

Supplemental Information

Supplemental Figures

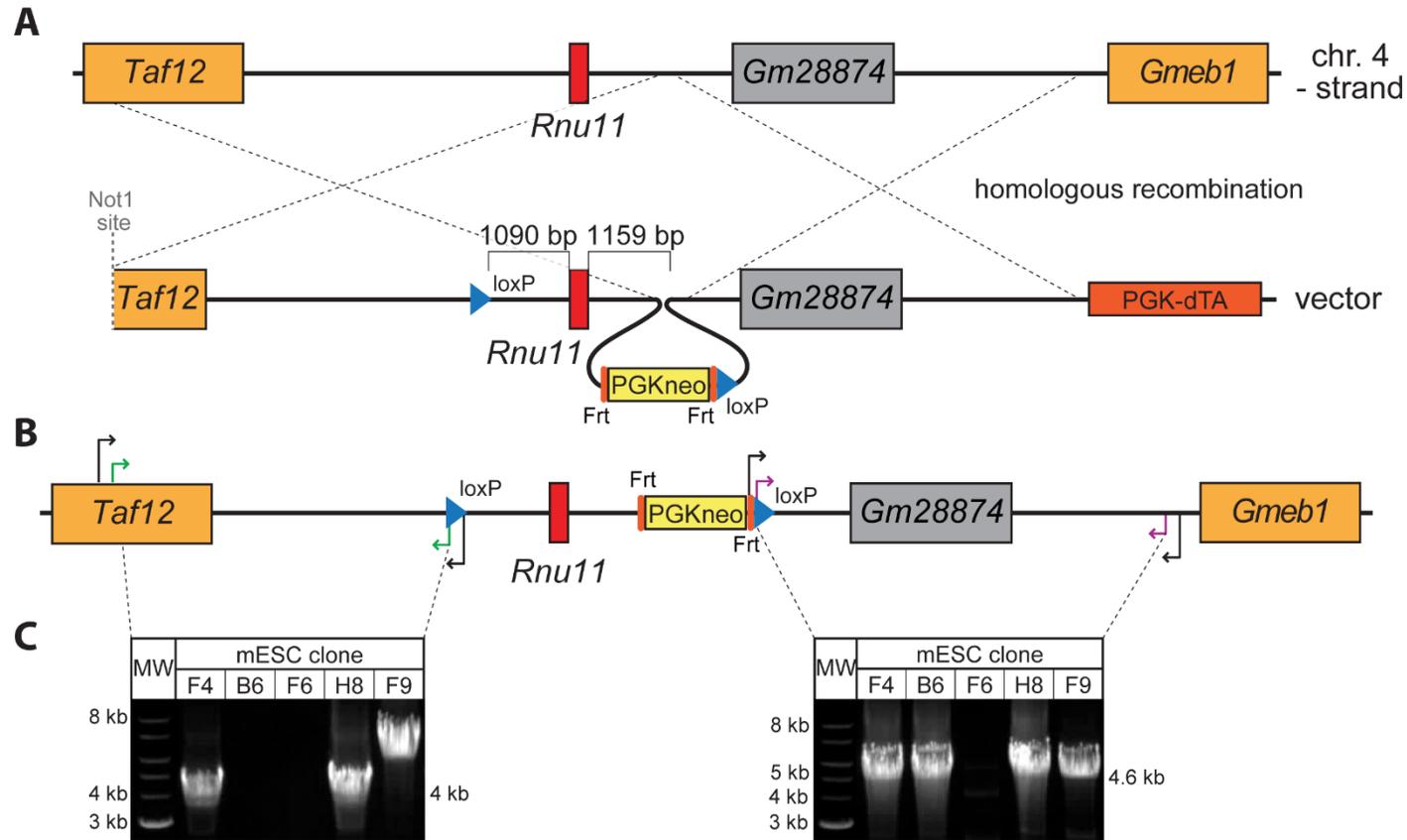


Figure S1. Generation and confirmation of the *Rnu11* cKO mice. Related to Figure 1. (A) Schematic of the *Rnu11* locus. The gray box represents the predicted gene *Gm28874*. Below the *Rnu11* locus schematic is the targeting construct used to introduce the loxP sites by homologous recombination (dashed lines). **(B)** Schematic representing the targeted allele, showing the primers used to interrogate the 5' loxP site (black and green arrows, left) and the 3' loxP site (black and purple arrows, right). For the 5' loxP site primer set, the outermost forward primer (black arrow, left) was designed outside the 5' arm of homology, with the outermost reverse primer (black arrow, left) positioned downstream of the 5' loxP site. The inner set of nested primers (green arrows) were designed in the 5' arm of homology and within the 5' loxP site, respectively. For the 3' loxP site primer set, the outermost forward primer (black arrow, right) was designed in the Frt site located upstream of the 3' loxP site, with the outermost reverse primer (black arrow, right) positioned downstream of the 3' arm of homology. The inner set of nested primers (purple arrows) were designed in the 3' loxP site and the 3' arm of homology, respectively. **(C)** Agarose gel images of long-range nested PCRs, performed on genomic DNA (gDNA) from targeted ES cells, using the 5' loxP site primer set (black and green arrows, left) and the 3' loxP site primer set (black and purple arrows, right). The number at the right of each gel image represents the expected product size produced from this strategy.

