Supplementary Table Legends

Table S1. Cherry picker siRNA list and Primer pairs utilized for qRT-PCR. siRNAs utilized for the differentiation and viability screen (Dharmacon). Primer pairs (forward and reverse) and TaqMan Probes used for qRT-PCR.

Table S2. siELF4 differentially expressed genes. All significantly differentially expressed genes following ELF4 knockdown in BE(2)-C cells, grouped by expression (up or down).

Table S3. Survival analysis of Versteeg and Seeger cohorts. Results of univariate and multivariate Cox proportional hazards regression analysis for the Versteeg and Seeger cohorts.

Table S4. PANTHER and mSigDB analysis of siELF4 down- and up-regulated genes. Results from pathway analysis using siELF4 differentially expressed genes (**Table S3**).

Table S5. miR-124 target list (mNSC Study and miRTarBase). miR-124-regulated genes identified from murine Neural Stem Cells were combined with high-confidence targets from miRTarBase.

Table S6. Overlap between miR-124 targets and siELF4 down-regulated genes. miR-124 targets (**Table S5**) were compared to siELF4 down-regulated genes (**Table S3**) to identify co-targeted genes.

Table S7. mSigDB analysis of overlap between miR-124 targets and siELF4 downregulated genes. Genes that are co-targeted by miR-124 and ELF4 (**Table S6**) were assessed for gene-set enrichment utilizing mSigDB.

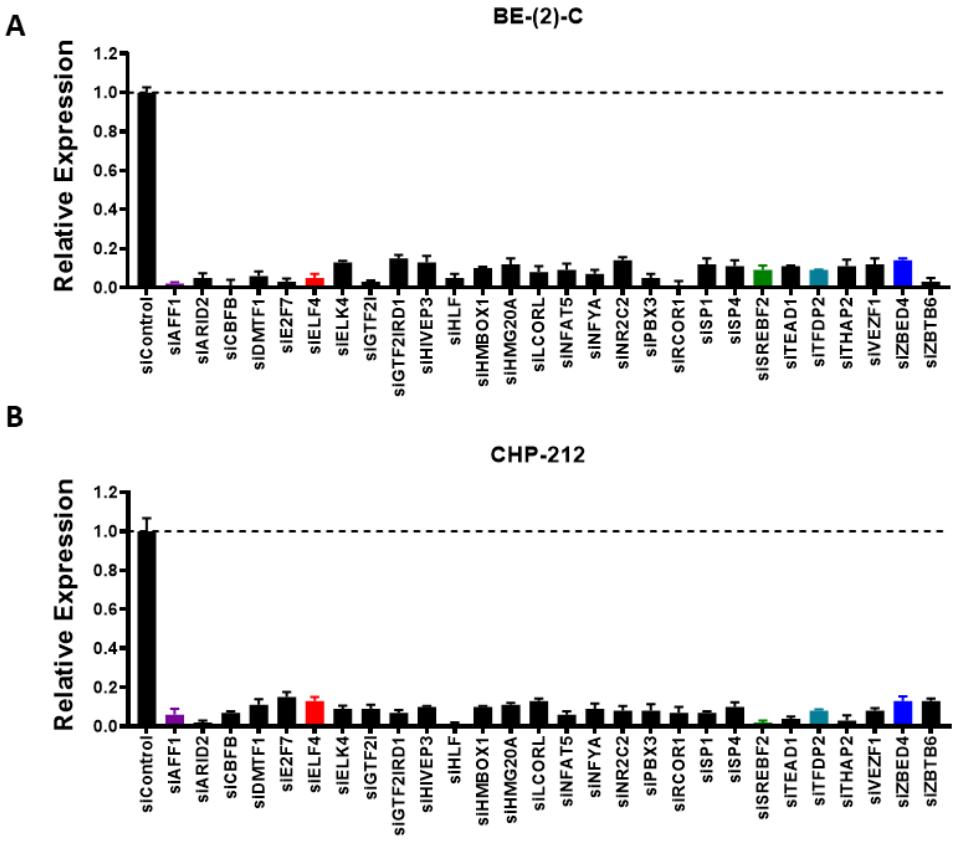

Figure S1. Knockdown efficiency 48 h after reverse transfection. BE(2)-C and CHP-212 cells were reverse transfected with siRNAs against each TF. After 48 h, RNA was isolated and qRT-PCR was performed to assess the knockdown efficiency for each siRNA. All knockdowns had a significant change in comparison to siControl based on a Student's t-test with a nominal significance threshold of $p = 0.05$.

