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Supplemental information

Selective regulation of chemosensitivity in glioblastoma by phosphatidylinositol 3-kinase beta

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Table S1. DepMap glioma cell lines re-annotated, related to Figure 1.

DepMap_ID	Cell_Line_Name	Tumor_Type		Note_in_Cellosaurus	Grades	PMID	MGMT_RNAseq	MGMT_Status*
		DepMap	Cellosaurus					
ACH-000323	**42-MG-BA**	Astrocytoma	glioblastoma		HGG	29740146		Deficient
ACH-000558	**A-172**	Glioblastoma	glioblastoma		HGG	22986464		Deficient
ACH-000269	**AM-38**	Glioblastoma	glioblastoma		HGG	25111384		Deficient
ACH-000464	**CAS-1**	Glioblastoma	glioblastoma		HGG		0.014	Deficient
ACH-000244	**DK-MG**	Glioblastoma	glioblastoma		HGG		0.070	Deficient
ACH-000738	**GB-1**	Glioblastoma	glioblastoma		HGG	33335215		Deficient
ACH-000756	**GI-1**	Glioblastoma	gliosarcoma		HGG		0.057	Deficient
ACH-000102	**GMS-10**	Glioblastoma	glioblastoma		HGG		0.070	Deficient
ACH-000027	**GOS-3**	Glioblastoma	glioblastoma		HGG		0.029	Deficient
ACH-000479	**KNS-81**	Glioblastoma	glioblastoma		HGG		0.111	Deficient
ACH-000631	**KS-1**	Astrocytoma	glioblastoma		HGG		0.251	Deficient
ACH-000595	**LN-229**	Glioblastoma	glioblastoma		HGG	25111384		Deficient
ACH-000673	**LN-443**	Glioblastoma	glioblastoma		HGG		0.000	Deficient
ACH-000676	**LN-464**	Glioblastoma	glioblastoma		HGG		0.124	Deficient
ACH-000152	**M059K**	Glioblastoma	glioblastoma		HGG	25111384		Deficient
ACH-001611	**NP 8**	Glioblastoma	glioblastoma		HGG		0.000	Deficient
ACH-001622	**Onda 7**	Glioblastoma	glioblastoma		HGG		0.000	Deficient
ACH-000376	**SF-295**	Glioblastoma	glioblastoma		HGG	23958055		Deficient
ACH-000609	**SF126**	Astrocytoma	glioblastoma		HGG	25111384		Deficient
ACH-000273	**SF539**	Glioma	gliosarcoma		HGG		0.163	Deficient
ACH-000504	**SNB75**	Glioma	glioblastoma		HGG		0.070	Deficient
ACH-000368	**SNU-1105**	Glioblastoma	glioblastoma		HGG		0.043	Deficient
ACH-000075	**U-87 MG**	Astrocytoma	glioblastoma		HGG	23958055		Deficient
ACH-000283	**A1207**	Glioblastoma	glioblastoma		HGG	22570426		Positive
ACH-001329	**ANGM-CSS**	Glioblastoma	glioblastoma		HGG		3.441	Positive
ACH-000098	**GAMG**	Glioblastoma	glioblastoma		HGG		0.556	Positive
ACH-000231	**KALS-1**	Glioblastoma	glioblastoma		HGG		3.995	Positive
ACH-000445	**KNS-60**	Glioblastoma	glioblastoma		HGG		2.799	Positive
ACH-000819	**LN-18**	Glioblastoma	glioblastoma		HGG	25111384		Positive
ACH-000634	**LN-340**	Glioblastoma	glioblastoma		HGG	32361246		Positive
ACH-000455	**LN-428**	Glioblastoma	glioblastoma		HGG	32982428		Positive
ACH-000215	**LN382**	Glioblastoma	glioblastoma		HGG		3.350	Positive
ACH-001605	**no.10**	Glioblastoma	glioblastoma		HGG		3.798	Positive
ACH-001623	**Onda 8**	Glioblastoma	glioblastoma		HGG		3.454	Positive
ACH-000887	**SF-172**	Glioblastoma	glioblastoma		HGG		3.446	Positive
ACH-000289	**SNU-466**	Glioblastoma	glioblastoma		HGG		3.981	Positive
ACH-000543	**SNU-489**	Glioblastoma	glioblastoma		HGG		1.982	Positive
ACH-000370	**SNU-626**	Glioblastoma	glioblastoma		HGG		3.901	Positive
ACH-000571	**T98G**	Glioblastoma	glioblastoma		HGG	25111384		Positive
ACH-000036	**U343**	Glioblastoma	glioblastoma		HGG	12503076		Positive
ACH-000469	**YH-13**	Glioblastoma	glioblastoma		HGG	25111384		Positive
ACH-000570	**YKG1**	Glioblastoma	glioblastoma		HGG		4.146	Positive
ACH-001125	MOGGCCM	Glioma	Anaplastic astrocytoma		HGG		No data**	
ACH-001126	MOGGUVW	Glioma	Anaplastic astrocytoma		HGG		No data	
ACH-001606	no.11	Glioblastoma	glioblastoma		HGG		No data	
ACH-002223	D-245MG	Glioblastoma	glioblastoma		HGG		No data	
ACH-002224	D-247MG	Glioblastoma	Gliosarcoma		HGG	29164620	No data	Deficient
ACH-002225	D-263MG	Glioblastoma	glioblastoma		HGG		No data	
ACH-002226	D-336MG	Glioblastoma	gliosarcoma		HGG		No data	
ACH-002227	D-392MG	Glioblastoma	glioblastoma		HGG		No data	
ACH-002228	D-423MG	Glioblastoma	glioblastoma		HGG		No data	
ACH-002229	D-502MG	Glioblastoma	glioblastoma		HGG		No data	
ACH-002230	D-542MG	Glioblastoma	glioblastoma		HGG		No data	
ACH-002231	D-566MG	Glioblastoma	glioblastoma		HGG		No data	
ACH-002259	KNS-81-FD	Glioblastoma	glioblastoma		HGG		No data	
ACH-002268	LN-405	Glioblastoma	glioblastoma		HGG		No data	
ACH-000622	KNS-42	Glioblastoma	glioblastoma	Misidentified/contaminated**	HGG	20935218		Deficient
ACH-001624	Onda 9	Glioblastoma	glioblastoma	Misidentified	HGG		3.379	Positive
ACH-002257	KINGS-1	Astrocytoma, anaplastic	Anaplastic astrocytoma	Misidentified	HGG			
ACH-000623	SNU-201	Glioblastoma	glioblastoma	Contaminated	HGG		0.971	Positive
ACH-000883	SW 1783	Astrocytoma	Anaplastic astrocytoma	Contaminated	HGG	24368590		Positive
ACH-000863	DBTRG-05MG	Glioblastoma	Anaplastic astrocytoma	Contaminated	HGG	25111384		Deficient

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ACH-000137	**8-MG-BA**	Astrocytoma	astrocytoma		II/III	29740146		Deficient
ACH-001016	**Becker**	Glioma	astrocytoma		II/III		0.163	Deficient
ACH-000389	**H4**	Glioma	astrocytoma		II/III	29749471		Deficient
ACH-000067	**Hs 683**	Oligodendroglioma	oligodendroglioma		II/III	20517307		Deficient
ACH-000128	**LN-319**	Astrocytoma	astrocytoma		II/III	23533755		
ACH-000200	**NMC-G1**	Glioblastoma	astrocytoma		II/III		0.124	Deficient
ACH-000655	**SF268**	Glioma	astrocytoma		II/III	20935218		Deficient
ACH-000807	**SNU-738**	Oligodendroglioma	oligodendroglioma		II/III		0.084	Deficient
ACH-000437	**SW 1088**	Astrocytoma	astrocytoma		II/III	***		Deficient
ACH-000592	**TM-31**	Glioma	astrocytoma		II/III		0.014	Deficient
ACH-000232	**U-251 MG**	Astrocytoma	astrocytoma		II/III	22986464		Deficient
ACH-000126	**KG-1-C**	Oligodendroglioma	oligodendroglioma		II/III		4.084	Positive
ACH-000591	**LN235**	Astrocytoma	astrocytoma		II/III	30140041		Positive
ACH-000040	**U-118 MG**	Astrocytoma	astrocytoma		II/III	26110872	1.655	Positive
ACH-001000	1321N1	Astrocytoma	astrocytoma		II/III		No data	
ACH-001118	M059J	Glioma	astrocytoma		II/III		No data	
ACH-001198	SNB19	Glioblastoma	astrocytoma		II/III	34201219	No data	Deficient
ACH-001214	U138MG	Glioma	astrocytoma		II/III	20935218	No data	Positive
ACH-002304	SK-MG-1	Astrocytoma	astrocytoma		II/III		No data	
ACH-000328	LN-215	Glioblastoma	astrocytoma	Misidentified	II/III		0.029	Deficient
ACH-002269	LNZTA3WT4	Astrocytoma	astrocytoma	Contaminated	II/III			
ACH-000208	U-178	Glioblastoma	astrocytoma	Contaminated	II/III		0.070	Deficient
ACH-000329	CCF-STTG1	Astrocytoma	astrocytoma	Contaminated	II/III	***		Deficient
ACH-000760	LNZ308	Glioblastoma	astrocytoma	Contaminated	II/III		0.014	Deficient
DepMap_ID	**Cell line_Name**	**Tissue_Type DepMap**	**Tissue_Type Cellosaurus**					
ACH-000064	SALE	lung	lung		Normal		2.949	
ACH-000170	PrEC LH	prostate	prostate epithelial cells		Normal		2.842	
ACH-000494	OELE	ovary	ovary		Normal		3.793	
ACH-000642	HMEL	breast	breast		Normal		0.722	
ACH-001142	NHAHTDD	central_nervous_system	central_nervous_system Epithelial cells		Normal		2.753	
ACH-001207	TIG-3 TD	fibroblast	lung fibroblast		Normal		4.184	
ACH-001310	HA1E	kidney	kidbey epithelial cells		Normal		3.353	
ACH-002463	RPE1-ss77	eye	Retinal pigment epithelial cell		Normal		0.000	

