Supporting Information

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SI Materials and Methods

List of Antibodies. Horseradish peroxidase (HRP)-associated secondary antibodies for Western blot analysis were stabilized goat anti-mouse IgG (H+L), peroxidase conjugated (#32430; Thermo Scientific) and stabilized goat anti-rabbit IgG (H+L), peroxidase conjugated (#32460; Thermo Scientific). The following primary antibodies were used: polyclonal ChIPAb+ REST (#17–641; Millipore), polyclonal anti-GAPDH (#SAB3500247; Sigma-Aldrich), monoclonal anti-His-tag (HIS.H8, #sc-57598; Santa Cruz Biotechnology), rabbit anti-GFP (#A-11122; Life Technologies), polyclonal anti-mSin3A (K-20) (#sc-994; Santa Cruz Biotechnology), and polyclonal anti-BDNF (H-117) (#sc-20981; Santa Cruz Biotechnology).

Protein Extraction. Total protein lysates were obtained from cells lysed on ice for 20 min in RIPA buffer [20 mM Tris·HCl, pH 7.4; 1% Triton X-100; 10% (vol/vol) glycerol; 150 mM NaCl; 1% PMSF; protease inhibitor mixture: Complete protease inhibitor mixture tablets; 04693116001; Roche Applied Science]. The final protein concentration was quantified by using the BCA protein assay kit (#23225; Pierce Biotech.). Nuclear extracts were prepared as follows: the pellet from 5×10^6 cells was resuspended in 400 μL cytoplasmic buffer (10 mM Hepes, pH 7.9, 1.5 mM $MgCl_2$, 10 mM KCl, 0.5 mM DTT, protease inhibitor mixture) and incubated 20 min on ice with constant shaking. After the addition of 25 μL of 10% (vol/vol) IGEPAL, the solution was briefly vortexed and centrifuged 2 min at 8,000 × *g* at 4 °C. The supernatant containing the cytoplasmic extracts was collected, the pelleted nuclei were suspended in 50 μL nuclear buffer [20 mM Hepes, pH 7.9, 1.5 mM $MgCl_2$, 420 mM NaCl, 0.2 mM EDTA, 25% (vol/vol) glycerol, protease inhibitor mixture] and incubated 20 min on ice with constant shaking. Nuclei were centrifuged 25 min at 10,000 × *g* at 4 °C, and the supernatant (nuclear extract) was collected. The final protein concentration was quantified by using the Bradford protein assay (#23200; Pierce Biotech.).

Western Blotting. Proteins were separated using precast 10% NuPAGE Novex Bis-Tris Gels (Life Technologies) and transferred to a nitrocellulose membrane. After incubation with primary antibodies, membranes were incubated with HRP-conjugated secondary antibodies and revealed by autoradiography using the SuperSignal West Pico Chemiluminescent Substrate (#34077; Thermo Scientific).

List of Plasmids and Cloning Strategies. Expression vectors encoding for REST and shREST were a kind gift from J. Meldolesi, Fondazione San Raffaele, Milano, Italy; pCS2+MTmSin3AN205 was a kind gift from T. Kouzarides, Gurdon Institute, Cambridge, UK; and pCS2+PRIKLE1 was a kind gift from A. Bassuk, University of Iowa, Ames, IA. pcDNA3.1His-*As*LOV2 was cloned as follows: the *As*LOV2 sequence was amplified from a bacterial plasmid encoding for *As*LOV2 (gift from T. R. Sosnick, University of Chicago, Chicago) and inserted in the pcDNA3.1V5/His vector (Life Technologies) between the HindIII and AgeI sites. pcDNA3.1His/*As*LOV2-PAH1 and pcDNA3.1His/*As*LOV2-RILPshort were obtained as follows: the PAH1 sequence was amplified from pCS2+MTmSin3AN205 and cloned in pcDNA3.1His-*As*LOV2 at the AgeI site; the RILP N313 sequence was amplified from pCS2+PRIKLE1 and cloned in pcDNA3.1-His-*As*LOV2 at the AgeI site. pGFP-REST was cloned as follows: mouse REST cDNA was amplified with modified primers and inserted in pPAGFP-C1 between the EcoRI and BamHI sites. pGL3-RE1/NRSE was a kind gift from K. Nakashima, Nara Institute of Science and Technology, Nara, Japan. All constructs were verified by DNA sequencing.

Lentivirus Production and Infection Procedures. The lentiviral pCCL.sin.cPPT.PGK.GFP.WPRE bidirectional expression vector and packaging plasmids were a kind gift from L. Naldini, TIGET, San Raffaele Sci. Institute, Milan, Italy. For the coordinated expression of AsLOV2-PAH1b and GFP or of scrambled/REST shRNA (described in ref. 9) and mCherry, the low-affinity nerve growth factor (NGF) receptor in the MA1 construct (49) was replaced with REST cDNA, leading to GFP expression from the miniCMV promoter and REST expression from the phosphoglycerate kinase (PGK) promoter. Third-generation lentiviruses were produced by transient four-plasmid cotransfection into HEK293T cells using the calcium phosphate transfection method. Supernatants were collected, passed through a 0.45-μm filter, and purified by ultracentrifugation as previously described. Viral vectors were titrated at concentrations ranging from 1×10^8 to 5×10^9 transducing units (TU)/mL and used at a multiplicity of infection (MOI) of 1–10. The efficiency of infection was estimated to range between 70% and 90% by counting neurons expressing GFP protein with respect to the total number of cells stained with DAPI. Primary cortical neurons were infected at 7 DIV. After 24 h, half of the medium was replaced with fresh medium. Experiments were performed 5–7 d after infection (between 12 and 14 DIV).