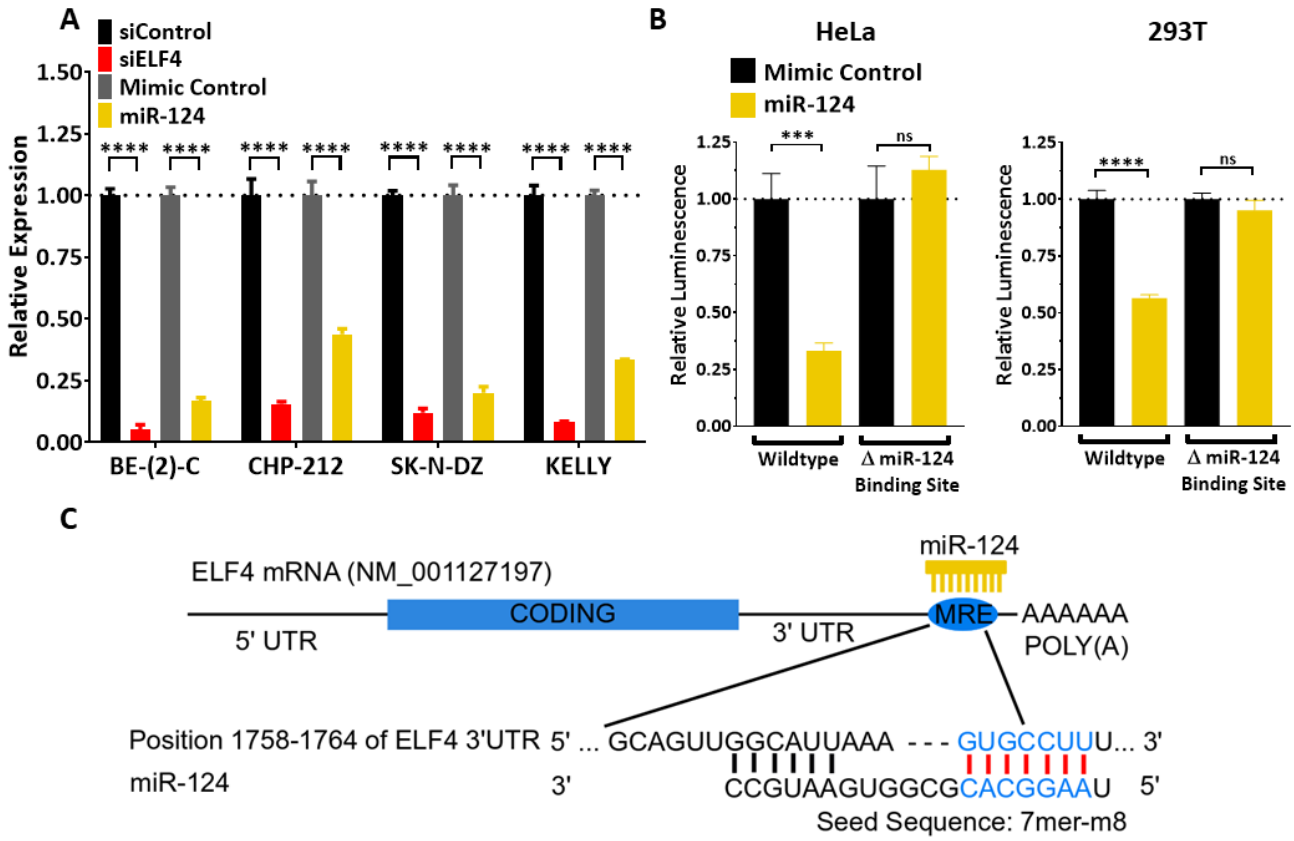

Figure S2. miR-124 directly binds ELF4 and regulates ELF4 expression. (A) ELF4 expression levels in BE(2)-C and CHP-212 cells 48 h after transfection. **(B)** HeLa and 293T cells co-transfected with an ELF4 3'UTR luciferase reporter and miRNA mimics. Luminescence was measured after 48 h. **(C)** The miR-124 binding site in ELF4 3'UTR. Statistical significance of observed changes was determined by Student's t test. * = $p<0.05$, *** = $p<0.001$, **** = $p<0.0001$.

