10.17305/bjbms.2022.8086

Biomolecules and Biomedicine
ISSN: 2831-0896 (Print) | ISSN: 2831-090X (Online)

Journal Impact Factor® (2021): 3.76
CiteScore® (2021): 5.2
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SUPPLEMENTAL DATA

Identification of RUNX1 and IFNGR2 as prognostic-relate biomarkers correlated with immune infiltration and subtype differentiation of low-grade glioma

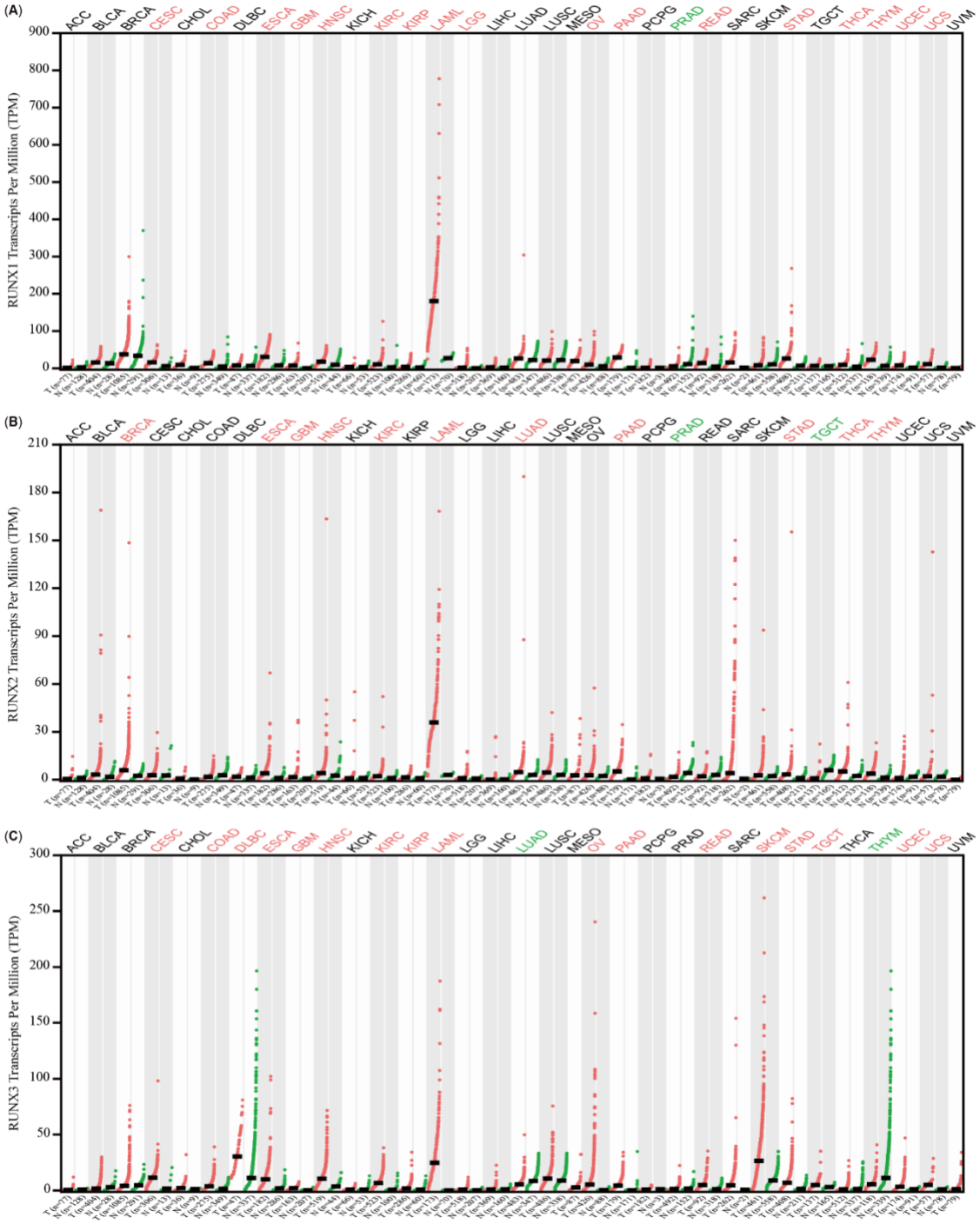

Figure S1. The transcription expression levels of the RUNXs in different types of human tumor and normal tissues based on GEPIA database. **(A)** RUNX1, **(B)** RUNX2, and **(C)** RUNX3. T: Tumor tissues, N: Normal tissues; RUNX: Runt-related transcription factors; GEPIA: Gene Expression Profiling Interactive Analysis