Electrophysiological Recordings. Patch electrodes, fabricated from thick borosilicate glasses, were pulled to a final resistance of 4–6 MΩ. Recordings with leak current <100 pA and series resistance >20 MΩ were discarded. All recordings were acquired at 50 kHz. Neuron excitability was performed in Tyrode extracellular solution in which D-(−)-2-amino-5-phosphonopentanoic acid (D-AP5; 50 μM), 6-cyano-7 nitroquinoxaline-2,3-dione (CNQX; 10 μM), bicuculline methiodide (30 μM), and (2*S*)-3-[[(1*S*)-1-(3,4-dichlorophenyl)ethyl]amino-2-hydroxypropyl](phenylmethyl) phosphinic acid hydrochloride (CGP; 5 μM) were added to block NMDA, non-NMDA, $GABA_A$, and $GABA_B$ receptors, respectively. The internal solution (K-gluconate) was composed of (in mM) 126 K gluconate, 4 NaCl, 1 $MgSO_4$, 0.02 $CaCl_2$, 0.1 BAPTA, 15 glucose, 5 Hepes, 3 ATP, and 0.1 GTP, pH 7.3. In voltage-clamp experiments, sodium currents were dissected using an extracellular solution containing 130 mM NaCl, 2 mM $CaCl_2$, 2 mM $MgSO_4$, 10 mM glucose, 5 mM tetraethylammonium chloride (TEA-Cl), and 10 mM Hepes/NaOH (pH 7.4). The patch pipette was filled with 130 mM $CsCl_2$, 10 mM NaCl, 20 mM TEA-Cl, 10 mM EGTA, 2 mM $MgCl_2$, 4 mM Mg-ATP, and 10 mM Hepes/CsOH (pH 7.4). All of the parameters were analyzed using the pClamp (Molecular Devices) and Prism (GraphPad Software) software.

Computational Methods. A proper size for the simulation box corresponding to a pressure of 1 atm and temperature of 300 K was determined in the constant pressure and temperature (NPT) ensemble using the Nosé-Hoover Langevin piston method for 500 ps (50). The simulation ensemble was then switched to constant volume and temperature (NVT) for the rest of the simulation by keeping the temperature stationary around 300 K using Langevin dynamics. Two simulation runs were performed, one for the full chimera and one for the truncated linker, each lasting 70 ns. We used the MD program nanoscale molecular dynamics (NAMD), the chemistry at Harvard macromolecular mechanics (CHARMM27) force field with the cross-term energy map (CMAP) correction for

protein backbone energetics (51), and transferable intermolecular potential, three-point (TIP3P) model for water molecules. Parameters for the flavin mononucleotide (FMN) chromophore were taken from ref. 52. Periodic boundary conditions were used to replicate the system and remove box surface effects. Short-range nonbonded interactions were cut off at 12 Å, whereas long-range electrostatic interactions were computed using the particle mesh Ewald method. Chemical bonds connecting hydrogen atoms to heavy atoms were kept fixed using the SHAKE algorithm (53). An integration time step of 1 fs was used to ensure stability of the dynamical algorithm.