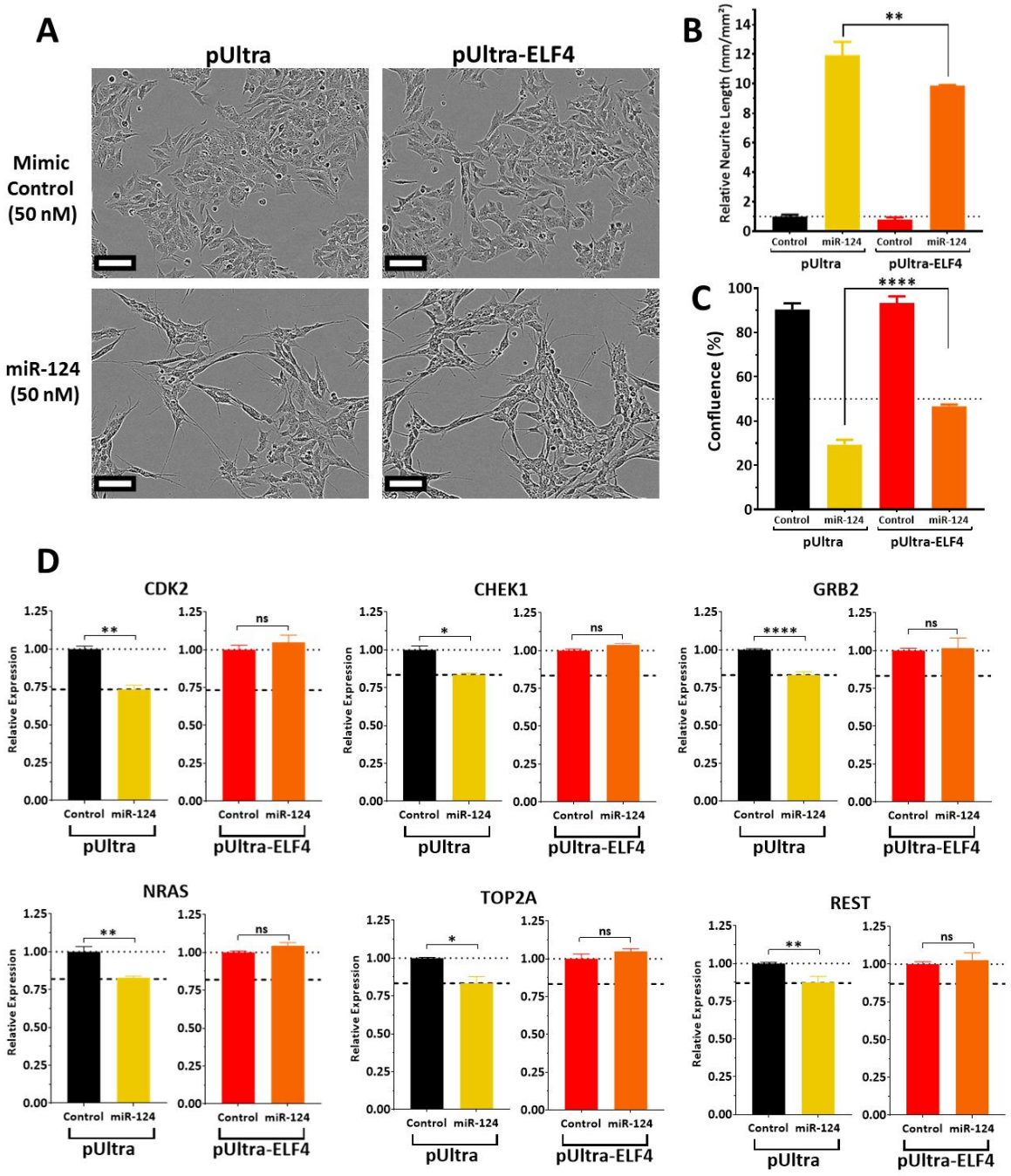

Figure S3. ELF4 ectopic expression antagonizes the effect of miR-124 on differentiation of KELLY cells. **(A)** KELLY cells were first infected with ELF4-expressing lentivirus or control then reverse transfected with miRNA mimics (miR-124 or control). 120 hours later, the impact on differentiation and confluence was measured. **(A)** Morphology of transfected cells (scale bar = 100 µm). **(B)** Quantification of neurite outgrowth of treated cells. **(C)** Confluence of treated cells. **D)** Control and ELF4-overexpressing cells were transfected with miRNA mimics; 48 hours later RNA was isolated and qRT-PCR was used to measure expression of a select group of co-targeted genes. Statistical significance of observed changes in neurite outgrowth was determined by a two-way ANOVA with Tukey's range test for multiple comparisons. A Student's t-test was used to assess differences in expression. * = $p<0.05$, **** = $p<0.0001$.