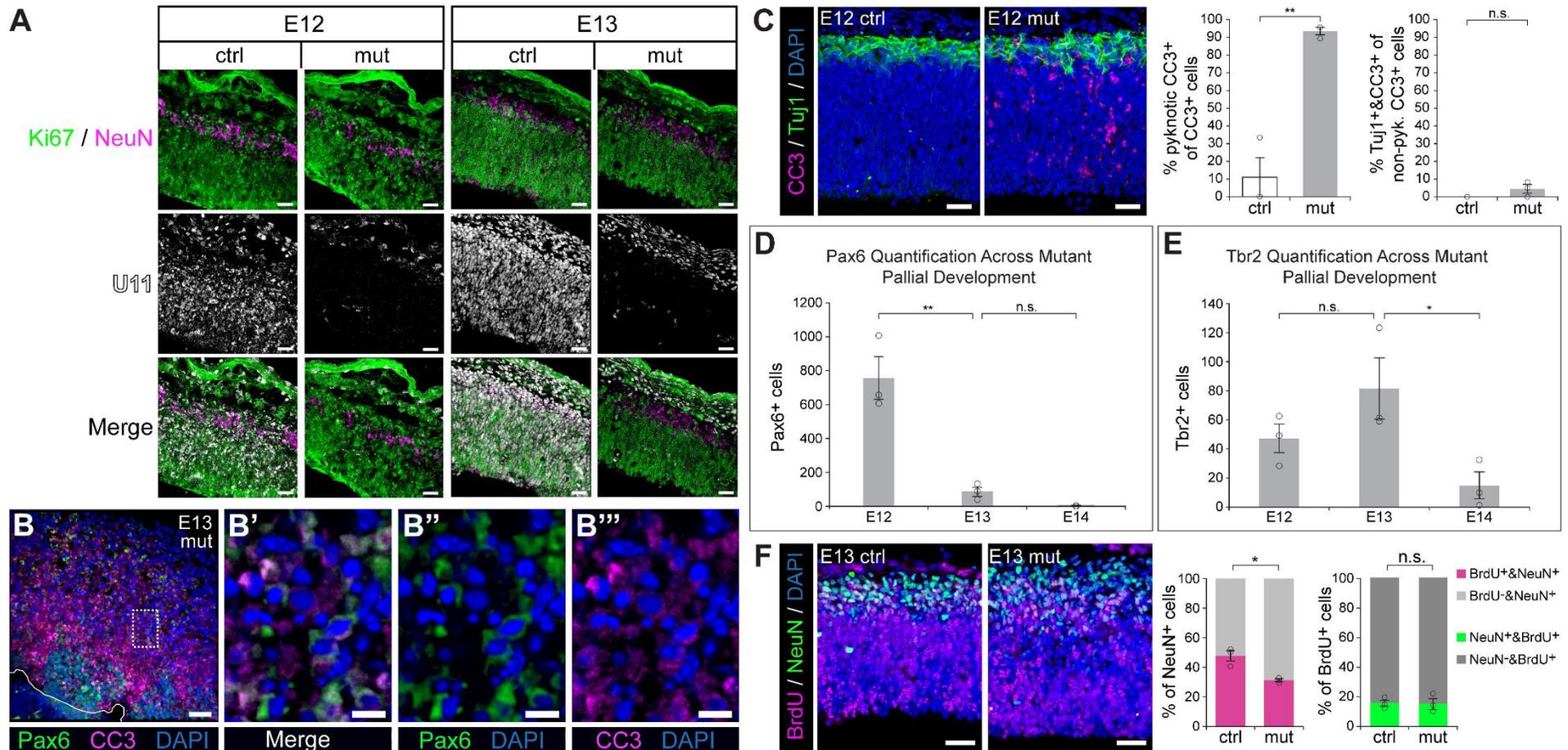


Figure S2. U11 loss and death of self-amplifying RGCs in the mutant pallium. Related to Figure 3. (A) Separated channels for sagittal section U11 FISH (middle row) and immunofluorescence (IF) for Ki67 (green) and NeuN (magenta) (top row), with overlay (bottom row), from the E12 and E13 control (ctrl) and mutant (mut) pallium. Scale bars=30 μ m. **(B)** IF for Pax6 (green) and cleaved caspase 3 (CC3, magenta) on sagittal section of E13 mutant pallium. White line marks the boundary between the subpallium and the pallium. Scale bar=30 μ m. **(B'-B''')** Magnified images of the dashed box in **(B)**, showing overlay of all channels (**B'**), Pax6 and DAPI (**B''**), and CC3 and DAPI (**B'''**). Scale bars=7 μ m. **(C)** IF for CC3 (magenta) and Tuj1 (green) in sagittal section of the E12 ctrl and mut pallium, with bar graphs showing the percentage of CC3⁺ cells that were pyknotic (left) and the percentage of non-pyknotic CC3⁺ cells that were Tuj1⁺ (right). Scale bars=30 μ m. **(D-E)** Bar graphs showing quantification of cells with nuclear Pax6 staining **(D)** or cells with nuclear Tbr2 staining **(E)** across development in the mutant pallium, from E12 and E14. Statistical significance across the three tested time-points was determined by one-way ANOVA, followed by Tukey's multiple comparison test to determine specific *P* values. **(F)** IF for BrdU (magenta) and NeuN (green) in the E13 ctrl and mut pallium, which had been pulsed with BrdU at E12. Scale bars=30 μ m. At right are bar graphs showing the percentage of NeuN⁺ cells that were BrdU⁺ in the E13 pallium, or the percentage of non-pyknotic BrdU⁺ cells that were NeuN⁺ in the E13 pallium. Statistical significance was determined by two-tailed student's *t*-tests (Table S8). Quantification data are represented as mean \pm SEM from *N*=3 for each condition per time-point; individual data points are superimposed on bar graphs. Non-pyk.=non-pyknotic. n.s.=not significant; *=*P*<0.05; **=*P*<0.01.