Notes:

*Determination of MGMT status:

1. Search literature and record MGMT status based on immunoblotting or sequencing of MGMT promoter methylation.
2. MGMT RNAseq data from DepMap: < 0.5, MGMT-deficient; >0.5, MGMT-positive.
3. MGMT-deficient is defined as methylated MGMT promoter and/or low levels of MGMT mRNAs or proteins.
4. MGMT-positive is defined as unmethylated MGMT promoter and high levels of MGMT mRNAs or proteins.

**Exclusion of cell lines:

1. No RNAseq or other data available at DepMap.
2. Misidentified or contaminated as noted in Cellosaurus.

***Citation of MGMT status in SW 1088 or CCF-STTG1:
https://slidetodoc.com/oncology-nursing-2016-temozolomide-resistance-in-glioblastoma-multiforme/

DepMap glioma cell lines used in this study are in bold.

Table S2. Immunohistochemical staining of gliomas, related to Figure 1.

Patient ID	Gender	Age	Tumor Grade	p110α (HPA009985)	p110β (CAB031938)	p110δ (CAB015420)	p110γ (HPA069976)	PTEN (HPA031335)	MGMT (HPA069497)
3	Male	68	HGG				ND (0%)		Low
1537	Male	77	HGG	ND (0%)		ND (0%)			
1587	Female	36	HGG	Low (25%)	Medium (75%)	ND (0%)	ND (0%)	Medium	ND
1608	Male	79	HGG				ND (0%)		ND
1627	Male	33	HGG	Low (25%)		ND (25%)			
1644	Female	63	HGG				ND (0%)		ND
2522	Male	61	HGG		Medium (75%)			ND	
2527	Male	72	HGG		Medium (75%)				
2726	Male	60	HGG	Medium (50%)		ND (0%)			
2728	Female	74	HGG		Low (50%)			Low	
2750	Male	47	HGG	Low (25%)		ND (25%)			
2790	Male	47	HGG		Medium (75%)				
3091	Male	71	HGG	High (75%)		ND (25%)			
3092	Male	48	HGG	Low (25%)		Low (50%)			
3203	Male	53	HGG		Medium (75%)			Low	
3226	Male	56	HGG		Medium (75%)		ND (0%)		ND
3241	Female	58	HGG	Medium (25%)		ND (25%)			
5479	Female	71	HGG				ND (0%)		ND
122	Female	32	LGG				ND (0%)		ND
156	Male	58	LGG			ND (0%)			
1613	Female	39	LGG		Medium (75%)				
2529	Female	37	LGG	ND (0%)					
2634	Female	45	LGG				ND (0%)		ND
2868	Male	27	LGG				ND (0%)		ND
2873	Male	58	LGG				ND (0%)		Low
3023	Female	1	LGG		ND (0%)				
3120	Male	38	LGG	Low (25%)		ND (0%)			
3137	Male	77	LGG	Low (25%)		ND (0%)			
3174	Female	22	LGG	Low (50%)	Low (50%)	ND (0%)		Low	
3365	Male	65	LGG		Low (25%)			ND	

Notes: Data were retrieved from THPA. Antibody catalog numbers starting with HPA or CAB and quantities of each staining are included. HGG: high-grade glioma (grade III/IV); LGG: low-grade glioma (grade I/II). ND: not detected.

Table S3. IC50s and AUCs of GBM cell lines, related to Figure 2.

Cell_Line	Grade	MGMT_Status	IC50s_of_TMZ (μM)	Log10IC50s	Source
42-MG-BA	IV	Deficient	123.755	2.093	Sanger GDSC1
A-172	IV	Deficient	669.400	2.826	Sheng lab
AM-38	IV	Deficient	260.073	2.415	Sanger GDSC1
CAS-1	IV	Deficient	649.190	2.812	Sanger GDSC1
DK-MG	IV	Deficient	560.638	2.749	Sanger GDSC1
GB-1	IV	Deficient	141.686	2.151	Sanger GDSC1
GI-1	IV	Deficient	41.641	1.620	Sanger GDSC1
KS-1	IV	Deficient	140.676	2.148	Sanger GDSC1
LN-229	IV	Deficient	612.900	2.787	Sheng lab
SF-295	IV	Deficient	930.800	2.969	Sheng lab
SF126	IV	Deficient	276.365	2.441	Sanger GDSC1
SF539	IV	Deficient	91.228	1.960	Sanger GDSC1
SNB75	IV	Deficient	684.412	2.835	Sanger GDSC1
U-87 MG	IV	Deficient	2297.000	3.361	Sheng lab
GAMG	IV	Positive	304.115	2.483	Sanger GDSC1
KALS-1	IV	Positive	388.582	2.589	Sanger GDSC1
LN-18	IV	Positive	5819.000	3.765	Sheng lab
no.10	IV	Positive	374.870	2.574	Sanger GDSC1
T98G	IV	Positive	1048.000	3.020	Sheng lab
YH-13	IV	Positive	342.779	2.535	Sanger GDSC1
YKG1	IV	Positive	358.998	2.555	Sanger GDSC1
8-MG-BA	II/III	Deficient	48.330	1.684	Sanger GDSC1
Becker	II/III	Deficient	206.917	2.316	Sanger GDSC1
H4	II/III	Deficient	213.604	2.330	Sanger GDSC1
Hs 683	II/III	Deficient	339.948	2.531	Sanger GDSC1
NMC-G1	II/III	Deficient	328.046	2.516	Sanger GDSC1
SF268	II/III	Deficient	328.700	2.517	Sheng lab
SW 1088	II/III	Deficient	172.264	2.236	Sanger GDSC1
U-251 MG	II/III	Deficient	151.000	2.179	Sheng lab
U-118 MG	II/III	Positive	776.085	2.890	Sanger GDSC1

Notes: MGMT status of cell lines was from Supplemental Table S1. TMZ IC50s were collected either from the DepMap Sanger GDSC1 dataset or acquired by the Sheng lab by measuring dose-dependent effect of TMZ in these cells.

Table S4. Datasets used for survival analyses, related to Figure 3.

Datasets	Primary/IDH-wt GII/III		Primary/IDH-wt GBM	
	MGMT-deficient	MGMT-positive	MGMT-deficient	MGMT-positive
TCGA	4	54	21	29
CGGA	8	20	16	17
Gravendeel			13	12
Rembrandt			17	13
LeeY			27	30
Murat			17	0
Ivy			3	0
Grzmil			2	4
Philips			14	14
Vital			7	7
Ducray			2	12
Gorovets	3	10		
Kamoun	3	9		
POLA	0	3		

Notes: Numbers of samples from the above datasets are shown.