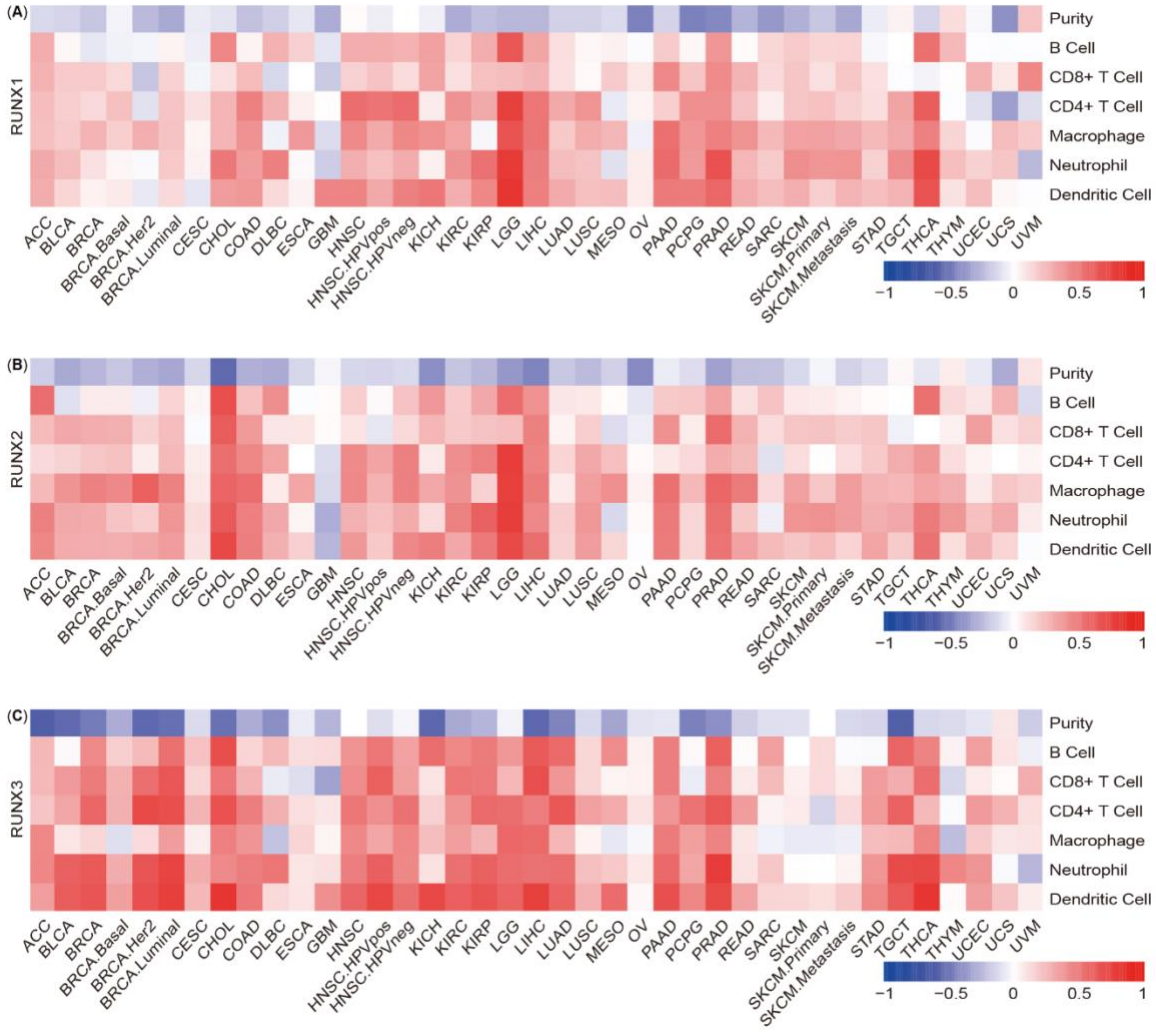

Figure S2. The correlations between RUNXs and immune infiltration (B cells, CD8$^+$ T cells, CD4$^+$ T cells, macrophages, neutrophils and dendritic cells) in multiple human cancers. (A) RUNX1, **(B)** RUNX2, and **(C)** RUNX3. RUNX: Runt-related transcription factors.