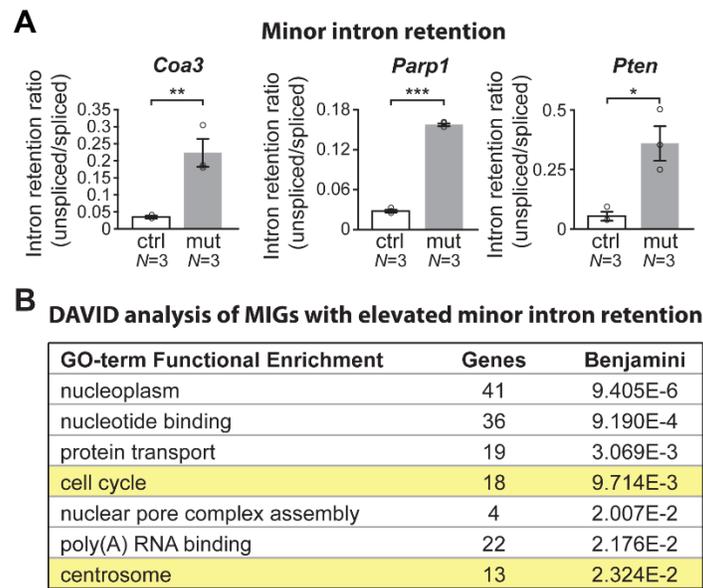


Figure S3. Validation of expression and minor intron retention changes in the E12 mutant pallium. Related to Figure 5. (A) Minor intron retention of *Coa3* (left), *Parp1* (middle), and *Pten* (right) in the E12 ctrl and mut pallium, as determined by qRT-PCR. The value plotted is the average ratio of normalized unspliced expression/normalized spliced expression \pm SEM from $N=3$ for each condition. Individual data points are superimposed on the bar graphs. Statistical significance was determined by two-tailed student's *t*-tests (Table S8). $*=P<0.05$, $**=P<0.01$, $***=P<0.001$. (B) GO Terms enriched for by DAVID analysis of MIGs with statistically significant elevated minor intron retention in the E12 mutant pallium, with cell cycle-related terms highlighted in yellow.