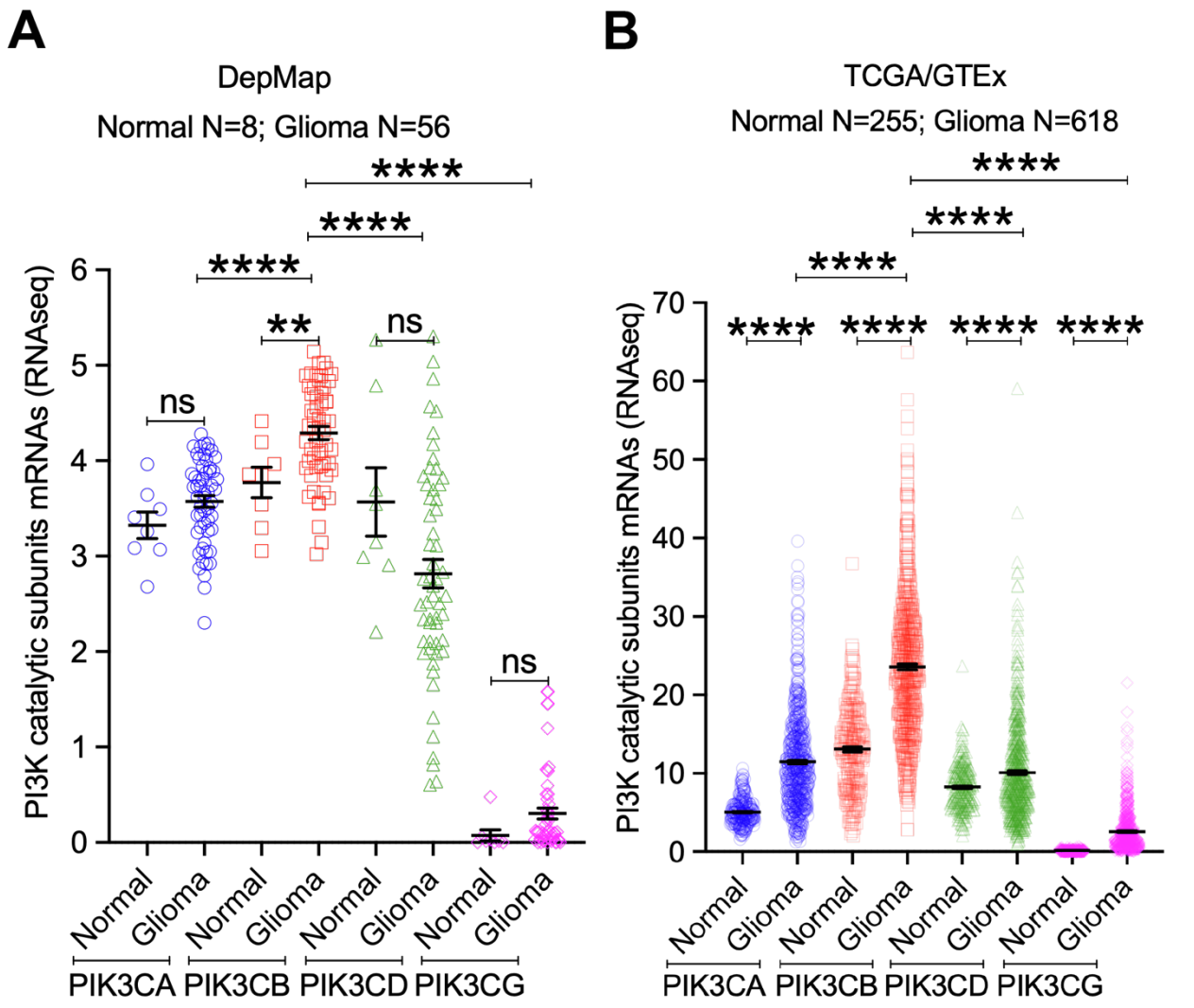

Figure S1. Expression of PI3K kinases in normal tissues and gliomas, related to Figure 1. RNAseq data were retrieved from DepMap, TCGA, and GTEx. (**A**) Comparison of PI3K mRNAs between DepMap normal cell lines (Normal) and glioma cell lines (Glioma). (**B**) Comparison of PI3K mRNAs between GTEx cerebral cortex (Normal) and TCGA gliomas (Glioma). Error bars are standard errors. Student *t*-test or one-way ANOVA was used to determine statistical significance. Sample sizes (N) of each group are shown. ns: not significant; **: $P < 0.01$; ****: $P < 0.0001$.

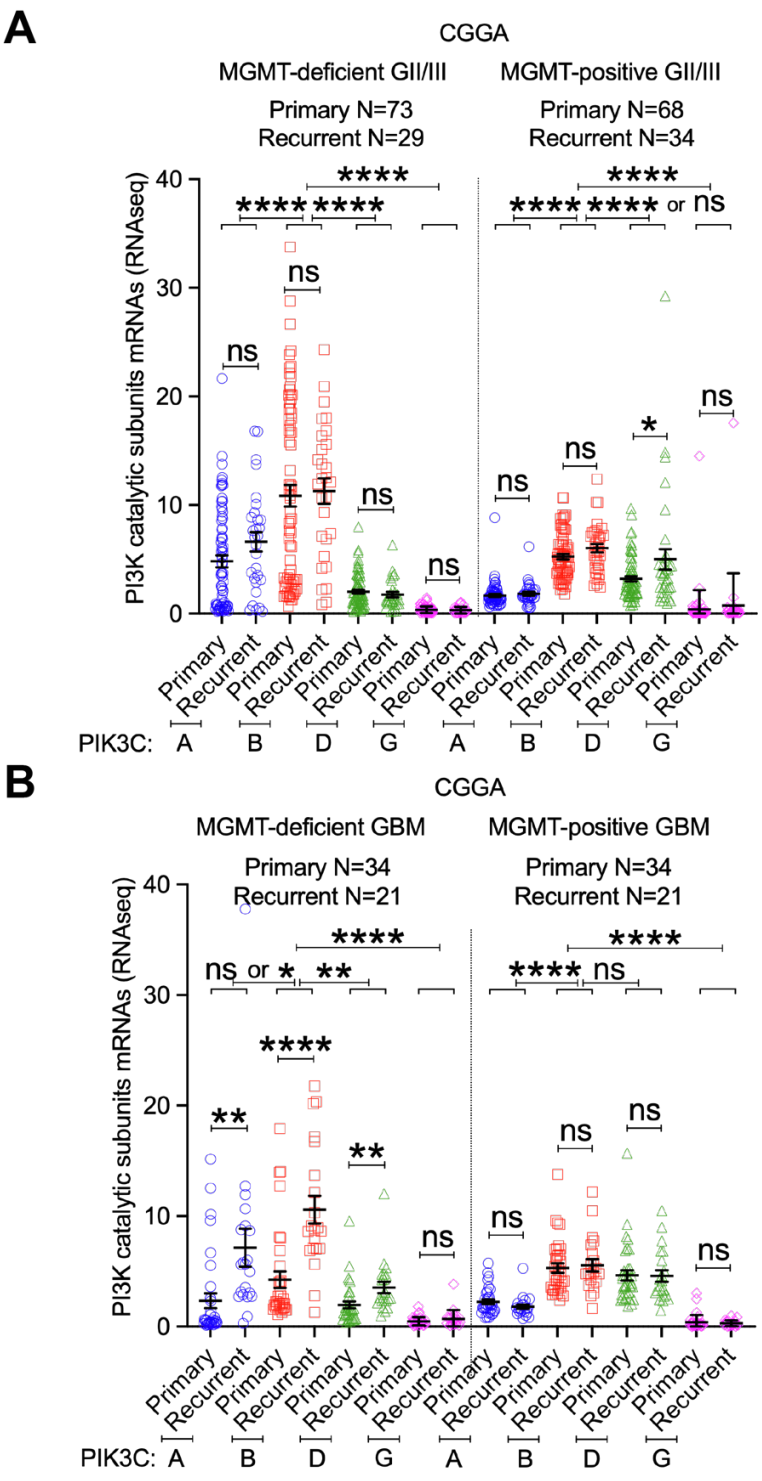

Figure S2. Expression of PI3K kinases in primary and recurrent gliomas, related to Figure 1. RNAseq data were retrieved from CGGA. mRNA levels of PI3K kinases in primary or recurrent grade II/III (GII/III) glioma (**A**) or grade IV GBM (**B**) are shown. Their levels were also compared in MGMT-deficient or MGMT-positive gliomas. Sample sizes (N) are shown. Error bars are standard errors. Student *t*-test or one-way ANOVA was used to determine statistical significance. ns: not significant; *: $P < 0.05$; **: $P < 0.01$; ****: $P < 0.0001$.

