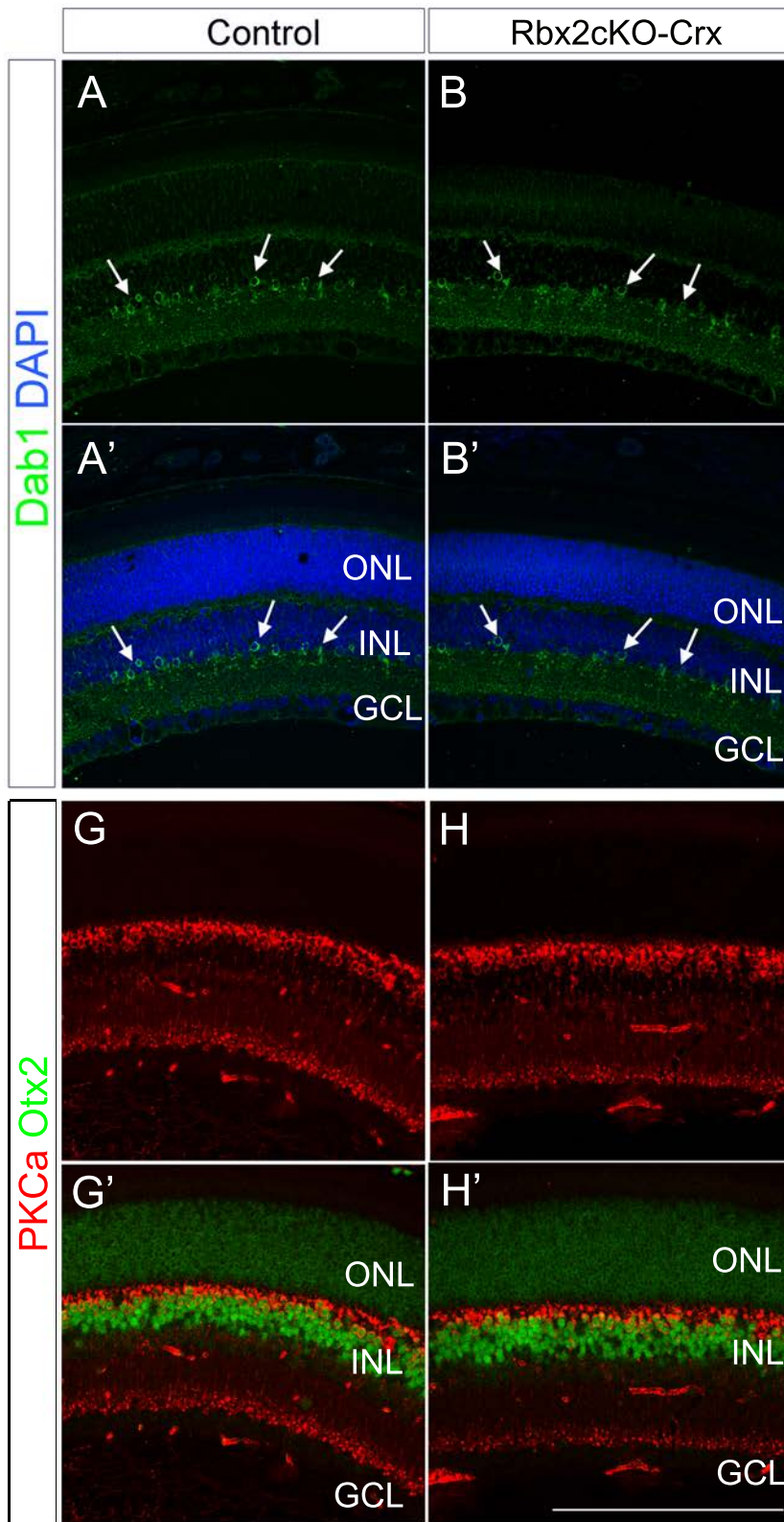
Supplementary Figures



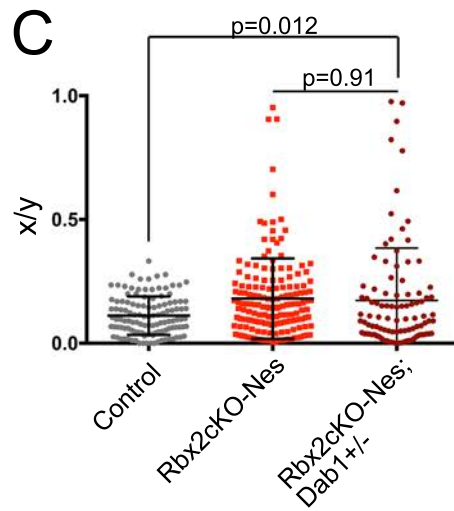
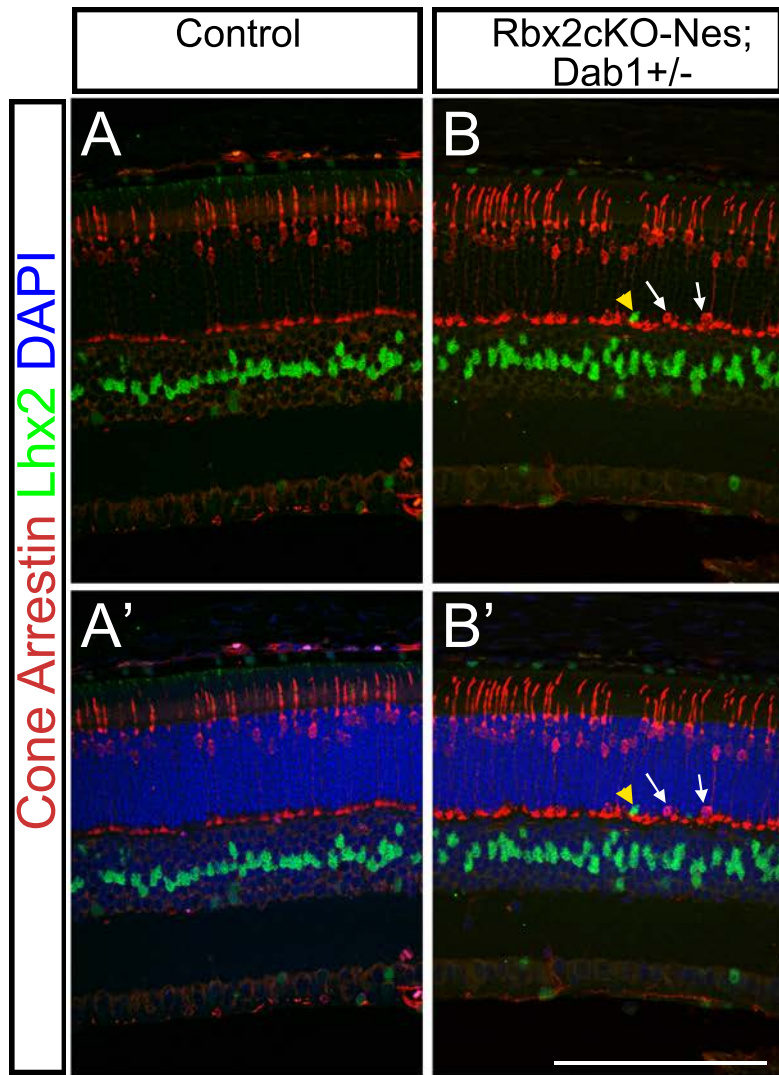
**Figure S1. CRL5 substrate adaptors are developmentally regulated in the retina.** RT-qPCR analyses represented as relative change (ddCT) between retinas collected at different stages of development and embryonic day 13 (n=3 for each stage). Data indicates mean  $\pm$  SD. P-values are obtained by one-way ANOVA with Tukey's post hoc test in each indicated sample (*Cul5*  $F(3,9)=5.514$ ; *SOCS7*  $F(3,10)=6.5$ ; *SOCS5*  $F(3,9)=5.8$ ; *CISH*  $F(3,10)=2.98$ ). P-values: \* $<0.05$ , \*\* $<0.01$ .