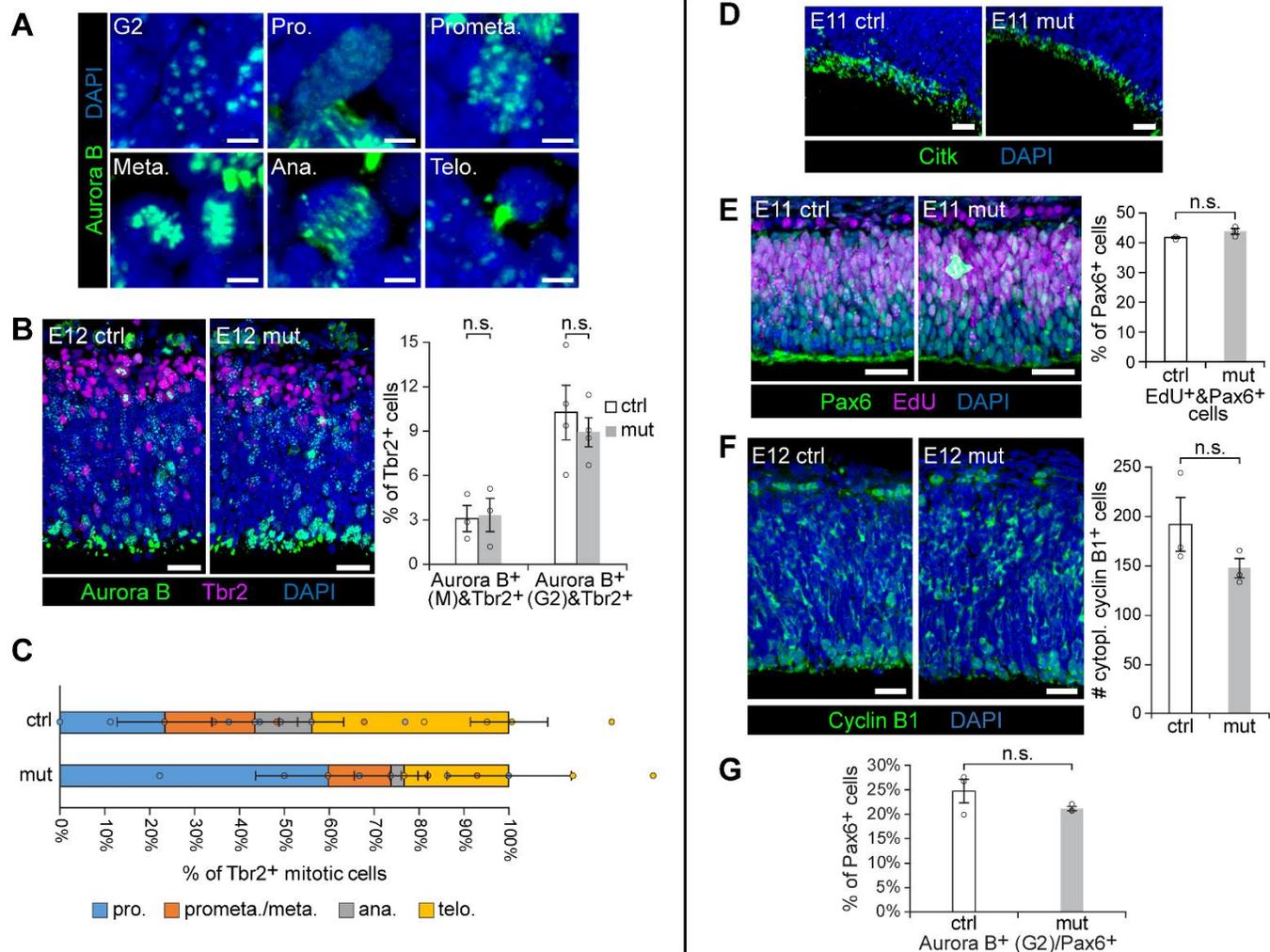


Figure S4. Cell cycle defects are not observed in the E11 mutant pallium, in IPCs of the E12 mutant pallium, or in G2 phase. Related to Figure 5. (A) IF for Aurora B (green, AurB) showing phase-specific Aurora B staining patterns in cells in G2, prophase (pro.), prometaphase (prometa.), metaphase (meta.), anaphase (ana.), and telophase (telo.) in the E12 pallium. Scale bars=4 μ m. (B) IF for Aurora B (green) and Tbr2 (magenta) in sagittal sections of control (ctrl) and mutant (mut) E12 pallium, with bar graph showing the percentage of Tbr2⁺ cells with mitosis- and G2-specific ($N=4$) Aurora B staining patterns. (C) Bar graph showing the percentage of Aurora B⁺/Tbr2⁺ mitotic cells in prophase, prometaphase/metaphase (prometa./meta.), anaphase, and telophase ($N=4$). (D) IF for Citk (green) in sagittal sections of E11 control and mutant pallium. (E) IF for Pax6 (green) and EdU detection (magenta) in sagittal sections from E11 control (ctrl, left) and mutant (mut, right) pallium, with bar graph showing the percentage of Pax6⁺ cells that were EdU⁺ in the E11 ctrl and mut pallium. (F) IF for cyclin B1 (green) in sagittal sections of the E12 ctrl and mut pallium, with bar graph showing the number of cell with cytoplasmic (cytopl.) cyclin B1. (G) Bar graph showing the percentage of Pax6⁺ cells with G2-specific Aurora B⁺ staining in the E12 ctrl and mut pallium. Scale bars=30 μ m, unless otherwise specified. Quantification data are represented as mean \pm SEM from $N=3$ for each condition per time-point, unless otherwise specified. Statistical significance was determined by two-tailed student's t -test (Table S8). n.s.=not significant.

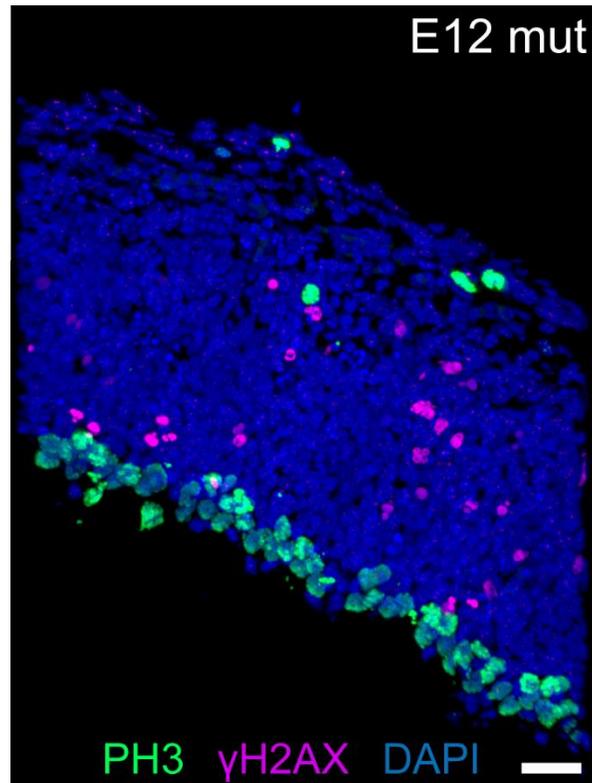


Figure S5. Cells with DNA damage are not in mitosis. Related to Figure 6. IF for PH3 (green) and γ H2AX (magenta) in a sagittal section of the E12 mutant pallium. Scale bar=30 μ m.

Table S1. RNAseq gene expression data. Related to Figure 4. FC=expression fold-change between control and mutant.