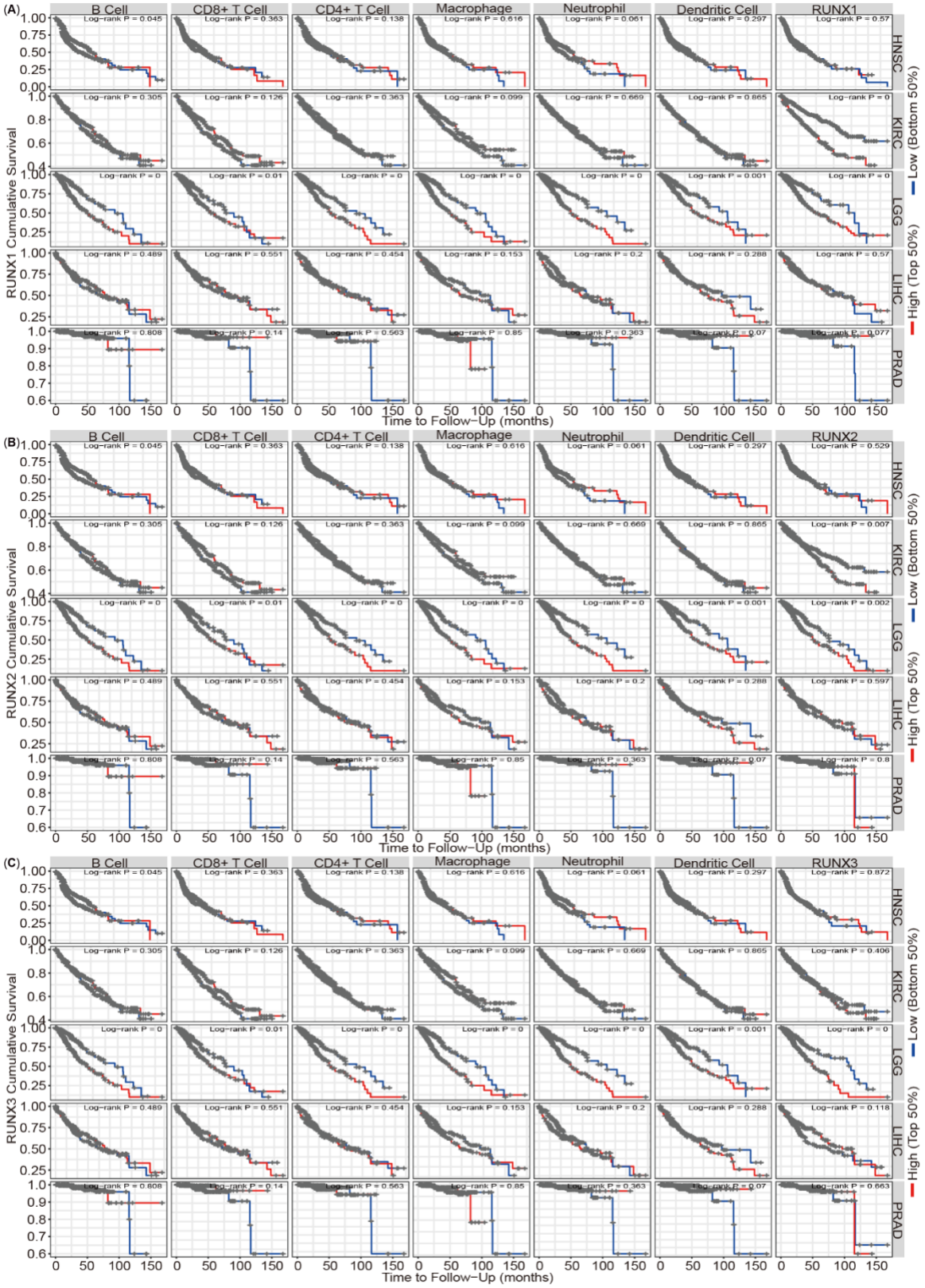

Figure S3. The relationship between RUNXs expression or immune infiltration and prognosis in HNSC, KIRC, LGG, LIHC and PRAD cancer types. (A) RUNX1, **(B)** RUNX2, and **(C)** RUNX3. HNSC: Head and Neck squamous cell carcinoma; KIRC: Kidney renal clear cell carcinoma; LGG: Low grade glioma; LIHC: Liver hepatocellular carcinoma; PRAD: Prostate adenocarcinoma; RUNX: Runt-related transcription factors.