**Figure S2. Defects on dendrite formation and polarity in displaced rBCs without neurodegeneration.** (A) Representative image of P20 Rbx2cKO-Nes retina stained with PKCalpha+ depicting three displaced rBCs (yellow arrows). Image represents a maximum projection of 90.4 x 167.3 x 29µm (x/y/z). (A'-A''') High magnification view of rBC indicated by a white box in (A), highlighting front (A'), side (A''), back (A'''), and top (A''') view (A). Yellow arrowheads indicate where apical rBC dendrite should be. (B-C'') Immunofluorescence using antibodies to the golgi marker RCAS1 (red) and PKCalpha (green) on control (B-B') and Rbx2cKO-Nes (C-C'') retinas. Only displaced rBCs (C'') showed evident defects on cell polarity indicated by RCAS1 staining. (D) Quantification of apoptotic (activated-Caspase 3+ cells) of P20 control (n=8), Rbx2cKO-Nes (n=5), and Rbx2cKO-Crx (n=3) retinas showed that depletion of RBX2 did not increased cell death. P= 0.3761 by one-way ANOVA test. ONL: outer nuclear layer, and INL: inner nuclear layer. Scale bars: 30µm in A, B, and C; 7µm in A'-A''', and B'-C''.

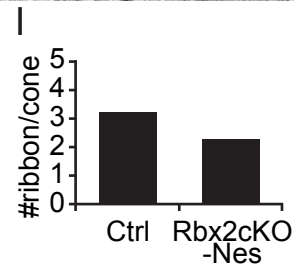
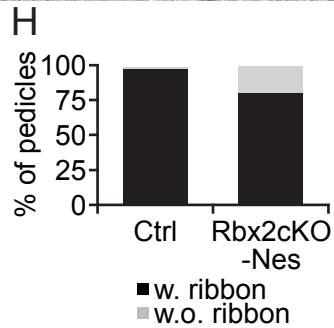
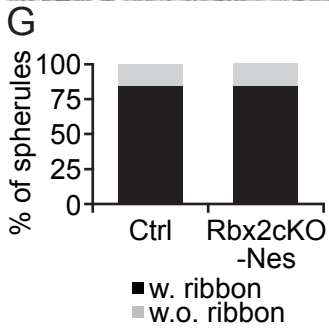
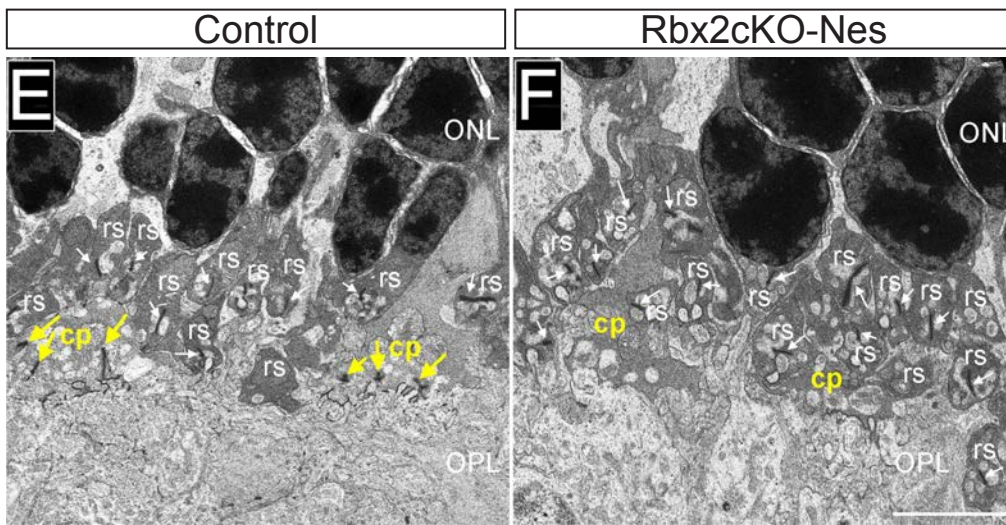
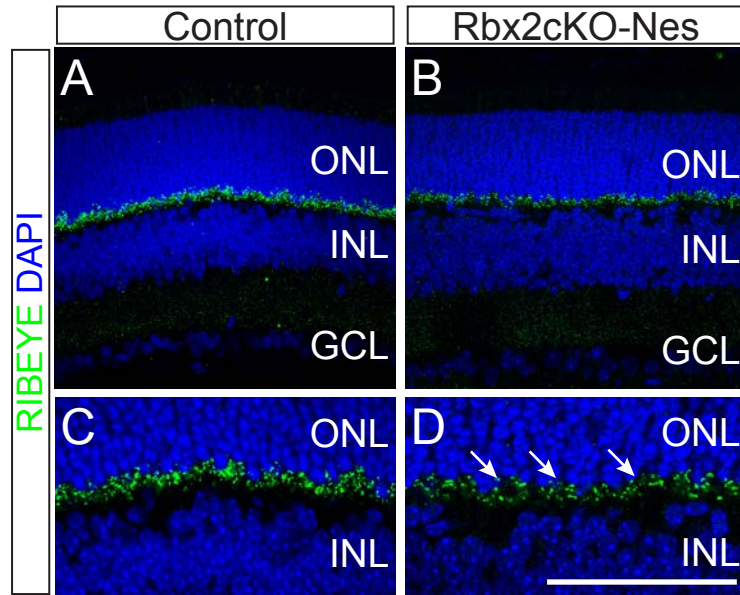