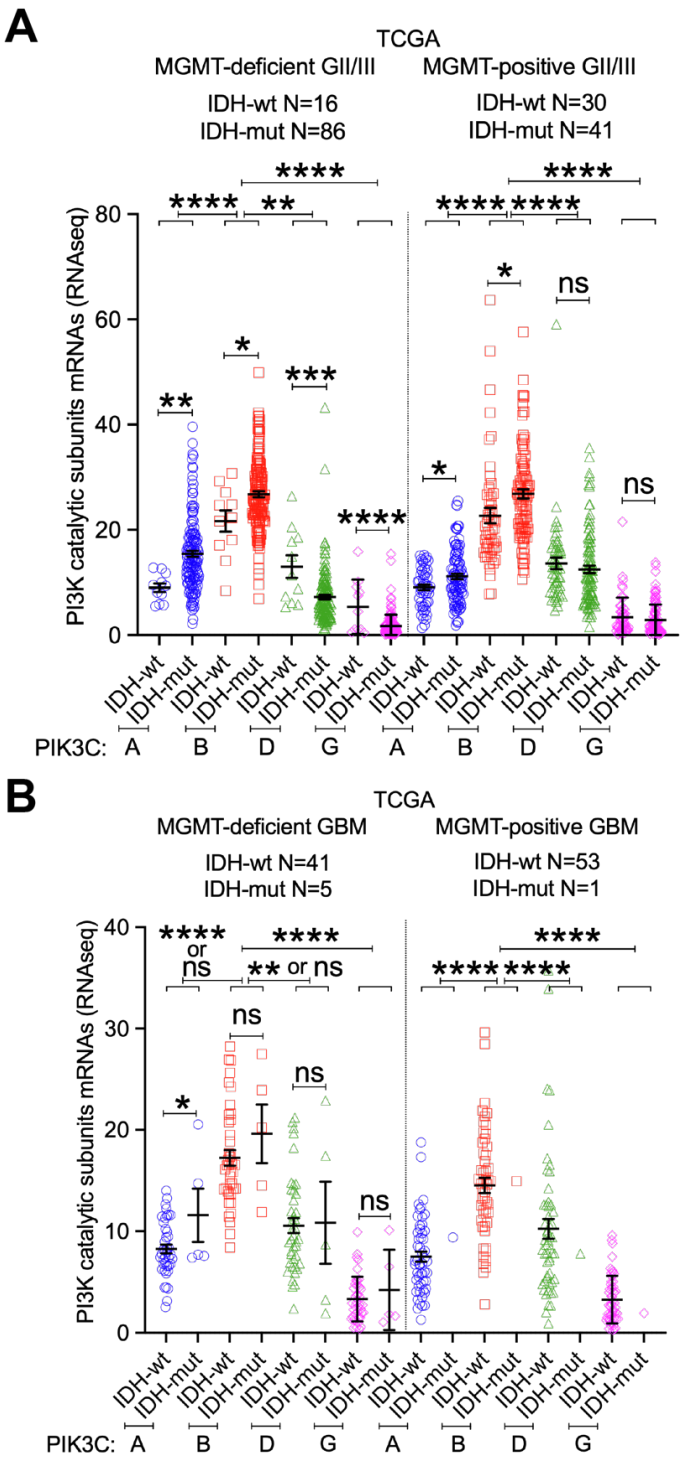

Figure S3. Expression of PI3K kinases in IDH wild-type or mutant gliomas, related to Figure 1. RNAseq data were retrieved from TCGA. mRNA levels of PI3K kinases in IDH wild-type (wt) or IDH mutant (mut) grade II/III (GII/III) glioma (**A**) or grade IV GBM (**B**) are shown. Their levels were also compared in MGMT-deficient or MGMT-positive gliomas. Sample sizes (N) are shown. Error bars are standard errors. Student *t*-test or one-way ANOVA was used to determine statistical significance. ns: not significant; *: $P < 0.05$; **: $P < 0.01$; ****: $P < 0.0001$.

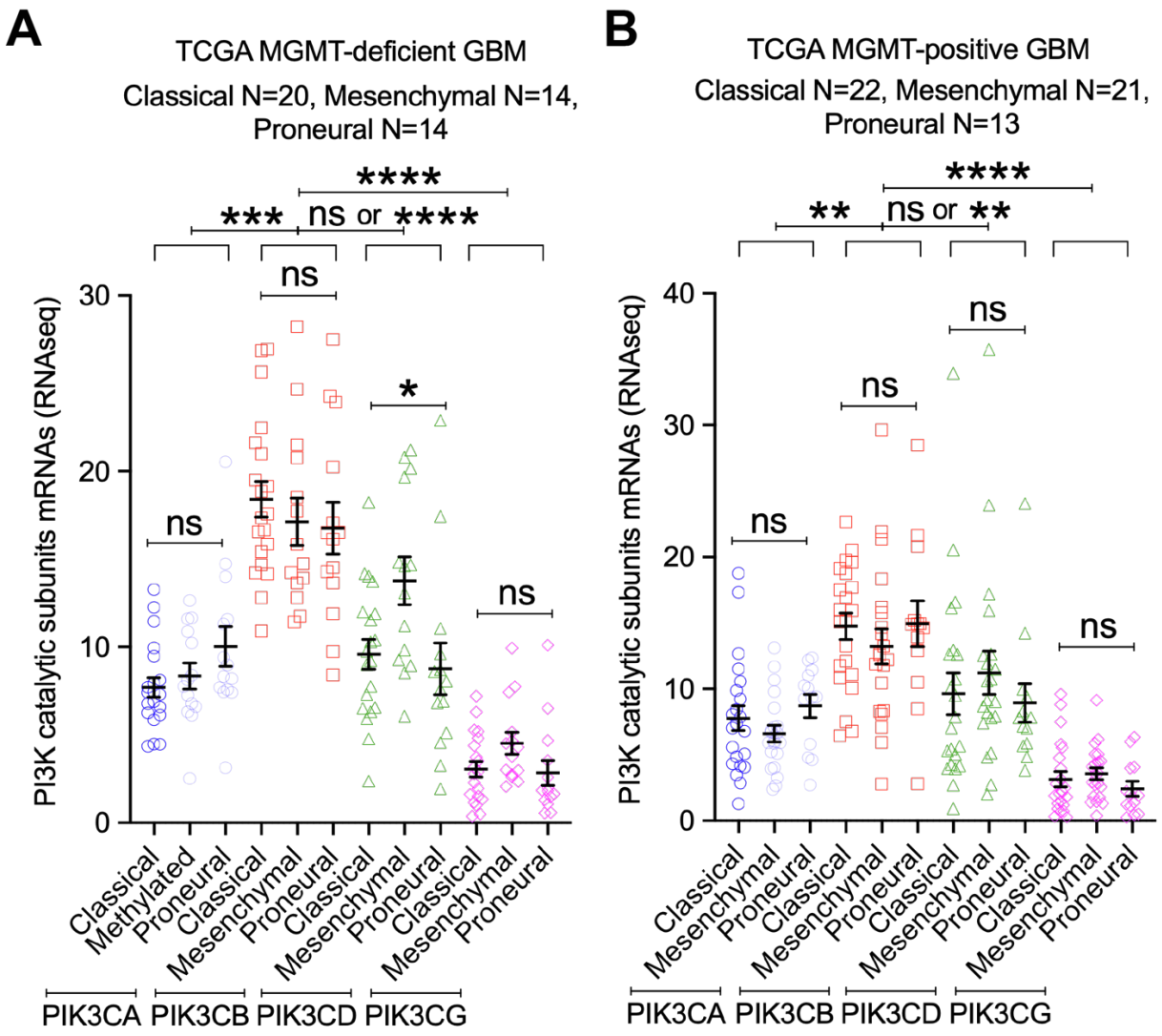

Figure S4. Expression of PI3K kinases in GBM subtypes, related to Figure 1. RNAseq data were retrieved from TCGA. mRNA levels of PI3K kinases in classical, mesenchymal, or proneural subtype in MGMT-deficient GBM (**A**) or MGMT-positive GBM (**B**) are shown. Sample sizes (N) are shown. Error bars are standard errors. Student *t*-test or one-way ANOVA was used to determine statistical significance. ns: not significant; *: $P < 0.05$; **: $P < 0.01$; ***: $P < 0.001$; ****: $P < 0.0001$.