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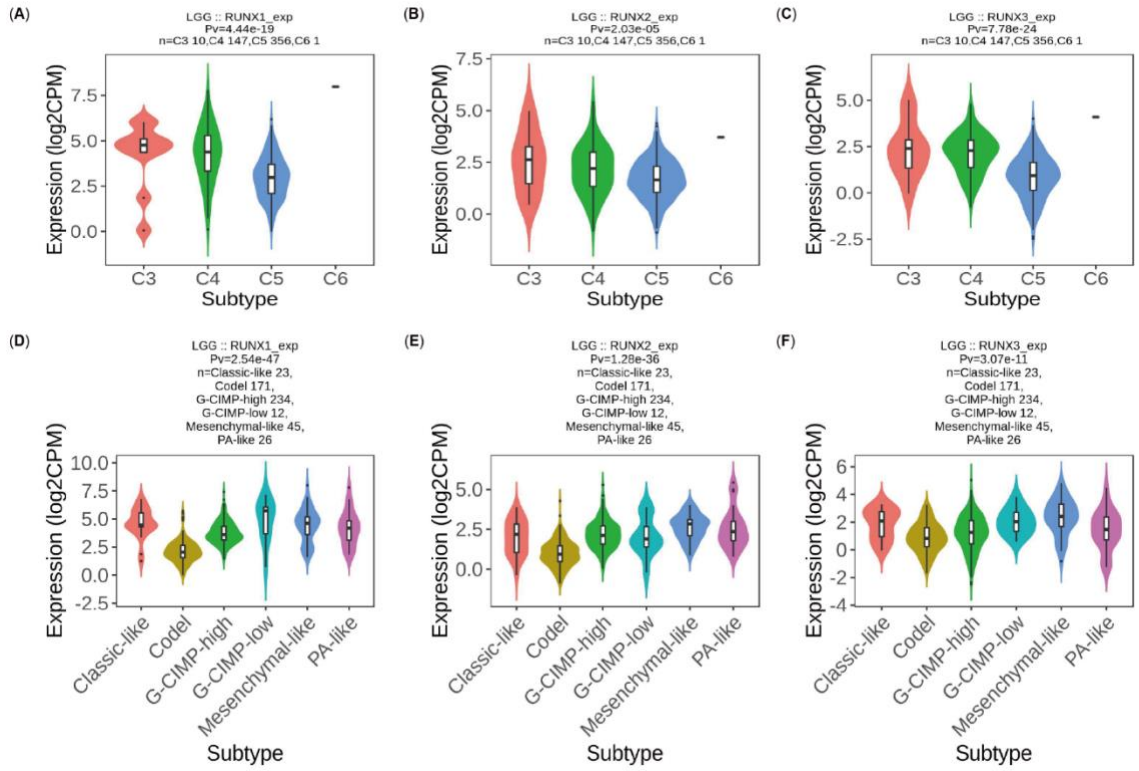

Figure S4. RUNXs were significantly correlated with the immune and molecular subtypes of LGG. Associations between RUNX1**(A)**, RUNX2 **(B)**, and RUNX3 **(C)** expressions and immune subtypes in LGG cancer based on TISIDB analysis. (C3: inflammatory; C4: lymphocyte depleted; C5: immunologically quiet; C6: TGF-β dominant). Associations between RUNX1**(D)**, RUNX2 **(E)**, and RUNX3 **(F)** expressions and molecular subtypes in LGG cancer based on TISIDB analysis. RUNX: Runt-related transcription factors; LGG: Low grade glioma. TGF-β: Tumor growth factor beta.