[Click here to Download Table S1](#)

Table S2. Mammalian functions of the 4 genes upregulated in the mutant pallium that enriched for “intrinsic apoptotic signaling pathway in response to DNA damage by p53 mediator” by DAVID. Related to Figure 4. FC=expression fold-change between control and mutant. Ave=average.

| Gene | Synonyms | Ave FPKM Ctrl | Ave FPKM Mut | FC | Function Description |
|---------------|----------------------------------|---------------|--------------|---------|--|
| <i>Bbc3</i> | <i>Puma, Jfy1, Jfy-1</i> | 0.7199858 | 1.8057943 | 2.43627 | p53 upregulated modulator of apoptosis, a BH3 domain-containing gene (PMID: 11572983); alongside <i>Pmaip1</i> (below), important activator of genotoxic stress-induced apoptosis in NPCs (PMID: 16822983); required for apoptosis driven by increased p53 expression (PMID: 12574499) |
| <i>Eda2r</i> | <i>Xedar</i> | 1.6703153 | 10.5807371 | 6.85729 | both <i>EDA2R</i> and its ligand, <i>EDA</i> , are transcriptionally activated by p53 (PMID: 19543321, 20501644); regulator of p53-mediated apoptosis via interaction with the death receptor FAS, and increased expression of <i>EDA2R</i> results in upregulation/stabilization of FAS protein levels (PMID: 19543321) |
| <i>Perp</i> | <i>Kcp1, Krtcap1, Pigp1, Thw</i> | 0.4671268 | 1.0282991 | 2.23922 | p53 apoptosis factor related to PMP-22; transcriptionally activated by p53 during p53-mediated apoptosis, but not during p53-mediated G1 arrest (PMID: 10733530, 14707288); induces p53 upregulation, post-translational p53 modifications that disrupt MDM2-p53 binding, and p53 nuclear translocation (PMID: 21451571); important positive regulator of p53-mediated apoptosis in the embryonic mouse brain (PMID: 14614825); DNA damage upregulates <i>Perp</i> expression (PMID: 14614825) |
| <i>Pmaip1</i> | <i>Noxa, Apr</i> | 0.7210288 | 2.3228602 | 2.56006 | pro-apoptotic, BH3 domain-containing gene whose transcription is activated by p53 (PMID: 10807576, 14500851); alongside <i>Bbc3</i> (above), important activator of genotoxic stress-induced apoptosis in NPCs (PMID: 16822983) |

Table S3. Minor intron retention data, derived from RNAseq data. Related to Figure 5. *P*-values were determined by two-tailed student's *t*-test. Rep=replicate; Avg=average. MSI=mis-splicing index.

[Click here to Download Table S3](#)

Table S4. Prediction of the effect of minor intron retention in the isoforms of MIGs with significant elevation of minor intron retention in the E12 mutant pallium. Related to Figure 5. ORF=open reading frame; NMD=nonsense-mediated decay.

[Click here to Download Table S4](#)

Table S5. Mammalian functions of the 21 MIGs regulating cell cycle with significantly elevated minor intron retention the mutant pallium. Related to Figures 5, S3, and S4. *P*-values were determined by two-tailed student's *t*-test. Syn=synonyms, ave=average. MSI=mis-splicing index.

[Click here to Download Table S5](#)

Table S6. The DNA damage response functions of 14 MIGs with significantly elevated minor intron retention in the mutant pallium. Related to Figure 4. MSI=mis-splicing index. *P*-values were determined by two-tailed student's *t*-test. Syn=synonyms, ave=average.

| MIG | Syns | Ave FPKM Ctrl | Ave FPKM Mut | Ave MSI Ctrl | Ave MSI Mut | MSI <i>P</i> -value | ΔMSI | DNA Damage Response Function Description |
|---------------|--|---------------|--------------|--------------|-------------|---------------------|---------------|---|
| <i>Baz1b</i> | <i>Wstf</i> , <i>Wbscr9</i> , <i>Wbscr10</i> | 21.907384 | 25.945518 | 5.447441 | 12.41971 | 0.019728 | 13.97227 | forms nucleosome remodeling complex with ISWI (complex: WICH), which phosphorylates and maintains this phosphorylation of H2AX (γH2AX), thereby regulating important steps in the DNA damage response process (PMID: 19092802) |
| <i>Ccnk</i> | <i>Cpr4</i> , <i>CycK</i> | 55.348807 | 5.326979 | 5.053107 | 27.09289 | 0.004641 | 22.03978 | cyclin K; when complexed with Cdk9, accumulates on chromatin in response to replication stress, and ssDNA in stressed cells (PMID: 20930849) |
| <i>Cep164</i> | <i>Nphp15</i> | 5.060874 | 5.324126 | 5.904086 | 30.06079 | 0.000429 | 24.15670 1 | centrosomal protein that is phosphorylated by the DNA damage response proteins ATR and ATM; siRNA-mediated knockdown in HeLa cells results in reduced phosphorylation of multiple DNA damage response proteins and chromosome missegregation (PMID: 18283122) |
| <i>Cull1</i> | | 41.697261 | 47.309440 | 2.804814 | 16.18051 | 0.001812 | 13.3757 | as part of the SCF E3 ubiquitin ligase complex, ubiquitinates Ku80, a component of the initiating complex of the NHEJ double-strand break DNA repair pathway, thereby regulating this complex's removal from DNA (PMID: 23324393) |
| <i>Cul4a</i> | | 11.101313 | 12.071160 | 3.608965 | 15.74483 | 0.044297 | 12.13587 | cullin; ubiquitin ligase that targets Ddb1 (another MIG, below) and Ddb2, both of which act to initiate the DNA damage response, for ubiquitination and degradation, thereby regulating the DNA damage response (PMID: 19481525) |
| <i>Dna2</i> | <i>Dna2l</i> | 3.9216094 | 3.7578457 | 4.807850 | 17.42876 | 0.015044 | 12.62091 | due to 5' to 3' endonuclease activity, involved in resection extension in homologous recombination-mediated DNA double stranded break repair (PMID: 28718810) |