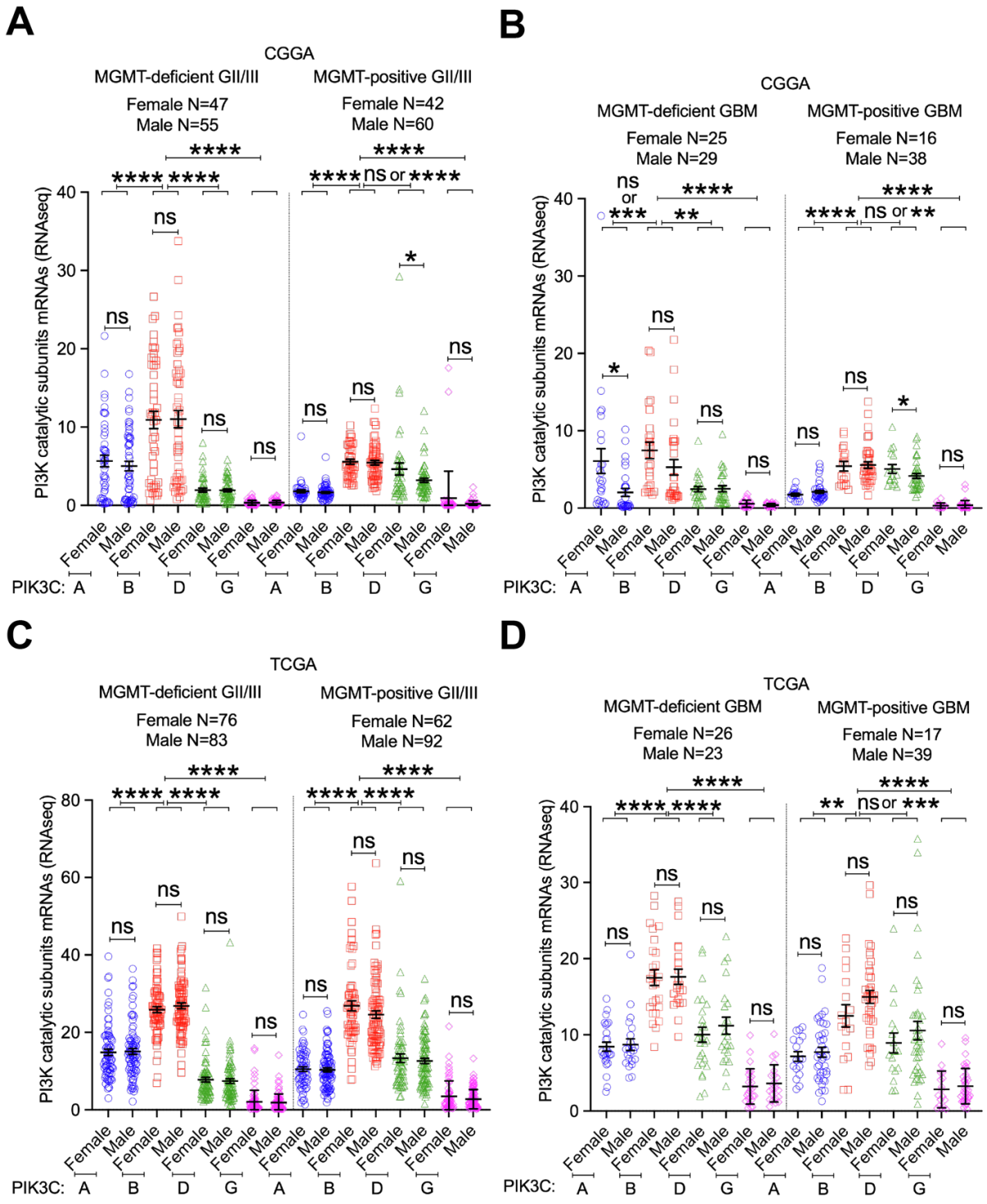

Figure S5. Gender difference in the expression of PI3K kinases, related to Figure 1. RNAseq data were retrieved from CGGA (**A** and **B**) or TCGA (**C** and **D**). mRNA levels of PI3K kinases in female or male grade II/III (GII/III) MGMT-deficient glioma (**A** and **C**) or MGMT-positive glioma (**B** and **D**). Sample sizes (N) are shown. Error bars are standard errors. Student *t*-test or one-way ANOVA was used to determine statistical significance. ns: not significant; *: $P < 0.05$; **: $P < 0.01$; ***: $P < 0.001$; ****: $P < 0.0001$.

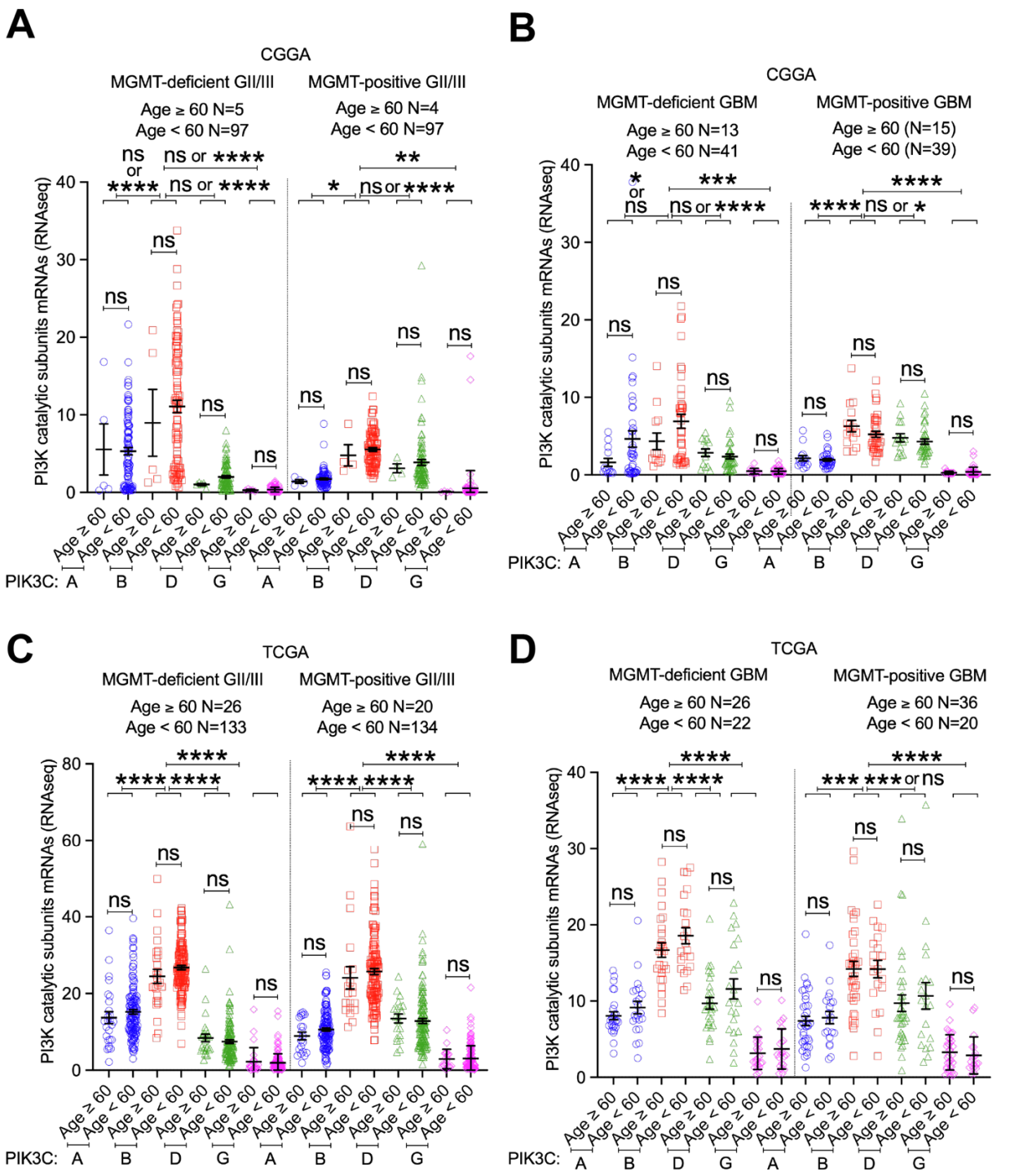

Figure S6. Expression of PI3K kinases in young or old glioma patients, related to Figure 1. RNAseq data were retrieved from CGGA (**A** and **B**) or TCGA (**C** and **D**). Patients were divided into two groups (Age >= 60 or Age < 60). mRNA levels of PI3K kinases in grade II/III (GII/III) glioma (**A** and **C**) or grade IV GBM (**B** and **D**) are shown. Their levels were also compared in MGMT-deficient or MGMT-positive gliomas. Sample sizes (N) are shown. Error bars are standard errors. Student *t*-test or one-way ANOVA was used to determine statistical significance. ns: not significant; *: $P < 0.05$; **: $P < 0.01$; ***: $P < 0.001$; ****: $P < 0.0001$.