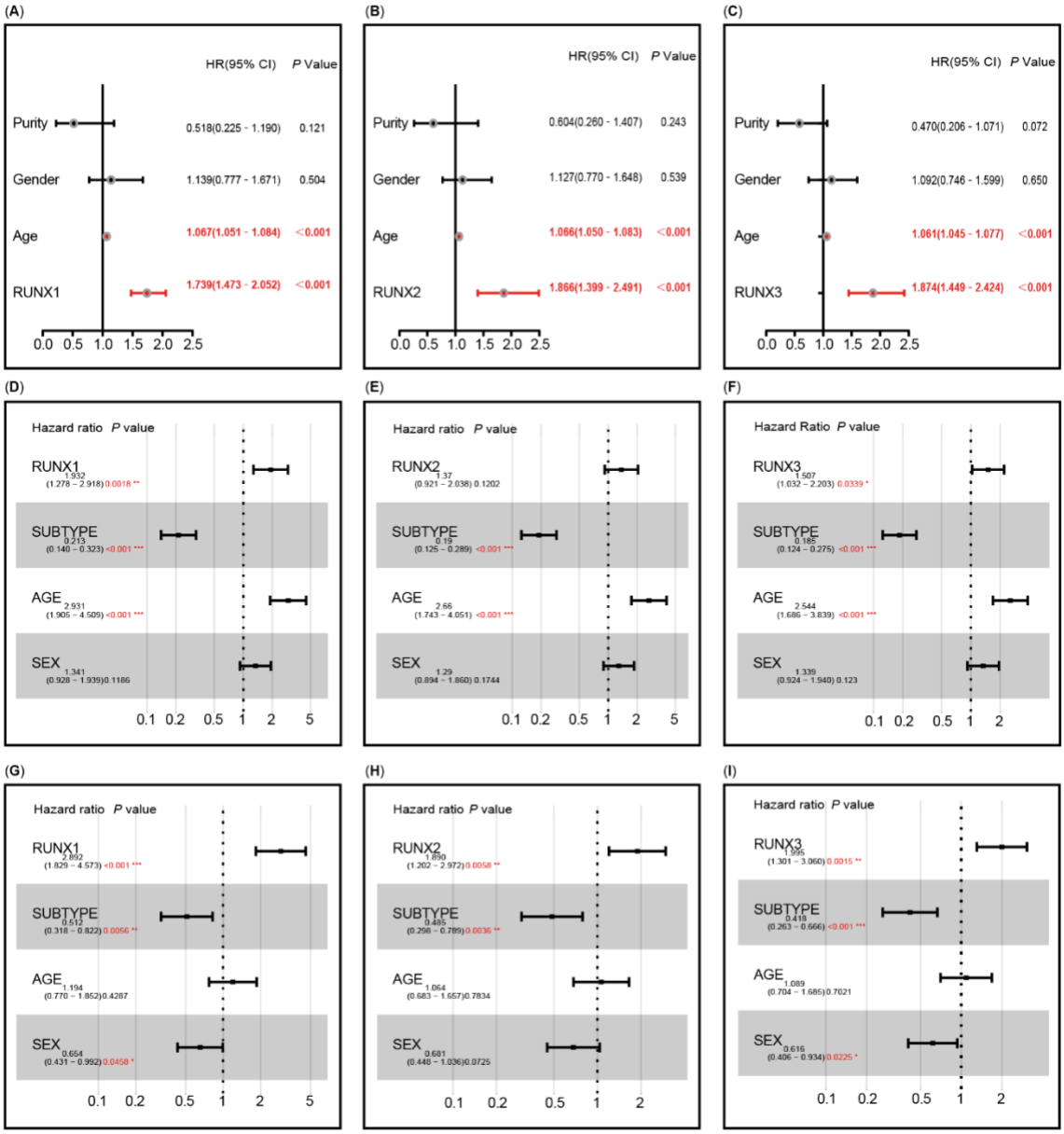

Figure S5. Multivariate Cox analysis of RUNXs expression and other clinical pathological factors. Age and RUNX1 **(A)**, RUNX2 **(B)**, and RUNX3 **(C)** expression are the independent prognostic factors based on TIMER analysis. **(D-F)** Multivariate Cox analysis of RUNXs expression and other clinical pathological factors (subtype, age, and sex) in LGG from TCGA dataset. **(G-I)** Multivariate Cox analysis of RUNXs expression and other clinical pathological factors (subtype, age, and sex) in LGG from CGGA dataset. RUNX: Runt-related transcription factors; TCGA: The Cancer Genome Atlas; CGGA: Chinese Glioma Genome Atlas; LGG: Low grade glioma.