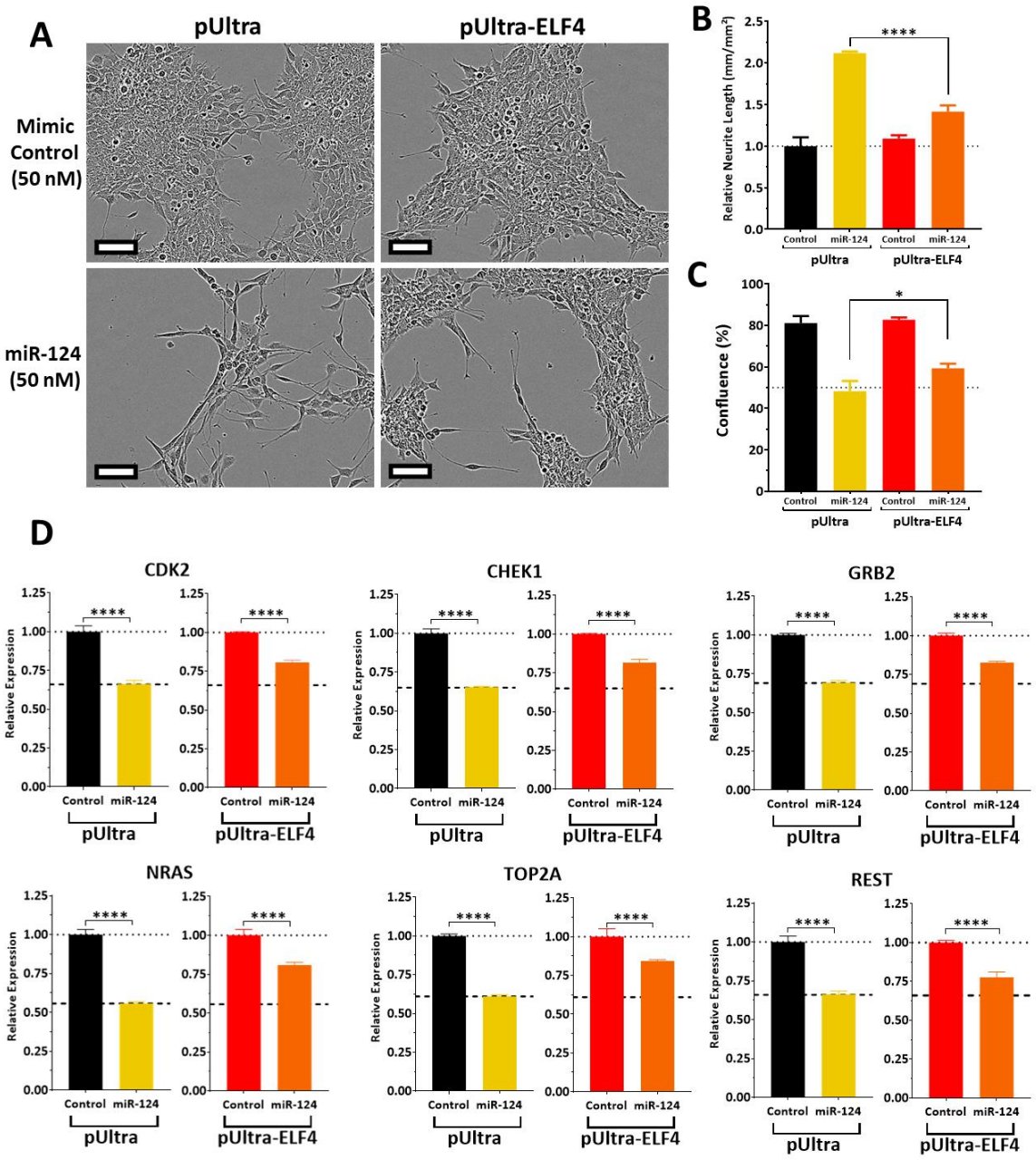

Figure S4. ELF4 ectopic expression antagonizes the effect of miR-124 on differentiation of SK-N-DZ cells. SK-N-DZ cells were first infected with ELF4-expressing lentivirus or control then reverse transfected with miRNA mimics (miR-124 or control). 120 hours later, the impact on differentiation and confluence was measured. **(A)** Morphology of transfected cells (scale bar = 100 µm); **(B)** Quantification of neurite outgrowth of treated cells. **(C)** Confluence of treated cells. **D)** Control and ELF4-overexpressing cells were transfected with miRNA mimics; 48 hours later RNA was isolated, and qRT-PCR was used to measure expression of a select group of co-targeted genes. Statistical significance of observed changes in neurite outgrowth was determined by a two-way ANOVA with Tukey's range test for multiple comparisons. A Student's t-test was used to assess differences in expression. * = $p<0.05$, **** = $p<0.0001$.