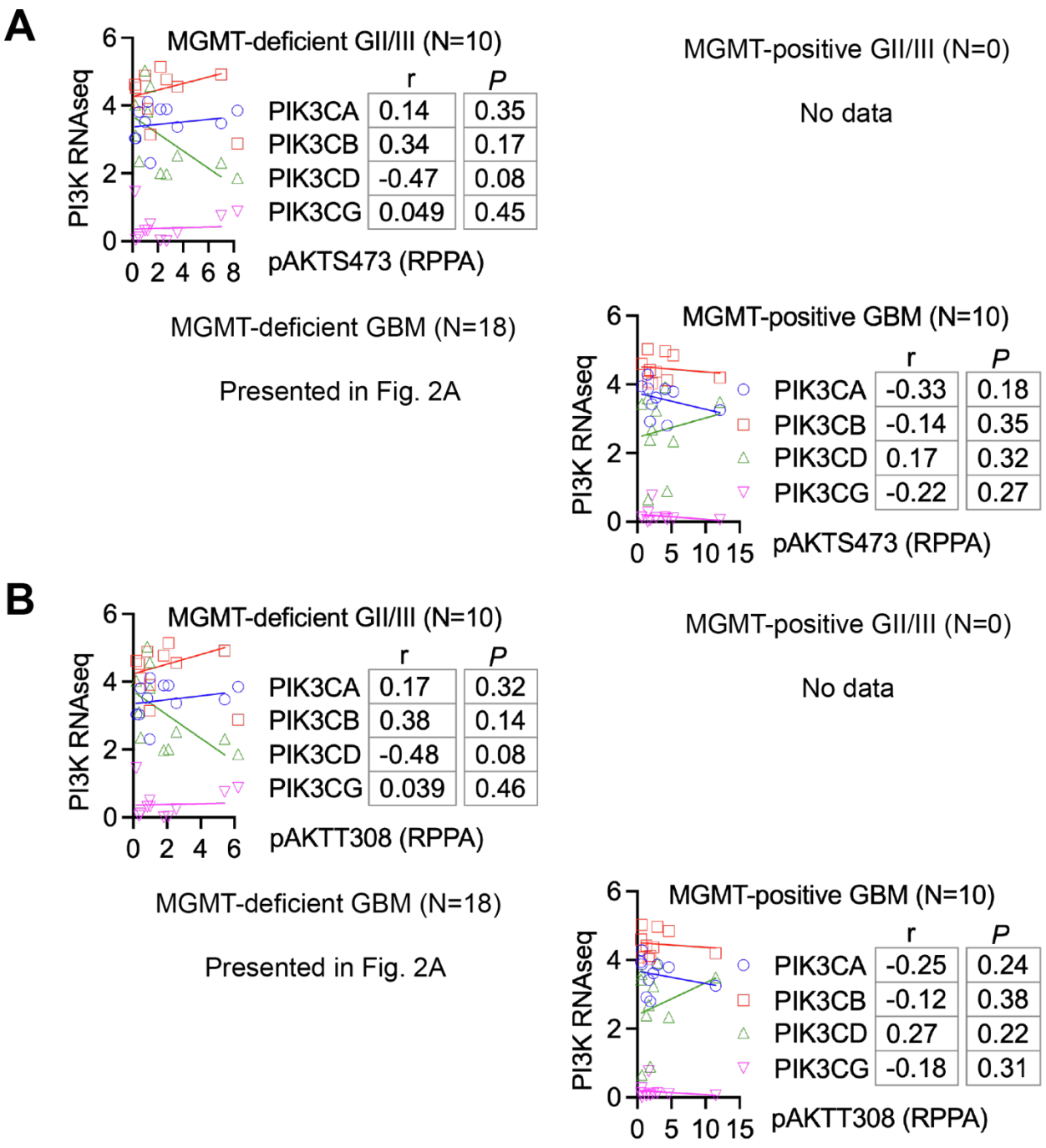

Figure S7. Correlation between PI3K mRNAs and pAKTS473/T308 in DepMap glioma cell lines, related to Figure 2. RNAseq or reverse phase protein array (RPPA) data were retrieved from DepMap. (**A**) Pearson correlation between PI3K mRNAs and pAKTS473 in DepMap MGMT-deficient or MGMT-positive grade II/III (GII/III) cell lines or GBM cell lines. (**B**) Pearson correlation between PI3K mRNAs and pAKTT308 in DepMap MGMT-deficient or MGMT-positive grade II/III (GII/III) cell lines or GBM cell lines. Prism 10 was used to determine Pearson coefficients. Protein levels of pAKTS473/T308 were normalized by those of total AKT and beta-actin. Pearson correlation coefficient r and *P* values as well as case numbers (N) are shown. Results in MGMT-deficient GBMs are shown in Figure 2A. No MGMT-positive GII/III cell lines were found in DepMap.

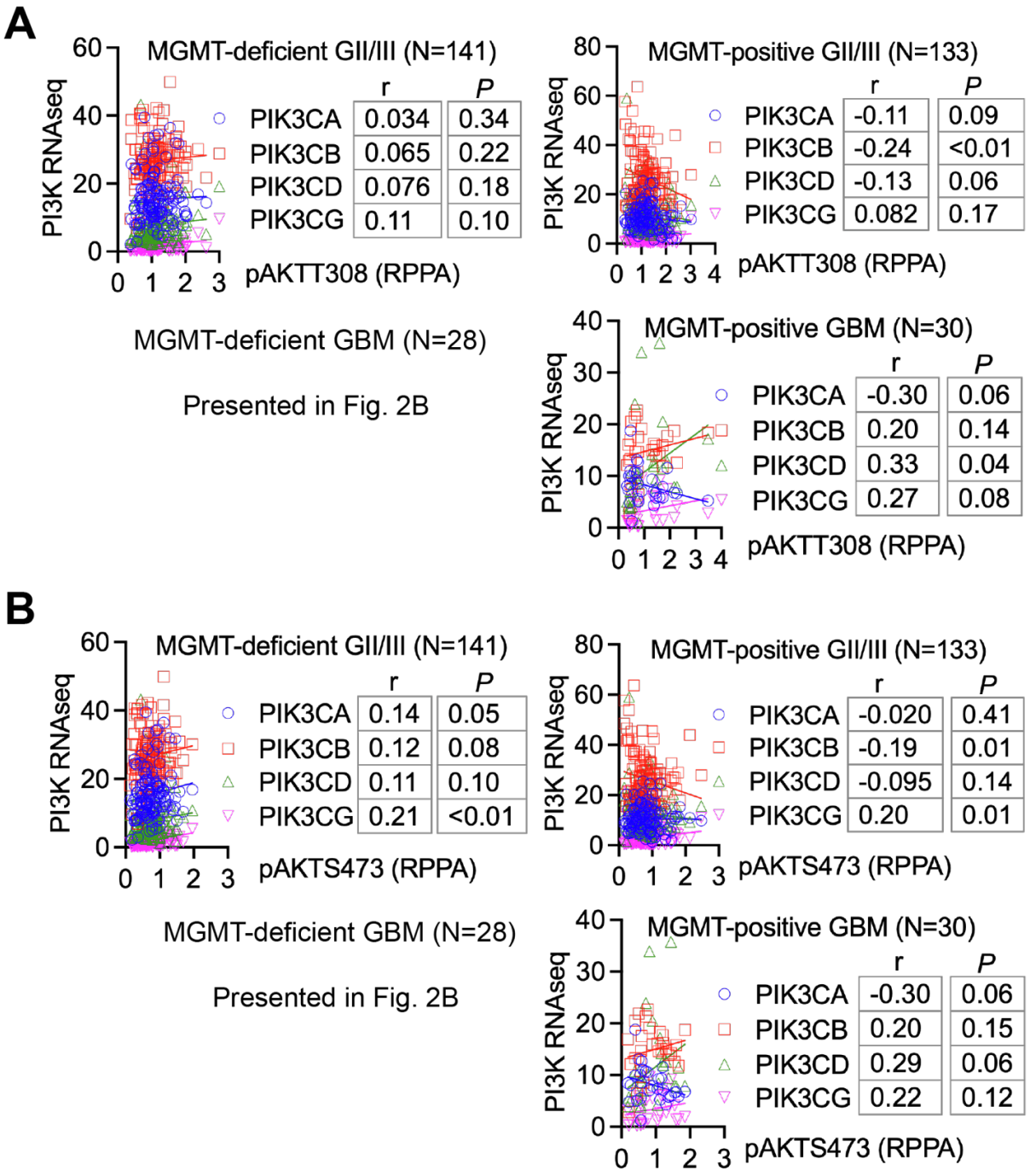

Figure S8. Correlation between PI3K mRNAs and pAKTS473/T308 in TCGA glioma specimens, related to Figure 2. RNAseq or reverse phase protein array (RPPA) data were retrieved from TCGA. (**A**) Pearson correlation between PI3K mRNAs and pAKTS473 in TCGA MGMT-deficient or MGMT-positive grade II/III (GII/III) specimens or GBM specimens. (**B**) Pearson correlation between PI3K mRNAs and pAKTT308 in TCGA MGMT-deficient or MGMT-positive grade II/III (GII/III) tumors or GBM tumors. Prism 10 was used to determine Pearson coefficients. Protein levels of pAKTS473/T308 were normalized by those of total AKT and beta-actin. Pearson correlation coefficient r and *P* values as well as case numbers (N) are shown. Results in MGMT-deficient GBMs are shown in Figure 2B.