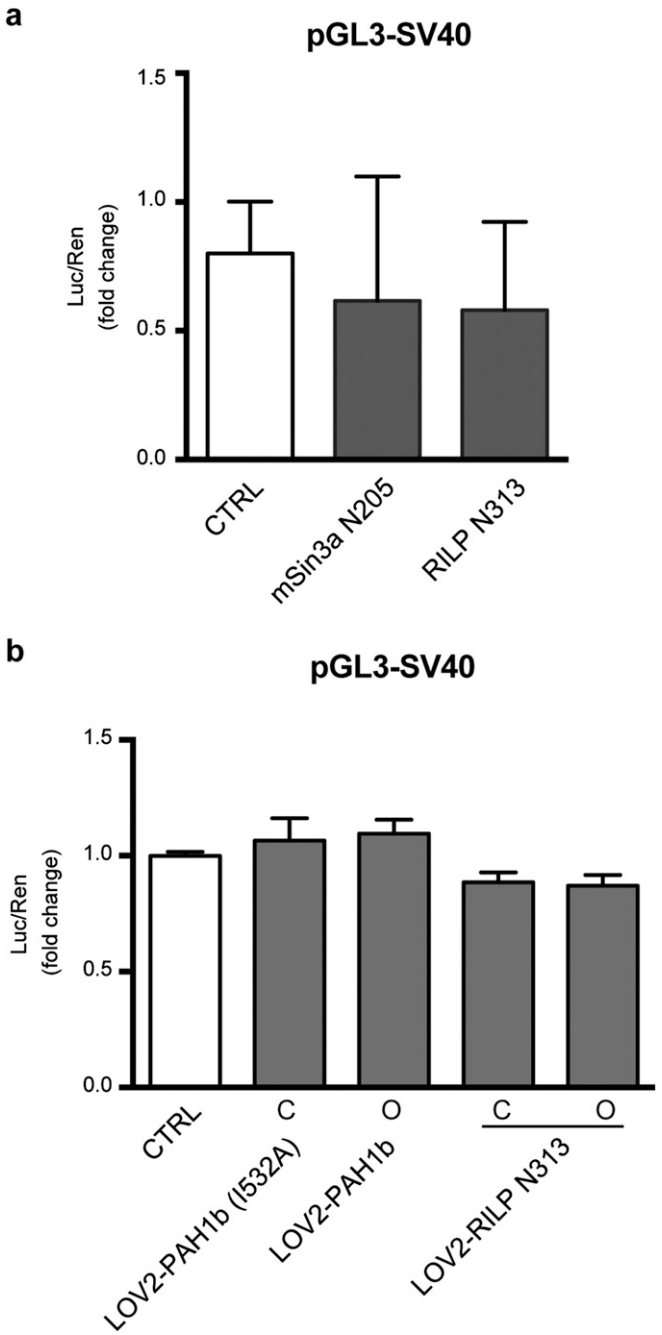

Fig. S1. (*A*) HeLa cells were transfected with the pGL3-SV40 reporter vector in the absence (white bars) or presence (gray bars) of expression plasmids encoding for mSin3a N205 or RILP N313. (*B*) HeLa cells were transfected with the pGL3-SV40 reporter vector in the absence (white bars) or presence (gray bars) of expression plasmids encoding for the closed (C) or open (O) forms of AsLOV2-PAH1b or RILP N313. Control samples were cotransfected with the empty vector corresponding to the effector plasmids. Luciferase activity was measured 48 h after transfection. Data were first normalized to the activity of the cotransfected TK-Renilla reporter vector and subsequently to the activity of the reporter gene alone, set to 1 (one-way ANOVA followed by the Tukey's multiple comparison test; $n = 3$ independent experiments).

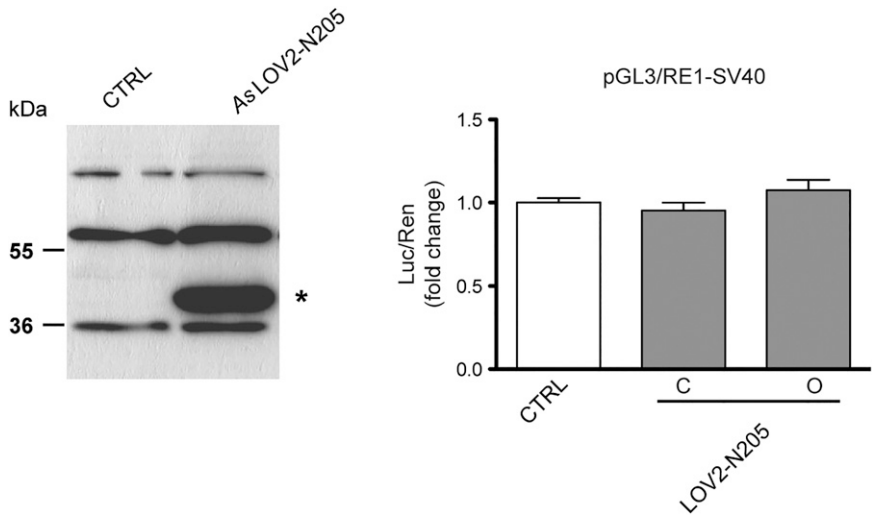

Fig. S2. (*Left*) N2a cells were transfected with a control plasmid (CTRL) or an effector vector encoding for AsLOV2-N205, as indicated. The expression of the construct (asterisk) in total cell lysate was analyzed by Western blot by using an anti-histidine tag antibody. (*Right*) HeLa cells were transfected with the pGL3-RE1/SV40 reporter vector in the absence (white bars) or presence (gray bars) of expression plasmids encoding for closed (C) or open (O) AsLOV2-N205 constructs, as indicated. Control samples were cotransfected with the empty vector corresponding to the effector plasmids. Luciferase activity was measured 48 h after transfection. Data were first normalized to the activity of the cotransfected TK-Renilla reporter vector and subsequently to the activity of the reporter gene alone, set to 1. Luc/Ren, luciferase/Renilla ratio (one-way ANOVA followed by the Tukey's multiple comparison test; $n = 3$ independent experiments).

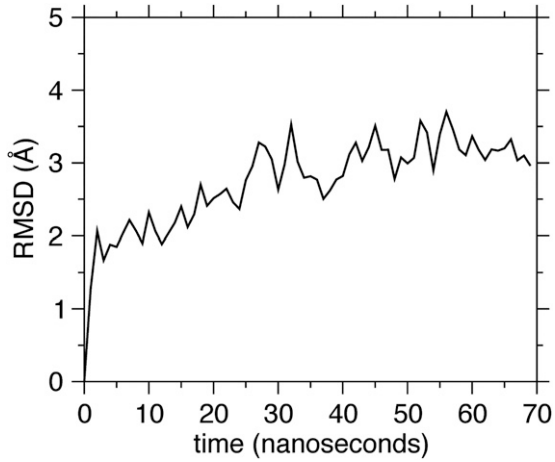

Fig. S3. Conformational stability of the dark state structure of the AsLOV2-PAH1 chimera after deletion of three residues at the linker region, calculated along an MD simulation as RMSD from the starting structure.