| | | | | | | | | |
|--------------|--|-----------|-----------|----------|----------|----------|----------|---|
| <i>Ddb1</i> | <i>XPE, DDBA, XAP1, XPCE</i> | 86.093717 | 88.333331 | 0.461818 | 2.706198 | 0.004385 | 2.244381 | large subunit of DNA damage-binding complex, which functions in nucleotide excision repair (PMID: 16951172); loss of <i>Ddb1</i> in mouse brain results in accumulation of cell cycle regulators and increased genomic instability, ultimately causing apoptosis of proliferating neuronal progenitors (PMID: 17129780) |
| <i>E2f1</i> | <i>Rbp3, Rbap1, Rbbp3</i> | 6.067779 | 6.486815 | 12.91249 | 53.23981 | 0.000964 | 40.32732 | upregulated in response to DNA damage, and promotes DNA damage-induced apoptosis (PMID: 11459832); indirectly regulates the transcription of the DNA damage response gene <i>GADD45A</i> (PMID: 20713352) |
| <i>Erc5</i> | <i>Xpg</i> | 5.363914 | 6.275879 | 1.333767 | 7.642992 | 0.048076 | 6.309225 | single strand-specific DNA endonuclease involved in the 3' incision step of nucleotide excision repair (PMID: 7657672) |
| <i>Exo1</i> | <i>Msa, Hex1</i> | 4.4268470 | 4.1420745 | 3.731127 | 9.386061 | 0.008405 | 5.654934 | 5' to 3' endonuclease involved in DNA mismatch repair and resection extension in homologous recombination-mediated DNA double stranded break repair (PMID: 24705021) |
| <i>Ints7</i> | | 8.821004 | 8.814813 | 3.590800 | 13.52046 | 0.012655 | 9.929663 | recruited to sites of DNA damage and interacts with SSB1, a DNA damage sensor recognized by ATM (PMID: 21659603) |
| <i>Parp1</i> | <i>Parp, Ppol, Adprt, Ard1, Adprt1</i> | 47.879274 | 47.679811 | 0.771961 | 14.72329 | 0.002560 | 13.95133 | nuclear protein that promotes formation of poly(ADP-ribose) chains (PARylation), which transfers ADP-ribose group from NAD ⁺ to target protein and forms a scaffold around DNA breaks, allowing for recruitment of essential DNA damage response factors (PMID: 21989215, 22431722) |
| <i>Usp10</i> | <i>UBPO, Uchrp</i> | 10.625277 | 8.050707 | 4.474797 | 17.26511 | 0.012563 | 12.79031 | after DNA damage, phosphorylated by ATM, resulting in <i>Usp10</i> stabilization and transport to the nucleus, where it deubiquitylates p53, thereby stabilizing p53 (PMID: 20096447) |

Table S7. Primer sequences for RT-PCR.

| Primer Name | Primer Direction | Sequence (5' to 3') |
|--------------------------------------|--------------------|------------------------------|
| <i>Rnu11</i> cKO, 5' loxP | Forward (primer 1) | ACCCTCCCCTACTGTTTTAC |
| | Reverse (primer 2) | AGGCTGCTACAGGATGACTC |
| <i>Rnu11</i> cKO, 3' loxP | Forward (primer 3) | CATGTGTTTGCTGGGAATTG |
| | Reverse (primer 4) | CTCATGAGGCAGATCTCTGAA |
| <i>Hist1h1a</i> expression | Forward | AGAAGAACAACAGCCGCATCAAAGTGG |
| | Reverse | CTTGGACTCAGCCTTCTTGTTCAGCTT |
| Cre genotyping | Forward | TATCCAGCAACATTTGGGCCAGCT |
| | Reverse | AACATTCTCCCACCGTCAGTACGTGA |
| <i>Emx1</i> -Cre zygosity, wild-type | Forward | AAGGTGTGGTTCCAGAATCG |
| | Reverse | CTCTCCACCAGAAGGCTGAG |
| <i>Emx1</i> -Cre zygosity, mutant | Forward | GATCTCCGGTATTGAAACTCCAGC |
| | Reverse | GCTAAACATGCTTCATCGTCGG |
| U11 expression | Forward | AAAGGGCTTCTGTCGTGAGTGGC |
| | Reverse | CCGGGACCAACGATCACCAG |
| <i>Neat1</i> expression | Forward | AATTGGCCAGAAGACAACAGGGTTTGC |
| | Reverse | GTATTCAGTGGCAAAGCACTCATGAGG |
| <i>Coa3</i> minor intron splicing | Forward | CAGTTGCAGTTTATGCGGCAGGTG |
| | Reverse (intronic) | CTAGCCACCCTTGCTGTTTTCCCAA |
| | Reverse (exonic) | CAGCTTTGGCTTCATCTTCCAGCTC |
| <i>Pten</i> minor intron splicing | Forward | CTCCCAGACATGACAGCCATCATCAA |
| | Reverse (intronic) | CTACTCCCACGTTATCAGAGTGACAGAA |
| | Reverse (exonic) | CAAGTCTTTCTGCAGGAAATCCCATAGC |
| <i>Parp1</i> minor intron splicing | Forward | AAAACCACCCCTGACCCTTCG |
| | Reverse (intronic) | GCACACAGCATAGCCAAGAAAGG |
| | Reverse (exonic) | AATGTACCTGGGGAGGGCAGTT |
| <i>Sfrs10</i> expression | Forward | AGCAGGTCTTACAGCCGAGATTATCG |
| | Reverse | CCAAACACGCCAAGACAACAGTTG |
| <i>Spc24</i> expression | Forward | CTCATACCTTGCACAGAAGTGGGGTT |
| | Reverse | AATAAAAAAGAAGCTGCAGGCCAGCC |
| Intergenic primers | Forward | GGATAGTTCATCTCCTGCAGGTCACAAG |
| | Reverse | GTCCCACCCATCTAGTTTAGCATCAGC |