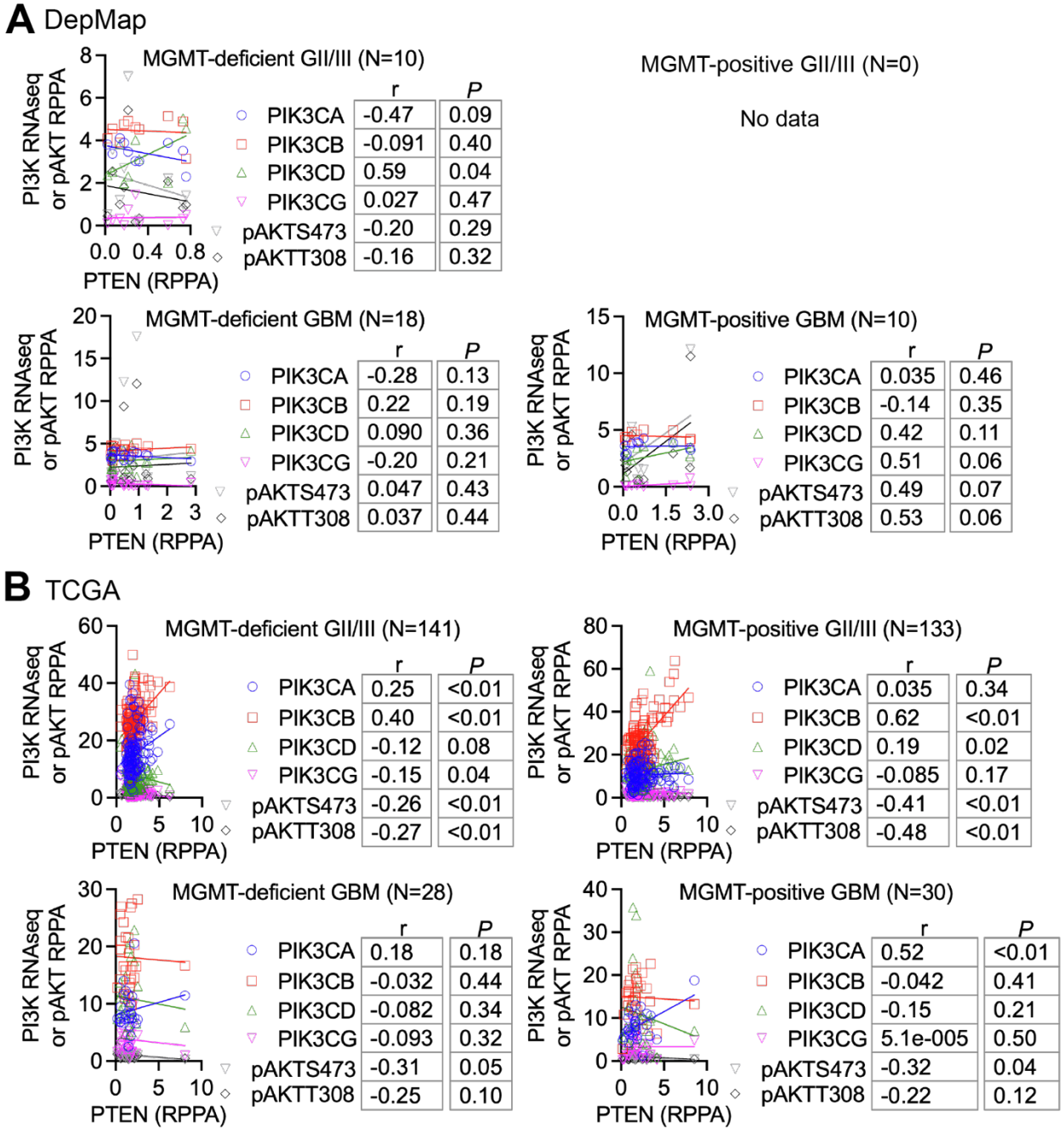

Figure S9. Correlation between PTEN proteins and PI3K mRNAs or AKTS473/T308, related to Figure 2. RNAseq or RPPA Data were retrieved from DepMap or TCGA. (**A**) Pearson correlation between PTEN proteins and PI3K mRNAs or pAKTS473/T308 in DepMap MGMT-deficient or MGMT-positive grade II/III (GII/III) cell lines or GBM cell lines. (**B**) Pearson correlation between PTEN proteins and PI3K mRNAs or pAKTS473/T308 in TCGA MGMT-deficient or MGMT-positive grade II/III (GII/III) tumors or GBM tumors. Prism 10 was used to determine Pearson coefficients. Protein levels of pAKTS473/T308 were normalized by those of total AKT and beta-actin. Pearson correlation coefficient r and *P* values as well as case numbers (N) are shown.

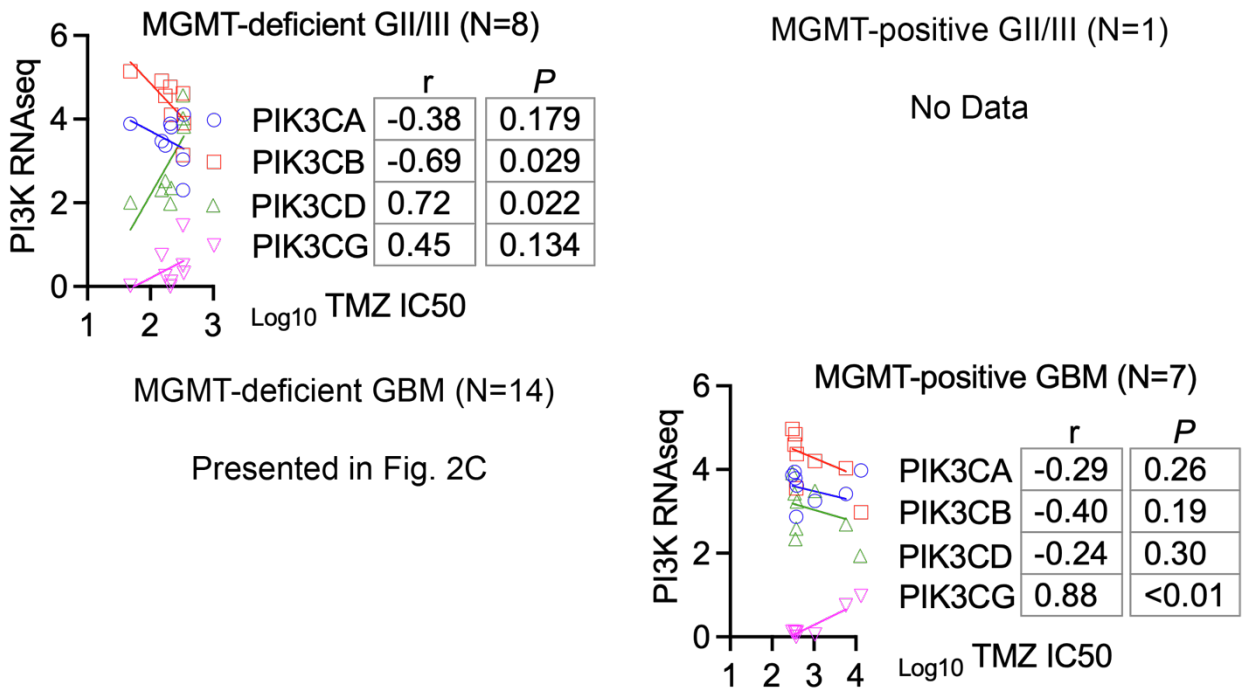

Figure S10. Correlation between TMZ IC50s and PI3K mRNAs, related to Figure 2. RNAseq or TMZ IC50 Data were retrieved from DepMap or acquired from the Sheng lab. Pearson correlations between TMZ IC50s and PI3K mRNAs in DepMap MGMT-deficient or MGMT-positive grade II/III (GII/III) cell lines or GBM cell lines were determined using Prism 10. Pearson correlation coefficient r and *P* values as well as case numbers (N) are shown. Results in MGMT-deficient GBMs are shown in Figure 2C.