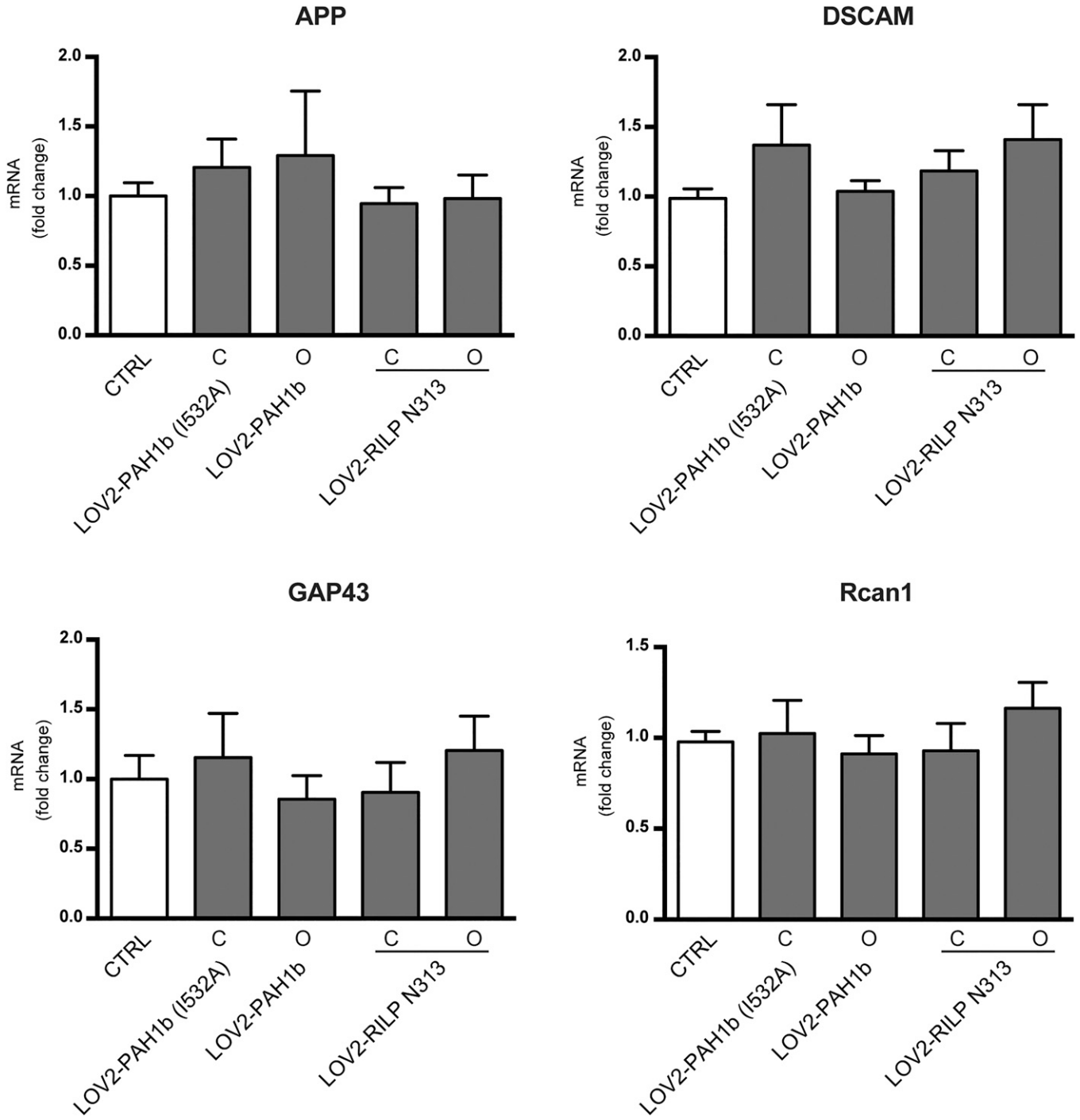

Fig. S4. Undifferentiated N2a cells were transfected with plasmids encoding for the closed (C) or open (O) variants of the LOV2-PAH1b or LOV2-RILPN313 chimeras, as indicated. The endogenous mRNA levels of the reported genes were quantified by qRT-PCR. *GAPDH* and *HPRT1* were used as control housekeeping genes (one-way ANOVA followed by the Tukey's multiple comparison test; $n = 3$ independent experiments).

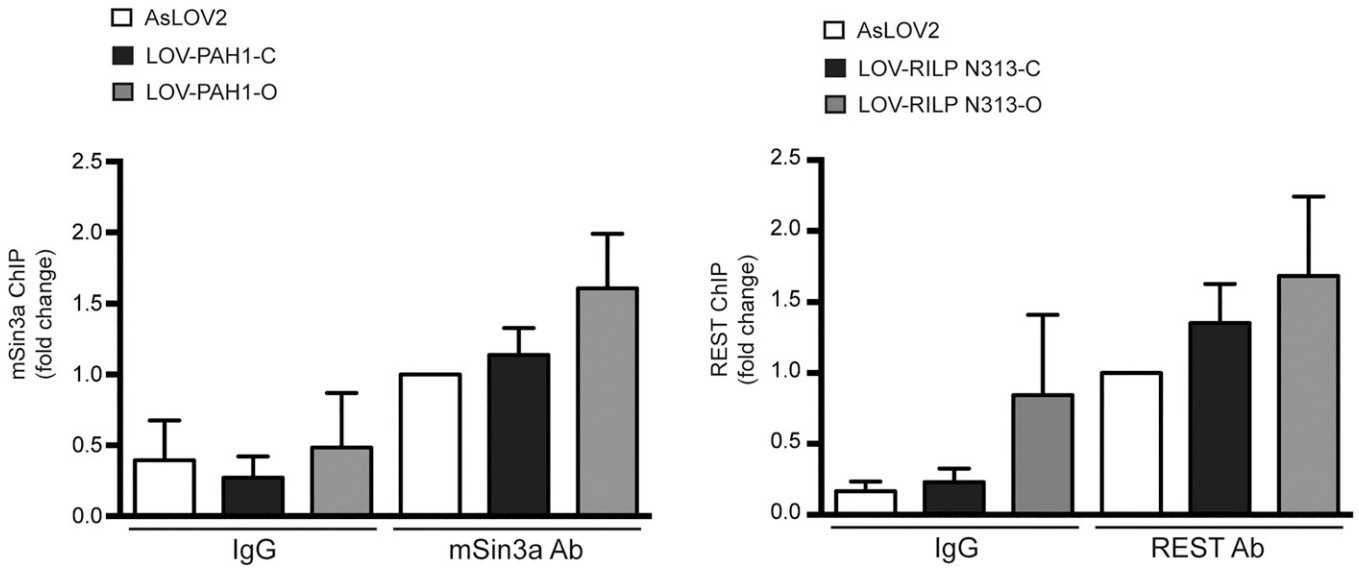

Fig. S5. N2a cells were transfected with the indicated expression vectors and then subjected to chromatin immunoprecipitation of the GAPDH promoter regions using 2 μg anti-mSin3a, anti-REST, or anti-rabbit IgG antibodies, as indicated. mSin3a or REST binding was normalized to the input DNA value and subsequently normalized to the binding in the control (*As*LOV2) sample, set to 1 (one-way ANOVA followed by the Tukey's multiple comparison test; $n = 3$ independent experiments).