**Figure S3. Normal rBC layering in Rbx2cKO-Crx retinas.** (A-B') P20 retinas from control and Rbx2cKO-Crx littermates were immunostained with antibodies to DAB1 (green) and counterstained with DAPI (blue). No DAB1 accumulation was observed in Rbx2cKO-Crx retinas. (C) Western blot comparison of P20 control and Rbx2cKO-Crx retina lysates showed that whereas Rbx2 was significantly depleted in Rbx2cKO-Crx retinas, no accumulation of DAB1 or FYN was detected. (D-E) Densitometric analyses of control (n=6) and Rbx2cKO-Crx (n=7) retina lysates normalized to Actin. Data indicates mean $\pm$ SD. P-values were obtained by Student t-test. (G-H') Normal localization of rBCs in Rbx2cKO-Crx retinas. P20 control and Rbx2cKO-Crx retinas were stained using antibodies to PKCalpha (red) and OTX2 (green). (I) Quantifications of the percentage of PKCalpha+ OTX2+ cells localized in the vitreal half of the INL in control mice (n=3), Rbx2cKO-Nes (n=3). P-value was obtained by Student t-test. ONL: outer nuclear layer, INL: inner nuclear layer, and GCL: ganglion cell layer. Scale bars: 100 $\mu$ m in A-B', and G-H'.



**Figure S4. RBX2 depletion in photoreceptors and bipolar cells leads to cone photoreceptor and Muller glia misplacement.**

(A-B') P20 retinas of control and Rbx2cKO-Nes; *Dab1* +/- mice labelled with antibodies to Cone Arrestin (red) and LHX2 (green), and counterstained with DAPI (blue). Note the ectopic position of some cones in Rbx2cKO-Nes; *Dab1* +/- retinas (white arrows in B-B') as well as LHX2+ Muller glia (yellow arrowhead in B-B'). (C) Scatter plot of the position of all cone nuclei obtained from three high magnification images of different retinas (n=3 for each genotype; control=120, Rbx2cKO= 167, and Rbx2cKO-Nes; *Dab1* +/- = 99 Cone Arrestin+ cells were quantified). The position of each individual nucleus in the ONL was recorded as the ratio (x/y) of the distance from the nucleus to the OLM (x) and the thickness of the ONL in that region of the retina (y). Scale bar: 100µm