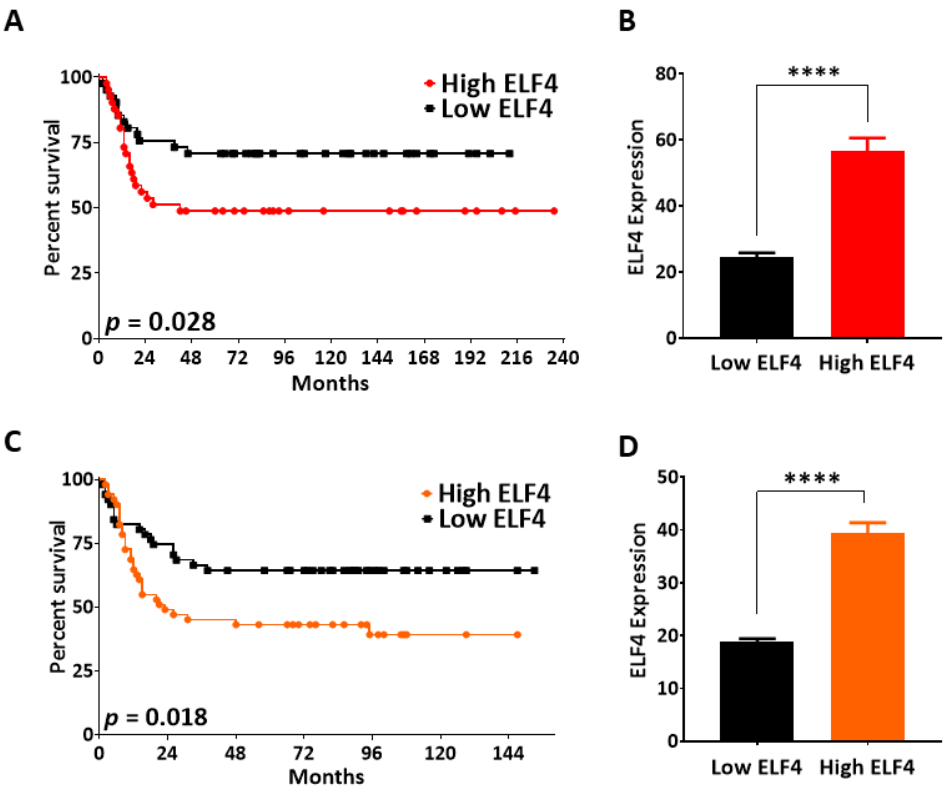

Figure S5. High ELF4 expression correlates with poor prognosis of neuroblastoma patients. (A) Overall survival (OS) of neuroblastoma patients from the Versteeg cohort stratified by ELF4 expression. **(B)** ELF4 expression levels (untransformed) in high and low groups. **(C)** Relapse-free survival for the Seeger cohort consisting of high-risk, non-amplified-MYCN neuroblastoma patients. Patients were stratified groups based on median ELF4 expression (untransformed). **(D)** ELF4 expression levels in the high and low groups. Survival analysis was performed using the log-rank (Mantel-Cox) test implemented in GraphPad Prism. Statistical significance of observed changes between the groups was determined by Student's t test. **** = p<0.0001.