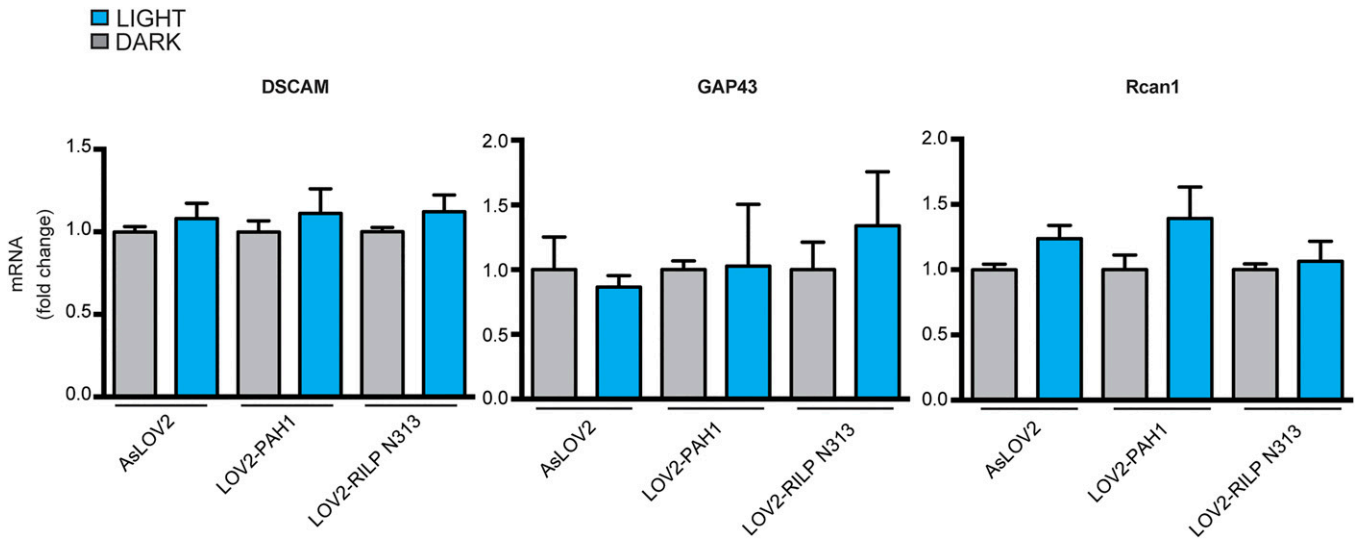

Fig. S6. Undifferentiated N2a cells were transfected with the indicated constructs and then subjected to 470-nm illumination (0.5 Hz) or kept in the dark, as indicated. After 48 h, the mRNA levels of the indicated genes were quantified via qRT-PCR. *GAPDH* and *HPRT1* were used as control housekeeping genes (one-way ANOVA followed by the Tukey's multiple comparison test; $n = 3$ independent experiments).