**Figure S5. RBX2 depletion seems to affect ribbon synapses in cone pedicles.** (A-D) P20 control and Rbx2cKO-Nes retinas stained with an antibody to RIBEYE and counterstained with DAPI. Less RIBEYE staining was observed in the outer plexiform layer in the absence of RBX2 (white arrows). (E, F) Representative electron microscopy images of control (E) and Rbx2cKO-Nes (F) retinas, highlighting cone pedicles (cp) and rod spherules (rs). Yellow and white arrows indicate ribbon synapses in cone pedicles and rod spherules, respectively. (G,H) Percentage of rod spherules (G) and cone pedicles (H) with or without the presence of synaptic ribbons. (I) For those cone pedicles containing ribbon synapses, no differences in the number of individual ribbon synapses within the pedicle were observed between control and Rbx2cKO-Nes. 56 control and 51 Rbx2cKO-Nes cone pedicles, and 416 control and 494 Rbx2cKO-Nes rod spherules were analysed from P15 control and Rbx2cKO-Nes retinas (n=1/condition). Data indicates mean $\pm$ SD. ONL: outer nuclear layer, INL: inner nuclear layer, and GCL: ganglion cell layer. Scale bars: 100 $\mu$ m in A-B, 50 $\mu$ m in C-D and 5 $\mu$ m in E and F.

**Table S1. Authentication of antibodies used in the present manuscript.**

Authentication methods used: 1: Antibody identification at <http://antibodyregistry.org>; 2: Genetic validation method; 3: Comparison with known published patterns in the literature; 4: Co-labeling of tagged protein with endogenous protein.

Antigen	Source	Host	Catalog (clone) #	Application	Authentication	Antibody ID	References (PMID)
Dab1 *	EMD Millipore	Rabbit	AB5840	WB, IF	1, 2, 3, 4	AB_11213946	17974915 20410119 24210661
Dab1	Rockland	Rabbit	100-401-225	WB	1, 2, 3, 4	AB_2245755	This paper
Rbx2	Santa Cruz	Rabbit	SC-166554 (G-8)	WB	1, 2	AB_2245755	24210661
Fyn *	Santa Cruz	Rabbit	SC-16 (FYN3)	WB	1, 2	AB_631528	26777117 28840468
Actin *	Santa Cruz	Goat	SC-1616 (I-19)	WB	1, 3	AB_630836	23541735 25283985 27704563 27874237
P-Tyr	EMD Millipore	Mouse	05-321 (4G10)	WB	1, 2	AB_309678	26334723 25814554 25915123
Dab1 *	Santa Cruz	Goat	SC-7827 (E-19)	IP	1, 2, 4	AB_638972	17974915 24210661
Dab1	Sigma-Aldrich	Rabbit	HPA052033	IF	1, 2, 3	AB_2681703	This paper
Cone Arrestin	Millipore	Rabbit	Ab15282	IF	1, 3	AB_11210270	26453550 25798616 24651551
Alpha-PKC	Sigma-Aldrich	Mouse	P5704	IF	1, 3	AB_477375	20127818 18671302
S-Opsin *	Santa Cruz	Goat	sc-14363	IF	1, 3	AB_2158332	26587737 28965762
Lhx2 *	Santa Cruz	Goat	sc-19344	IF	1, 3	AB_2135660	28089909
Glutamine Synthetase	EMD Millipore	Mouse	MAB302 (GS-6)	IF			26283925 26527153 25907681
Otx2	R&D Systems	Goat	AF1979	IF	1, 3	AB_2157172	27097562
GFAP	NeuroMab	Mouse	73-240 (N206/8)	IF	1, 3	AB_10672298	28111074
RIBEYE	Synaptic Systems	Rabbit	192103	IF	1, 3	AB_2086775	25232303 21179232
RCAS1	Cell Signaling	Rabbit	12290T	IF	3		28687497 26887977
Activated Caspase-3	Cell Signaling	Rabbit	9664S	IF	3		28767729 24210661

\*: Discontinued; WB: Western blot; IF: Immunofluorescence; IP: Immunoprecipitation