Table S8. Summary of statistical analyses performed.

| Fig. | Experimental Paradigm (ex: immunostaining) | N-value | P-value | t/f-value | Statistical test | Test for multiple testing? | Does data meet assumption of normality? |
|------|--|--------------------------|---|---|----------------------------------|----------------------------|---|
| 1B | qPCR | WT=3 WT/KO=3 | 6.43E-05 | 17.38388 | Student's t-test ¹ | No | Yes |
| 1C | Genotype frequency | WT=20 Het=45 Mut=0 | 0.0001067 | N/A | Chi-squared goodness of fit test | No | Yes |
| 1J | Pallium thickness | Ctrl=3 Mut=3 | 0.63251 (E12) 0.96958 (E13) 0.02533 (E14) | -0.5168 (E12) 0.0405 (E13) 3.4805 (E14) | Student's t-test ¹ | No | Yes |

| | | | | | | | |
|----|--------------|---|---|---|--|----|-----|
| 2A | TUNEL | Ctrl=3 Mut=3 | 0.587853 | -0.58845 | Student's t-test ¹ | No | Yes |
| 2B | TUNEL | Ctrl=4 Mut=4 | 0.000364 | -7.19759 | Student's t-test ¹ | No | Yes |
| 2C | TUNEL | Ctrl=3 Mut=3 | 0.001647 | -7.55204 | Student's t-test ¹ | No | Yes |
| 2D | TUNEL | Ctrl=4 Mut=4 | 0.017318 | -3.25681 | Student's t-test ¹ | No | Yes |
| 2E | CC3 | Ctrl=3 Mut=3 | 0.348641 | -1.06066 | Student's t-test ¹ | No | Yes |
| 2F | CC3 | Ctrl=4 Mut=4 | 0.006263 | -4.11334 | Student's t-test ¹ | No | Yes |
| 3D | Ki67 | Ctrl=3 Mut=3 | 0.94384 (E12) 0.00114 (E13) 9.86E-05 (E14) | 0.07497 (E12) 8.31546 (E13) 15.6004 (E14) | Student's t-test ¹ | No | Yes |
| 3D | NeuN | Ctrl=3 Mut=3 | 0.58591 (E12) 0.50374 (E13) 0.00256 (E14) | 0.59165(E12) 0.73383 (E13) 6.71922 (E14) | Student's t-test ¹ | No | Yes |
| 3H | Pax6 | Ctrl=3 Mut=3 | 0.91943 (E12) 0.00063 (E13) 2.83E-06 (E14) | 0.1076 (E12) 9.6834 (E13) 38.116 (E14) | Student's t-test ¹ | No | Yes |
| 3L | Tbr2 | Ctrl=3 Mut=3 | 0.983274 (E12) 0.030437 (E13) 0.001203 (E14) | -0.0223 (E12) 3.2822 (E13) 8.2035 (E14) | Student's t-test ¹ | No | Yes |
| 4B | qRT-PCR | Ctrl=4 Mut=4 | 0.000109 | 8.944523 | Student's t-test ¹ | No | Yes |
| 4C | qRT-PCR | Ctrl=4 Mut=4 | 0.004161 | 4.487097 | Student's t-test ¹ | No | Yes |
| 4E | yH2AX | Ctrl=3 Mut=3 (E11) 0.027471 (E12) Ctrl=3 Mut=4 (E12) | 0.039021 (E11) 0.027471 (E12) | -3.02372 (E11) -4.02969 (E12) | Student's t-test ¹ (E11) T-test: two sample assuming unequal variances (E12) | No | Yes |
| 4E | P53 | Ctrl=3 Mut=3 (E11) Ctrl=3 Mut=4 (E12) | 0.57902 (E11) 0.01497 (E12) | 0.60302 (E11) -5.05123 (E12) | Student's t-test ¹ (E11) T-test: two sample assuming unequal variances (E12) | No | Yes |
| 4G | Pax6/ p53 | Mut=3 | P<0.001 (Fisher's method) 0.1651 (N=1) 2.53E-04 | N/A | Fisher Exact test ² | No | Yes |