Table S1. NanoString nCounter code set design

Gene name	Accession no.	REST-target	Position	Target sequence
REST-target genes				
Stat1	NM_009283.3	YES	1591–1690	ACGCTGGGAACAGAACTAATGAGGGGCCTCTCATTGTCACCGAAGAACTTCACTC-TCTTAGCTTTGAAACCCAGTTGTGCCAGCCAGGCTTGGTGATTGA
Nfasc	NM_001160316.1	YES	4936–5035	CTCTTCGTATGCCTTATACAGCTCGGATCTAACCCTGCGGTTCCCAGATCCCTAG-CCCTATTCTGCAAGTCTTGATGCCTCCAGTGATGTGTGTCTCGGG
Nrxn1	NM_177284.2	YES	721–820	AGATGCCCTCACTTAAACATCATCGGATGCAAATGGATCAGTGATCGCTCTTGAG-CCTCGGTGGCCCTCTTTTTCAGAACGTTGCCTCCAAAGTGTATCC
Grin2b	NM_008171.3	YES	6341–6440	GGGAAAGCTCTTCGTATAAGGCTTTGTGAAAGAGCCATTACAGTAGGGTGAGAGA-GGGGGATGTTTTTAGTCATTAACGGTAGGGTTAGTGAGAAAGGGG
Cacna1h	NM_021415.4	YES	3213–3312	ATCGTCACTGTGTTTCAGATCTTGACACAGGAAGACTGGAATGTGGTTCTTTACA-ACGGCATGGCTTCCACCTCGTCCTGGGCTGCCCTTTACTTTGTGG
Syp	NM_009305.2	YES	733–832	TGTTTGGCTTCCTGAACCTGGTGCTCTGGGTTGGCAACCTATGGTTCGTGTTCAA-GGAGACAGGCTGGGCCGCCCCATTCATGCGCGCACCTCCAGGCGC
Grin1	NM_008169.2	YES	493–592	ACAGATGGCCCTGTCAGTGTGTGAGGACCTCATCTCTAGCCAGGTCTACGCTATC-CTAGTTAGTCACCCGCCTACTCCCAACGACCACTTCACTCCCACC
Cacna1i	NM_001044308.2	YES	9301–9400	TGCCCTGGCTTATTCCTGTCTTCGGCTCTGGTCCTGGCTTTCCCTCAGAGGAGGA-TGAACGAATCATGAGCAGTACTTACCTGTCTGCTTGTCCCTAATA
Calb1	NM_009788.4	YES	344–443	ATGGAAAAATAGGAATTGTAGAGTTGGCTCACGTCTTACCCACAGAAGAGAATTT-CTTGCTGCTCTTTCGATGCCAGCAACTGAAGTCCTGCGAGGAATT
Aplp1	NM_007467.3	YES	2301–2400	CTGGGTCCCAGGTATGTATGTCACTCCCTGGAATTCACCATCCCACGTTTCTTCA-CTAACATCCCAATAAAGTCCTCTTTCCCACCCGGCCAAAAAAAAA
Syt4	NM_009308.3	YES	1121–1220	TCTGGTCTCTCTCTGTTATCAGTCCACTACAAACACGCTCACTGTGGTGGTCTTA-AAAGCGCGGCACCTACCGAAATCTGATGTGTCTGGACTTTCAGAT
L1cam	NM_008478.3	YES	3561–3660	TGAAGACTAATGGAACTGGCCCTGTGCGAGTTTCTACTACAGGTAGCTTTGCCTC-CGAGGGCTGGTTCATCGCCTTTGTCAGCGCTATCATTCTCTTGCT
Sst	NM_009215.1	YES	47–146	CCTGCGACTAGACTGACCCACCGCGCTCCAGCTTGGCTGCCTGAGGCAAGGAAGA-TGCTGTCCTGCCGTCTCCAGTGCGCCCTGGCTGCGCTCTGCATCG
Neurod1	NM_010894.2	YES	565–664	CCTGCGCTCAGGCAAAAGCCCTGATCTGGTCTCCTTCGTACAGACGCTCTGCAAA-GGTTTGTCCCAGCCCACTACCAATTTGGTCGCCGGCTGCCTGCAG
Bdnf	NM_007540.4	YES	3261–3360	AGTCCCGTCTGTACTTTACCCTTTGGGGTTAGAAGTCAAGTTGGAAGCCTGAATG-AATGGACCCAATGAGAACTAGTGTTAAGCCCATTTCCCTAGTCAG
Penk	NM_001002927.2	YES	1011–1110	CGCTTTGCTGAGTCTCTGCCCTCCGATGAAGAAGGCGAAAATTACTCGAAAGAAG-TTCCTGAGATAGAGAAAAGATACGGGGGCTTTATGCGGTTCTGAA
Glra3	NM_080438.2	YES	757–856	GGACTTGCGGTACTGCACTAAACACTACAATACAGGAAAGTTTACATGCATAGAA-GTGCGATTCCATCTTGAGCGTCAGATGGGCTATTACTTGATCCAG
Gabrg2	NM_177408.5	YES	1614–1713	TTAGAACAAGAGCTGTTACACTGAGCAAGATACCTTTGAGCAACAGCAATGAAAA-CAGTGGAAGCTGGGAGGGTTTAAAGTGGCATTATCAGTCTTTGAC
Oprm1	NM_001039652.1	YES	1197–1296	GATTCCAGAAACCACTTTCCAGACTGTTTCCTGGCACTTCTGCATTGCCTTGGGT-TACACAAACAGCTGCCTGAACCCAGTTCTTTATGCGTTCCTGGAT
Cacng2	NM_007583.2	YES	645–744	GACGCGGACTACGAAGCTGACACCGCAGAGTATTTCCTCCGGGCCGTGAGGGCCT-CGAGTATCTTCCCGATCCTGAGTGTGATCCTGCTTTTCATGGGTG
Nrxn3	NM_001198587.1	YES	6111–6210	CTCAAAATCTACCATGGCATTCACTCCATGTAGCAGGTTGTGGTGTCTCTAGAAC-CAATTGTTAGTTTCCTAATGCTTTGGTGAACCCATGTGGGATGAC
Non–REST-target and housekeeping genes				
Map2k2	NM_023138.4	NO	2268–2367	CACATTCCTAAATACTAGGAAGGCTGAGTCGGGAAAACGACAGGTTTTGGGCCAC-TGTGGGCTACCTAGTGAGAATGTCTTACATCATGGAAATGGTGCA
Gtf3c1	NM_207239.1	NO	4587–4686	ACCAACGGCATGCTAGACCAGCCTGATCATTTTTCTTTCAAGGACCTGGATAGCA-GTGACCCTTCAAATGACCTGGTGGCATTTTCTTTGGACAGCCCTG
Tuba1a	NM_011653.2	NO	1379–1478	GGGAGGAAGAAGGAGAGGAATACTAAATTAAATGTCACAAGGTGCTGCTTCCACA-GGGATGTTTATTGTGTTCCAACACAGAAAGTTGTGGTCTGATCAG
Camkk2	NM_145358.2	NO	1321–1420	AAAGGCGTTGGATGTTTGGGCCATGGGTGTGACGCTGTACTGCTTTGTCTTTGGC-CAGTGCCCTTTCATGGATGAACGAATCATGTGTTTGCACAGTAAG
Syt17	NM_138649.1	NO	1391–1490	AACTGGAAAACGCCAGCCTAGTATTCACAGTGTTCGGCCACAACATGAAAAGCAG-CAATGACTTCATCGGGAGGATCGTCATCGGCCAGTATTCCTCCGG
Ubc	NM_019639.4	NO	22–121	GCGGAGTCGCCCGAGGTCACAGCCCTGCCCTCCCACACAAAGCCCCTCAATCTCT-GGACGCCACCGTGAAACAACTCCGTGAGAGAGACGATGCAGATCT
Gpi1	NM_008155.3	NO	2676–2775	CAGGGATGAACTCGATTATACCCTTCAAAAGACAGTTTTAGCAGGGTGTCATTAC-TGTCACATTCATATCCTGGCTATCAGTGAGTCCTTGTGAGACTGA
Map3k5	NM_008580.4	NO	641–740	CATCATCCTCTACTGCGATACTAATTCCGATTCACTCCAGTCCCTGAAGGAAATT-ATTTGCCAGAAGAATACTGTGTGCACCGGGAACTACACCTTCATC

Table S1. Cont.