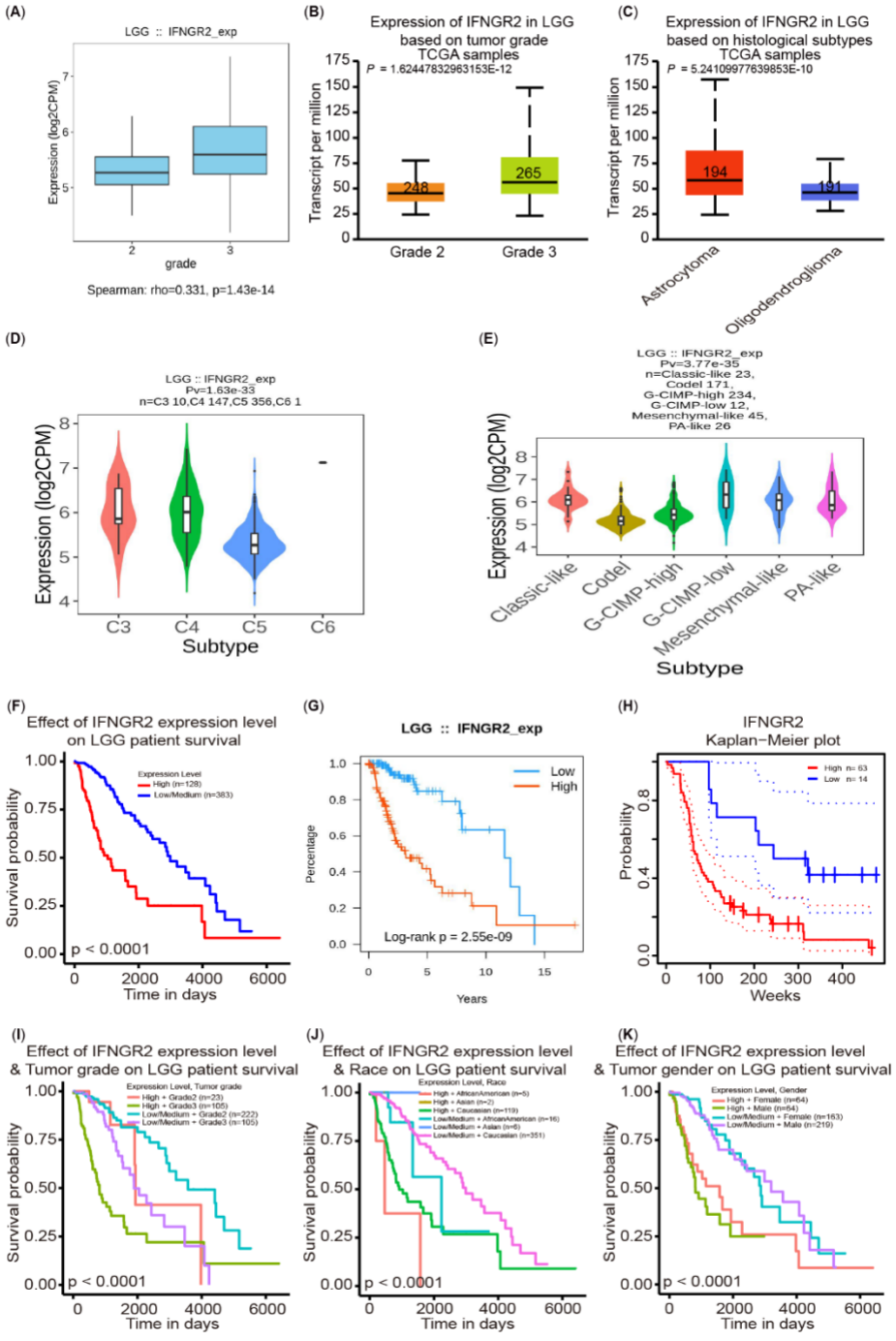

Figure S6. Comprehensive analysis of IFNGR2 in LGG. (A) The IFNGR2 transcription levels was significantly elevated in grade III gliomas compared with grade II LGG based on TISIDB analysis. **(B)** The IFNGR2 transcription level was significantly elevated in grade III gliomas compared with grade II LGG based on UACLAN database. **(C)** The different expression of IFNGR2 in different histological subtypes of LGG tissues based on UACLAN database. **(D)** Associations between IFNGR2 expression and immune subtypes in LGG cancer based on TISIDB analysis (C3: inflammatory; C4: lymphocyte depleted; C5: immunologically quiet; C6: TGF-β dominant). **(E)** Associations between IFNGR2 expression and molecular subtypes in LGG cancer based on TISIDB analysis. Correlation analysis between IFNGR2 expression and prognosis of LGG cancer based on the UACLAN **(F)**, TISIDB **(G)**, and PrognoScan databases **(H)**. **(I-K)** The expression of IFNGR2

related to survival probability based on tumor grades stages, races, genders of LGG cancers in UACLAN database. LGG: Low grade glioma; TGF-β: Tumor growth factor beta; IFNGR2: Interferon gamma receptor 2.