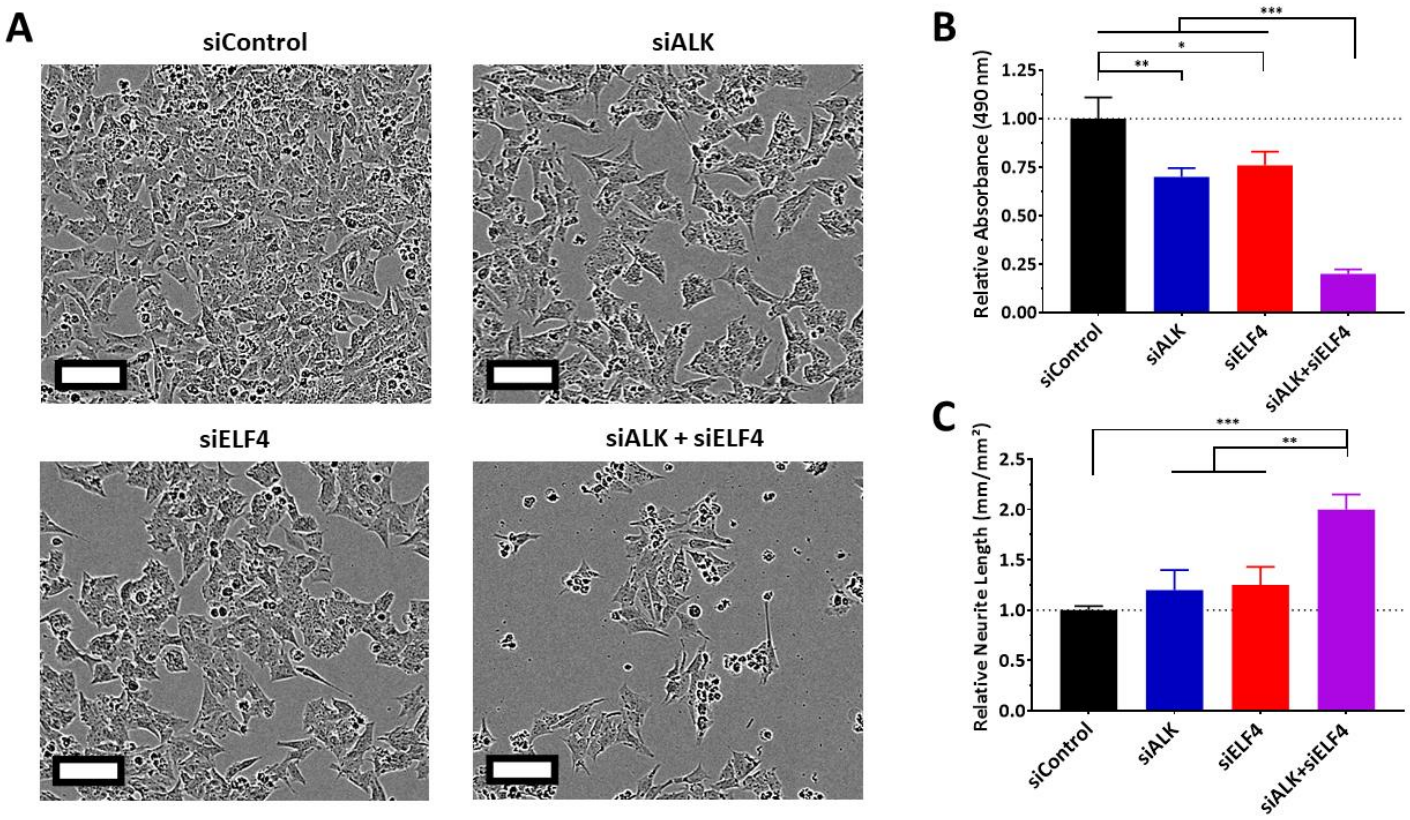

Figure S6. Synergistic effect of ALK and ELF4 inhibition on differentiation and proliferation of KELLY cells. **(A-C)** KELLY cells were reverse transfected with siRNAs (siALK, siELF4 and control). After 120 h: **(A)** Morphology was observed (scale bar = 100 µm); (**B**) Numbers of viable cells were assessed; and (**C**) Neurite length was measured. Statistical significance of observed changes was determined by Student's t test with a nominal threshold for significance of p = 0.05. * = $p<0.05$, ** = $p<0.01$, *** = $p<0.001$.