| | | | | | | | |
|----|----------------------|--|---|--|--|----|-----|
| | | | (N=2) 6.72E-04 (N=3) | | | | |
| 5C | Ki67/ PH3 | Ctrl=5 Mut=5 | 0.036877 | -2.50113 | Student's t-test ¹ | No | Yes |
| 5D | Pax6/ AuroraB (M) | Ctrl=3 Mut=3 | 0.281507 | -1.24374 | Student's t-test ¹ | No | Yes |
| 5E | Pax6/ AuroraB | Ctrl=4 Mut=4 | 0.050205 (prophase) 0.022396 (prometaphase/ metaphase) 0.399731 (anaphase) 0.853745 (telophase) | 2.443901 (prophase) -3.05402 (prometaphase/ metaphase) 0.906253 (anaphase) 0.19244 (telophase) | Student's t-test ¹ | No | Yes |
| 5F | Citk | Ctrl=3 Mut=3 (E11) Ctrl=4 Mut=4 (E12) | 0.909922 (E11) 0.001502 (E12) | -0.1205 (E11) 5.5091 (E12) | Student's t-test ¹ | No | Yes |
| 5G | Pax6/ EdU | Ctrl=3 Mut=4 | 0.00013 | 10.58504 | T-test: two sample assuming unequal variances | No | Yes |
| 5H | Tbr2/ EdU | Ctrl=3 Mut=3 | 0.143103 | 1.972462 | Student's t-test ¹ | No | Yes |
| 6A | yH2AX/ EdU | Mut=3 | 0.5<P<0.6 (Fisher's method) 0.0953 (n=1) 0.762 (n=2) 1.0 (n=3) | N/A | Fisher Exact test ² | No | Yes |
| 6B | P53/EdU | Mut=3 | P<0.001 (Fisher's method) 0.0095 (N=1) 0.4087 (N=2) 4.61E-06 (N=3) | N/A | Fisher Exact test ² | No | Yes |
| 6C | P53/ AuroraB (M) | Mut=4 | P<0.001 (Fisher's method) 3.94E-06 (N=1) 3.24E-04 (N=2) 0.0135 (N=3) 0.0115 (N=4) | N/A | Fisher Exact test ² | No | Yes |
| 6C | P53/ AuroraB (G2) | Mut=4 | P<0.001 (Fisher's method) 9.18E-05 | N/A | Fisher Exact test ² | No | Yes |

| | | | | | | | |
|-----|-----------------------|---|---|--|-----------------------------------|-------------------|-----|
| | | | (N=1) 2.76E-06 (N=2) 0.0321 (N=3) 2.01E-07 (N=4) | | | | |
| 6D | EdU/CC3 | Mut=3 | 0.4<P<0.5 (Fisher's method) 0.345 (N=1) 0.586 (N=2) 0.265 (N=3) | N/A | Fisher Exact test ² | No | Yes |
| S2C | CC3/pyknotic cells | Ctrl=3 Mut=3 | 0.001891 | -7.28116 | Student's t-test ¹ | No | Yes |
| S2C | CC3/Tuj1 | Ctrl=3 Mut=3 | 0.140357 | -1.83533 | Student's t-test ¹ | No | Yes |
| S2D | Pax6 | E12=3 E13=3 E14=3 | 0.001 (ANOVA) 0.002 (E12- E13) 0.719py (E13- E14) | 30.835 (ANOVA) | One-way ANOVA | Post-hoc Tukey | Yes |
| S2E | Tbr2 | E12=3 E13=3 E14=3 | 0.046 (ANOVA) 0.284 (E12- E13) 0.039 (E13- E14) | 5.351 (ANOVA) | One-way ANOVA | Post-hoc Tukey | Yes |
| S2F | BrdU/ NeuN | Ctrl=3 Mut=3 | 0.0115 (%NeuN) 0.96281 (%BrdU) | 4.416383 (%NeuN) 0.049606 (%BrdU) | Student's t-test ¹ | No | Yes |
| S3A | qRT-PCR | Ctrl=3 Mut=3 | 0.009763 (Coa3) 0.014807 (Pten) 2.13E-06 (Parp1) | -4.6360 (Coa3) -4.1038 (Pten) -40.90389 (Parp1) | Student's t-test ¹ | No | Yes |
| S4B | Tbr2/ AuroraB | Ctrl=4 Mut=4 (G2) Ctrl=3 Mut=3 (M) | 0.544268 (G2) 0.890293 (M) | 0.642447 (G2) -0.14693 (M) | Student's t-test ¹ | No | Yes |
| S4C | Tbr2/ AuroraB | Ctrl=4 Mut=4 | 0.109109 (prophase) 0.640059 (prometaphase/ metaphase) 0.266296 (anaphase) 0.258209 (telophase) | -1.8803 (prophase) 0.492213 (prometaphase/ metaphase) 1.225527 (anaphase) 1.248928 (telophase) | Student's t-test ¹ | No | Yes |
| S4E | Pax6/ EdU | Ctrl=3 Mut=3 | 0.114046 | -2.01577 | Student's t-test ¹ | No | Yes |
| S4F | Cyclin B1 | Ctrl=3 | 0.196738 | 1.547139 | Student's | No | Yes |

| | | | | | | | |
|-----|-----------------------|-----------------|----------|----------|----------------------------------|----|-----|
| | | Mut=3 | | | t-test ¹ | | |
| S4G | Pax6/ AuroraB (G2) | Ctrl=3 Mut=3 | 0.223782 | 1.438129 | Student's t-test ¹ | No | Yes |

¹ Two-sample t-test assuming equal variances

² Followed by Fisher's method to combine *P*-values