Gene name	Accession no.	REST-target	Position	Target sequence
Rcan1	NM_001081549.1	NO	1061–1160	ACACAAGGACACTGGGGACATCCTGAGAAAACTGATAGTTCTTGTAATTGCTCATTTCTAGGTTCTGTTTTTGGCAAGGACAGGTTGACTGGTGGCCCAG
Tbx21	NM_019507.1	NO	626–725	CACTAAGCAAGGACGGCGAATGTTCCCATTCCTGTCCTTCACCGTGGCTGGGCTGGAGCCCACAAGCCATTACAGGATGTTTGTGGATGTGGTCTTGGTG
Tdo2	NM_019911.2	NO	496–595	AGAGTCTACAGTTCCGGCTGCTGGAAAATAAGATTGGTGTTCTTCAGAGCTTGAGAGTCCCTTACAACAGGAAACACTATCGTGATAACTTTGGAGGAGA
Gapdh	NM_001001303.1	HK	891–990	AGGTTGTCTCCTGCGACTTCAACAGCAACTCCCACTCTTCCACCTTCGATGCCGGGGCTGGCATTGCTCTCAATGACAACTTTGTCAAGCTCATTTCCTG
Hdac3	NM_010411.2	HK	1026–1125	GGCCATTAGTGAGGAACTTCCCTATAGTGAATACTTCGAGTACTTTGCCCCAGATTTCACACTCCATCCAGATGTCAGCACCCGCATCGAGAATCAGAAC
Hprt	NM_013556.2	HK	31–130	TGCTGAGGCGGCGAGGGAGAGCGTTGGGCTTACCTCACTGCTTTCCGGAGCGGTAGCACCTCCTCCGCCGGCTTCCTCCTCAGACCGCTTTTTGCCGCGA
Pgk1	NM_008828.2	HK	37–136	CCGGCATTCTGCACGCTTCAAAAGCGCACGTCTGCCGCGCTGTTCTCCTCTTCCTCATCTCCGGGCCTTTCGACCTCACGGTGTTGCCAAAATGTCGCTT
Ppia	NM_008907.1	HK	391–490	CCAAGACTGAATGGCTGGATGGCAAGCATGTGGTCTTTGGGAAGGTGAAAGAAGGCATGAACATTGTGGAAGCCATGGAGCGTTTTGGGTCCAGGAATGG

Optimal sequences were designed on the chosen genes by Nanostring Technologies. The table reports the following for each gene: (*i*) the gene name; (*ii*) the accession number; (*iii*) whether it is a REST-target gene (YES/NO) or a housekeeping (HK) gene; the position (*iv*) and the sequence (*v*) of the targeted region.