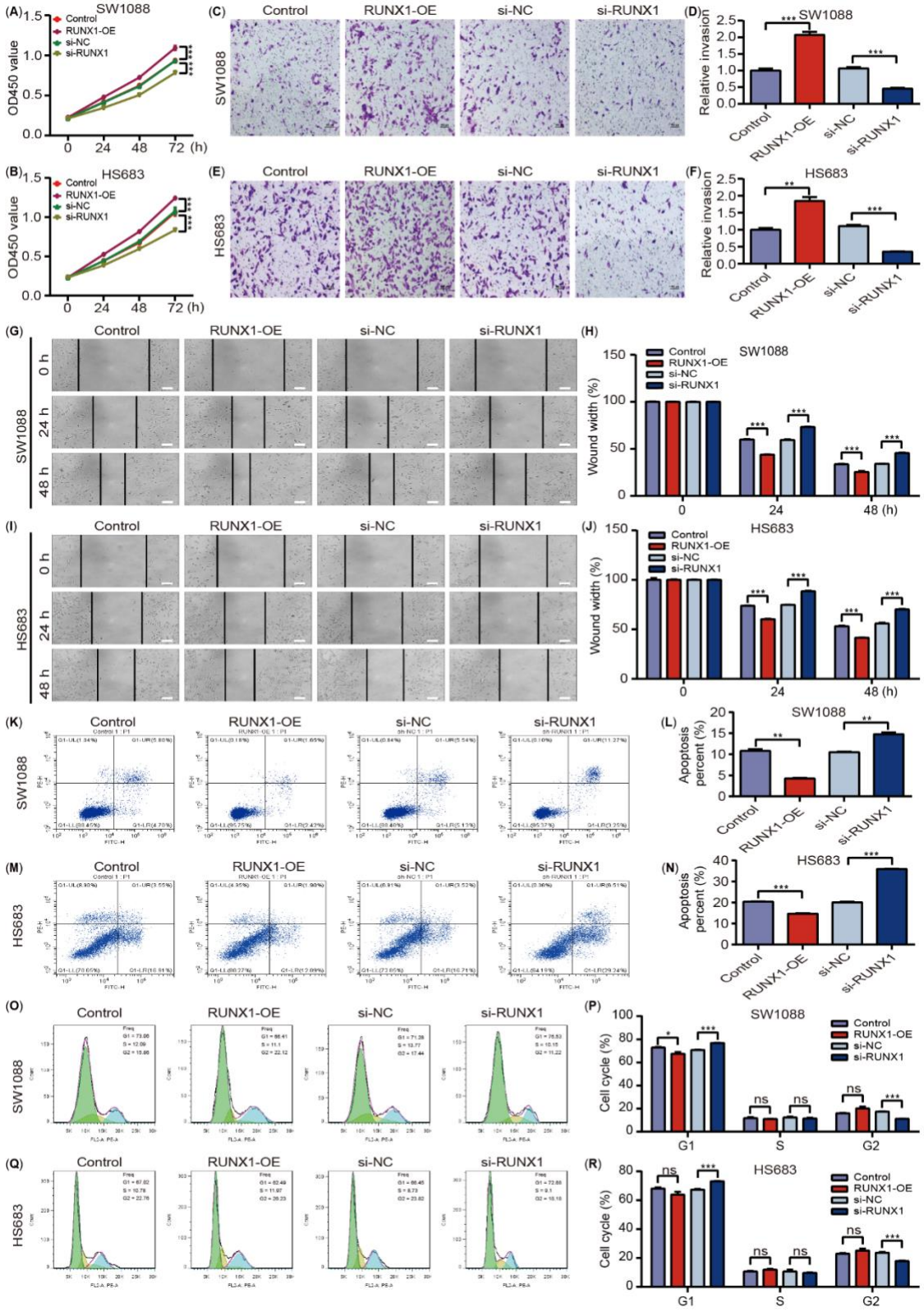

Figure S7. RUNX1 can monitor the proliferation, invasion, migration, and apoptosis in human LGG cell lines. (A and B) The CCK-8 assay was performed to detect the proliferative activity of SW1088 and HS683 cells in different groups (n = 3). **(C-F)** Transwell invasion assays were performed to detect the invasion abilities of SW1088 and HS683 cells in different groups and the related statistical analysis was performed (Scale bar: 400 μm; n = 3). **(G-J)** The scratch assays were

performed to detect the migration abilities of SW1088 and HS683 cells in different groups and the related statistical analysis was performed (Scale bar: 100 μm; n = 3). **(K-N)** Flow cytometry was performed to detect the apoptosis rate of SW1088 and HS683 cells in different groups and the related statistical analysis was performed (n = 3). **(O-R)** Flow cytometry was performed to detect the cell cycle of SW1088 and HS683 cells in different groups and the related statistical analysis was performed (n = 3). Data are shown as presented as mean ± SEM; $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; OE: Overexpression; NC; Negative control; si: Short interfering; LGG: Low grade glioma; CCK-8: Cell counting kit 8; RUNX: Runt-related transcription factors.

Table S1. Clinical characteristics of patients with LGG from TCGA database.

Characteristic	**No. of patients**
Number	514 (100%)
Age (years)	
Median	41
Range	14-87
Sex	
Male	285 (55.4%)
Female	228 (44.4%)
Unknown	1 (0.2%)