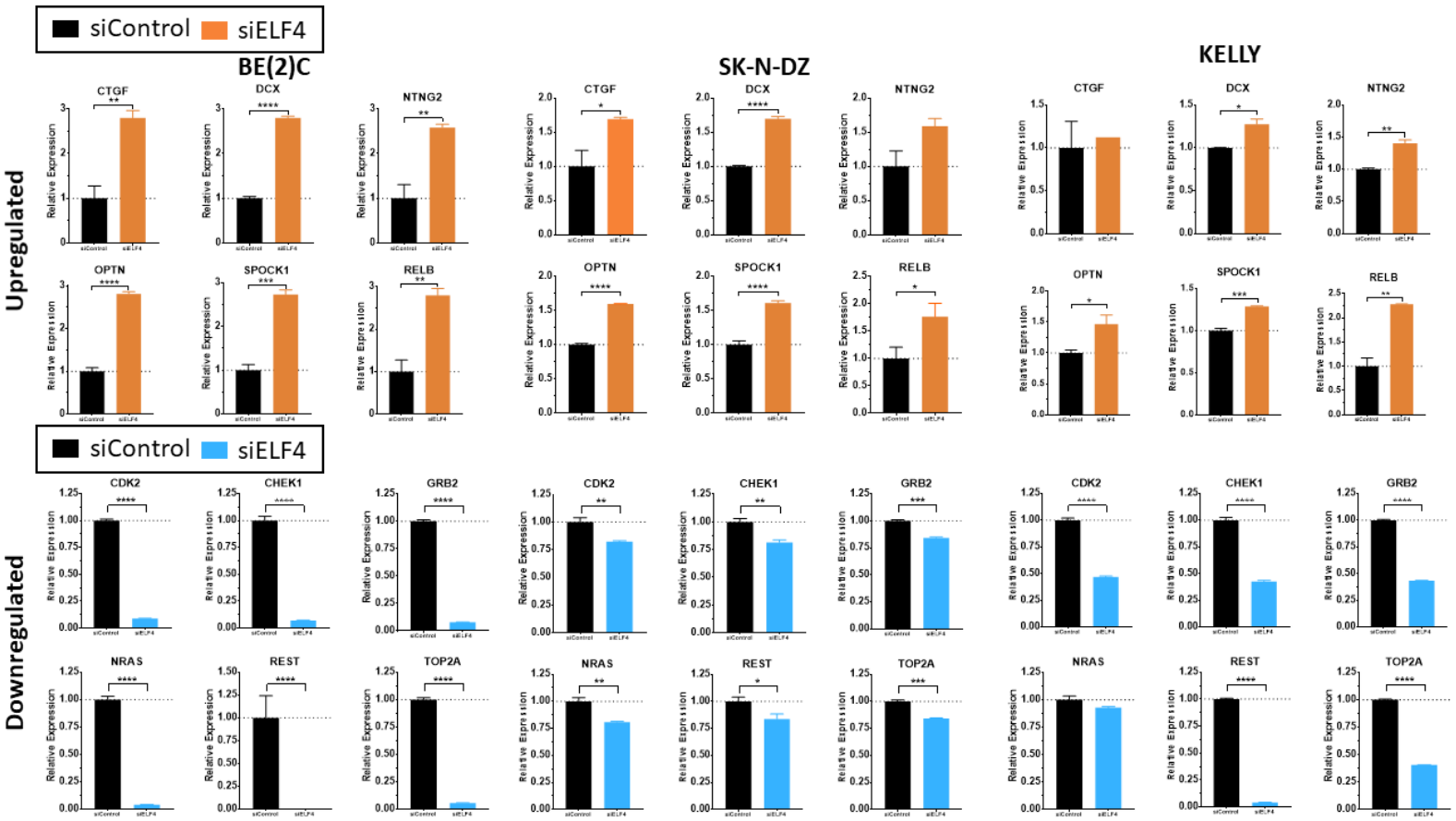

Figure S7. Validation of RNA-seq Results. (A-C) A group of downregulated genes identified in the RNA-seq analysis was validated by qRT-PCR in SK-N-BE(2)-C, SK-N-DZ, and KELLY cells 48 hours after reverse transfection with either siControl or siELF4. **(D-E)** A group of upregulated genes identified in the RNA-seq analysis was validated by qRT-PCR in the same cell lines following knockdown. Statistical significance of observed changes was determined by Student's t-test with a nominal threshold for significance of p = 0.05. * = p<0.05, ** = p<0.01, *** = p<0.001, **** = p<0.0001.