Table S2. NanoString nCounter gene expression data

Gene name	Accession no.	REST-target	mSin3 N205_1	mSin3 N205_2	mSin3 N205_3	RILP N313_1	RILP N313_2	RILP N313_3	shREST_1	shREST_2	shREST_3	CTRL_1	CTRL_2	CTRL_3
REST-target genes														
Stat1	NM_009283.3	YES	−1.05	−1.32	−1.13	1.9	1.85	2.06	1.87	1.72	1.83	1.02	−1.09	1.07
Nfasc	NM_001160316.1	YES	2.08	−1.13	2.31	1.75	1.27	1.04	1.84	1.64	1.06	1.25	1.06	−1.32
Nrxn1	NM_177284.2	YES	3.78	2.71	−1.07	8.92	−1.07	1.62	4.75	5.89	−1.07	−1.07	−1.03	1.09
Grin2b	NM_008171.3	YES	2.56	4.79	1.99	5	2.35	−1.1	2.66	2.74	5.08	−1.9	3.17	−1.67
Cacna1h	NM_021415.4	YES	2.25	1.82	1.64	2.17	1.62	2.4	1.96	−1.06	1.1	−1.86	−1.13	2.11
Syp	NM_009305.2	YES	1.23	1.11	1.22	1.26	1.22	1.14	1.37	1.58	1.54	−1.02	1.1	−1.08
Grin1	NM_008169.2	YES	2.34	2.11	2.81	1.42	1.94	1.76	1.21	1.5	1.51	−1.03	−1.13	1.17
Cacna1i	NM_001044308.2	YES	2.39	2.18	2.29	2.03	2.28	2.2	1.06	−1.01	−2.58	1.3	−1.22	−1.06
Calb1	NM_009788.4	YES	2.98	1.98	1.24	4.53	4.19	2.59	4.75	−1.05	−1.07	−1.07	−1.03	1.09
Aplp1	NM_007467.3	YES	1.44	1.42	1.46	1.37	1.31	1.38	1.54	1.59	1.56	1	−1.01	1.01
Syt4	NM_009308.3	YES	1.9	1.72	1.72	1.67	1.7	1.64	1.48	1.39	1.34	1.01	−1.04	1.02
L1cam	NM_008478.3	YES	1.29	1.18	1.2	1.07	1.06	1.12	1.31	1.26	1.25	1	−1.02	1.02
Sst	NM_009215.1	YES	1.09	1.17	1.16	1.03	1.12	1.11	1.19	1.21	1.19	1.09	−1.19	1.09
Neurod1	NM_010894.2	YES	1.98	3.8	1.37	1.08	6.49	1.44	1.15	1.22	−1.02	−1.07	−1.03	1.09
Bdnf	NM_007540.4	YES	−1.07	−1.07	1.37	1.08	1.02	1.44	1.15	1.22	−1.02	−1.07	−1.03	1.09
Penk	NM_001002927.2	YES	−1.07	−1.07	1.37	1.08	1.02	1.44	1.15	1.22	−1.02	−1.07	−1.03	1.09
Glra3	NM_080438.2	YES	−1.07	−1.07	1.37	1.08	1.02	1.44	1.15	1.22	−1.02	−1.07	−1.03	1.09
Gabrg2	NM_177408.5	YES	−1.07	−1.07	1.37	1.08	1.02	1.44	1.15	1.22	−1.02	−1.07	−1.03	1.09
Oprm1	NM_001039652.1	YES	−1.07	−1.07	1.37	1.08	1.02	1.44	1.15	1.22	−1.02	−1.07	−1.03	1.09
Cacng2	NM_007583.2	YES	−1.07	−1.07	1.37	1	1.02	1.44	1.15	1.22	−1.02	−1.07	−1.03	1.09
Nrxn3	NM_001198587.1	YES	−1.07	−1.07	1.37	1	1.02	1.44	1.15	1.22	−1.02	−1.07	−1.03	1.09
Non–REST-target and housekeeping genes														
Map2k2	NM_023138.4	NO	−1.48	−2.34	−1.75	−1.84	−1.9	−1.63	−1.87	−2.03	−2.05	−1.86	−1.6	−1.93
Gtf3c1	NM_207239.1	NO	−1.36	−1.41	−1.26	−1.3	−1.31	−1.18	−1.21	−1.18	−1.29	−1.2	−1.24	−1.1
Tuba1a	NM_011653.2	NO	−1.63	−1.82	−1.7	−1.49	−1.59	−1.49	−1.28	−1.43	−1.38	−1.44	−1.46	−1.49
Camkk2	NM_145358.2	NO	−1.01	−1.14	−1.04	−1.05	−1.02	1	−1.04	−1.28	−1.27	1.02	−1.05	1.05
Syt17	NM_138649.1	NO	−1.22	−1.63	−1.33	−1.26	−1.14	−1.11	−1.9	−1.99	−2.02	1.03	−1	−1.03
Ubc	NM_019639.4	NO	−1.74	−1.76	−1.25	−1.08	−1.33	−1.15	−1.21	−1.51	1.09	1.14	−1.31	1.15
Gpi1	NM_008155.3	NO	−1.15	1.06	−1.13	−1.01	−1.27	−1.05	1.2	−1.21	−1.01	−1.01	1.1	−1.09
Map3k5	NM_008580.4	NO	−1.22	−1.12	−1.15	−1.15	−1.09	−1.25	−1.29	−1.16	−1.39	1.03	−1.09	1.06
Rcan1	NM_001081549.1	NO	−1.07	−1.04	−1	−1.11	−1.06	−1.07	−1.03	−1.04	−1.11	−1.08	−1.05	1.14
Tbx21	NM_019507.1	NO	−1.38	−1.23	−1.08	−1.21	−1.43	−1.26	−1.23	−1.39	−1.01	−1.28	1.34	−1.05
Tdo2	NM_019911.2	NO	−1.13	−1.65	1.34	−1.65	−1.65	1.86	2.87	−1.27	−1.58	2.26	−1.59	−1.42
Gapdh	NM_001001303.1	HK												
Hdac3	NM_010411.2	HK												
Hprt	NM_013556.2	HK												
Pgk1	NM_008828.2	HK												
Ppia	NM_008907.1	HK												

NanoString nCounter gene expression data. The table reports the following for each gene: (*i*) the gene name; (*ii*) the accession number; (*iii*) whether it is a REST-target gene (YES/NO) or a housekeeping (HK) gene; and (*iv*) the fold change of expression. Values were processed by using the nSolver Analysis Software Version 2.5 (nanoString Technologies). by removing the experimental background; normalizing to the HK genes; calculating the ratio with respect to the control samples; and visualizing the obtained data as a heat map (Fig. 1*E*).

Table S3. Electrophysiological properties of LOV2-PAH1–transduced primary neurons

Cell parameter	LOV2-PAH1 dark	LOV2-PAH1 light
n	21	21
Maximum firing rate (Hz)	18.57 ± 0.98*	48.67 ± 4.86
Current threshold (pA)	152.4 ± 14.01	179.8 ± 11.52
Membrane capacitance (pF)	49.02 ± 4.56	50.05 ± 4.00
Membrane resistance (MΩ)	259.5 ± 23.32	210.4 ± 11.47
Voltage threshold (mV)	−17.20 ± 2.31	−18.31 ± 1.93
AP amplitude (mV)	49.51 ± 3.21	53.79 ± 2.37
AHP (mV)	−21.58 ± 2.17	−20.74 ± 1.59
AP width 50% (ms)	1.195 ± 0.061	1.171 ± 0.059
Rising slope (V/s)	22.71 ± 2.04	28.92 ± 2.66
Repolarizing slope (V/s)	−9.490 ± 0.61	−10.09 ± 0.57

Electrophysiological parameters describing the passive membrane properties and the shape of the first elicited action potential measured for neurons transduced with LOV2-PAH1 and kept under either dark or lit conditions for 24 h. The analysis refers to the experiments and data reported in Fig. 6.

*$P < 0.001$; unpaired Student *t* test.

Other Supporting Information Files

Dataset S1 (DOCX)