Race	
White	474 (92.2%)
Black or African American	21 (4.1%)
Asian	7 (1.4%)
American Indian or Alaska Native	1 (0.2%)
Unknown	11 (2.1%)
***IDH* mutation status**	
IDH wt	92 (17.9%)
IDH mut-codel	167 (32.5%)
IDH mut-non-codel	248 (48.2%)
Unknown	7 (1.4%)
Radio status	
Treated	296 (57.6%)
Un-treated	183 (35.6%)

Unknown	35 (6.8%)

Table S2. Clinical characteristics of patients with LGG from CGGA database.

Characteristic	**No. of patients**
Number	182 (100%)
Age (years)	
Median	39
Range	10-74
Sex	
Male	111 (61.0%)
Female	71 (39.0%)
Race	
Asian	182 (100%)
***IDH* mutation status**	
IDH wt	48 (26.4%)
IDH mut-codel	57 (31.3%)
IDH mut-non-codel	75 (41.2%)
Unknown	2 (1.1%)
Radio status	
Treated	142 (78.0%)
Un-treated	32 (17.6%)
Unknown	8 (4.4%)

Table S3. The siRNA sequence used in the present study.

Primer Name		Sequence(5'to3')
RUNX1-siRNA	Sense	ACTGTGATGGCTGGCAATGATG
	Anti-sense	TCTGTGGTAGGTGGCGACTTG
IFNGR2-siRNA	Sense	TTCAATGTCACTCTACGCCTTCG
	Anti-sense	TCAGCGATGTCAAAGGGAGAGG
BCL2-siRNA	Sense	ATCGCCCTGTGGATGACTGAG
	Anti-sense	CAGCCAGGAGAAATCAAACAGAGG
MCL1-siRNA	Sense	TTCAGCGACGGCGTAACAAAC
	Anti-sense	GAAGAACTCCACAAACCCATCCC
MMP9-siRNA	Sense	GGTCCTGGTGCTCCTGGTG
	Anti-sense	GCCTGTCGGTGAGATTGGTTC
VEGF-siRNA	Sense	GCCTTGCCTTGCTGCTCTAC
	Anti-sense	ATTCTGCCCTCCTCCTTCTGC
CCND1-siRNA	Sense	GCATCTACACCGACAACTCCATC
	Anti-sense	TTGTTCTCCTCCGCCTCTGG
CCND2-siRNA	Sense	TGGATGCTGGAGGTCTGTGAG
	Anti-sense	CTGAGGCTTGATGGAGTTGTCG
CCND3-siRNA	Sense	ACTGGATGCTGGAGGTATGTGAG
	Anti-sense	AGCGTGGTCGGTGTAGATGC