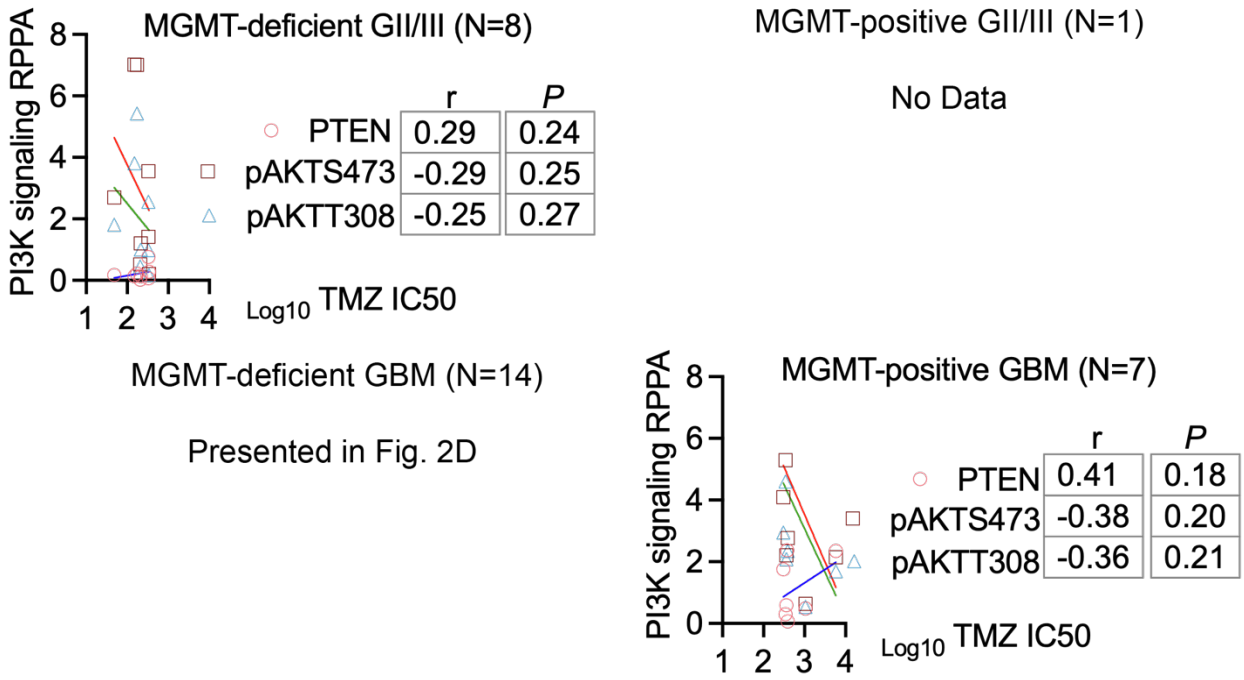

Figure S11. Correlation between TMZ IC50s and pAKTS473/T308 or PTEN, related to Figure 2. RPPA or TMZ IC50 Data were retrieved from DepMap or acquired from the Sheng lab. Pearson correlations between TMZ IC50s and pAKTS473/T308 or PTEN in DepMap MGMT-deficient or MGMT-positive grade II/III (GII/III) cell lines or GBM cell lines were determined using Prism 10. Pearson correlation coefficient r and *P* values as well as case numbers (N) are shown. Protein levels of pAKTS473/T308 were normalized by those of total AKT and beta-actin. Results in MGMT-deficient GBMs are shown in Figure 2D.

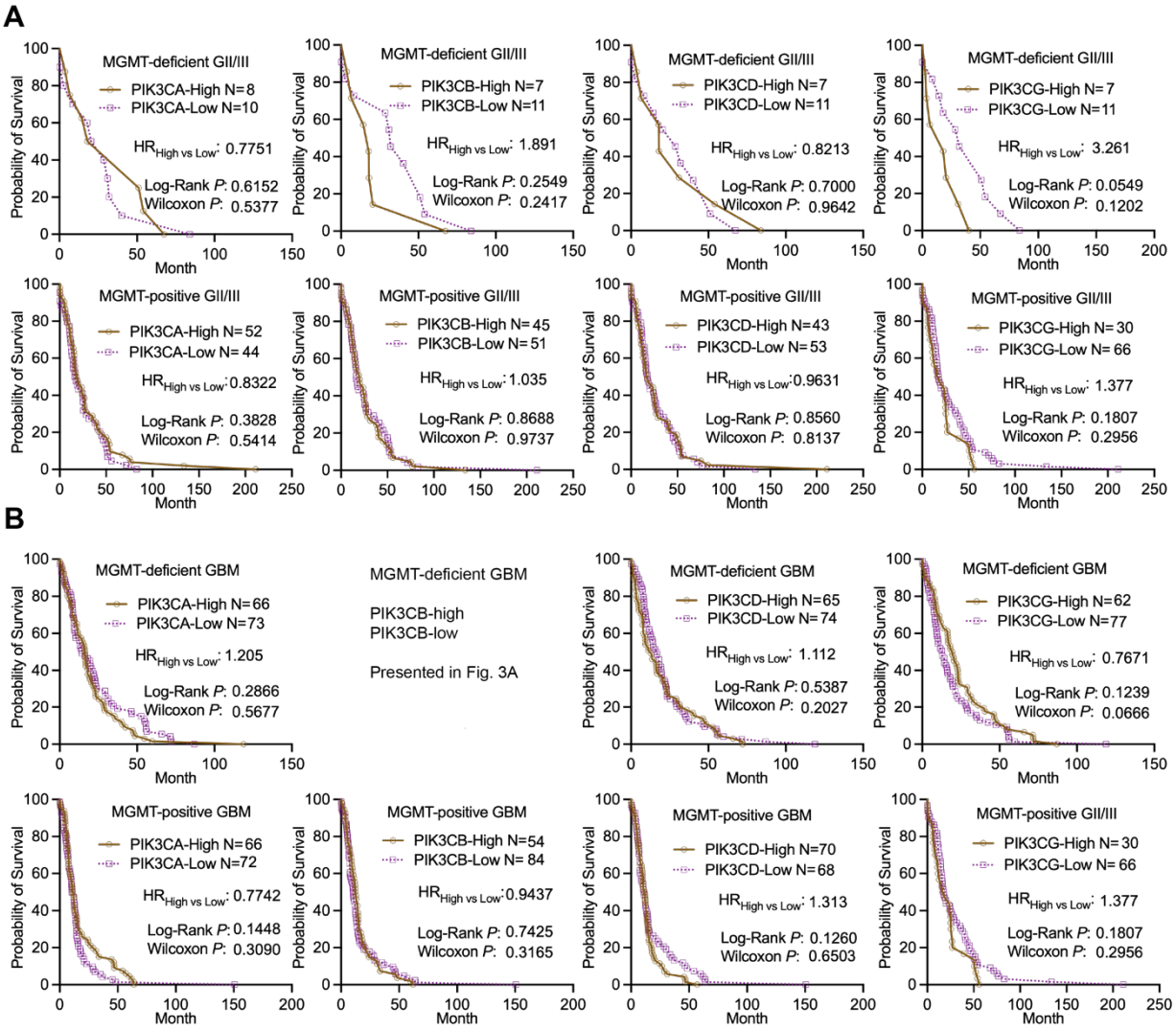

Figure S12. Survival analyses of PI3K kinases in IDH-wt gliomas, related to Figure 3. RNAseq and associated clinical data were retrieved from TCGA and CGGA. Additional datasets (e.g., Rembrandt, Murat, Ducay, LeeY Gravendeel, Ivy, Gorovets, Grzmil, Kamoun, Philips, POLA, and Vital) were downloaded from the GlioVis program (heep://gliovis.bioinfo.cnio.es). Primary gliomas with wild-type isocitrate dehydrogenase (IDH-wt) were included in the analysis, whereas recurrent gliomas or gliomas with mutant IDH were excluded. Kaplan-Meier analysis was performed in MGMT-deficient or MGMT-positive grade II/III (GII/III) glioma (**A**) and GBM (**B**) by comparing the survival of patients with high or low levels of PI3K kinases. Hazard ratios (HRs) of patients with high levels over those with low levels of PI3K genes (high vs low), case numbers (N), Log-Rank *P* values, and Wilcoxon *P* values are shown. Results in MGMT-deficient GBMs are shown in Figure 3A.

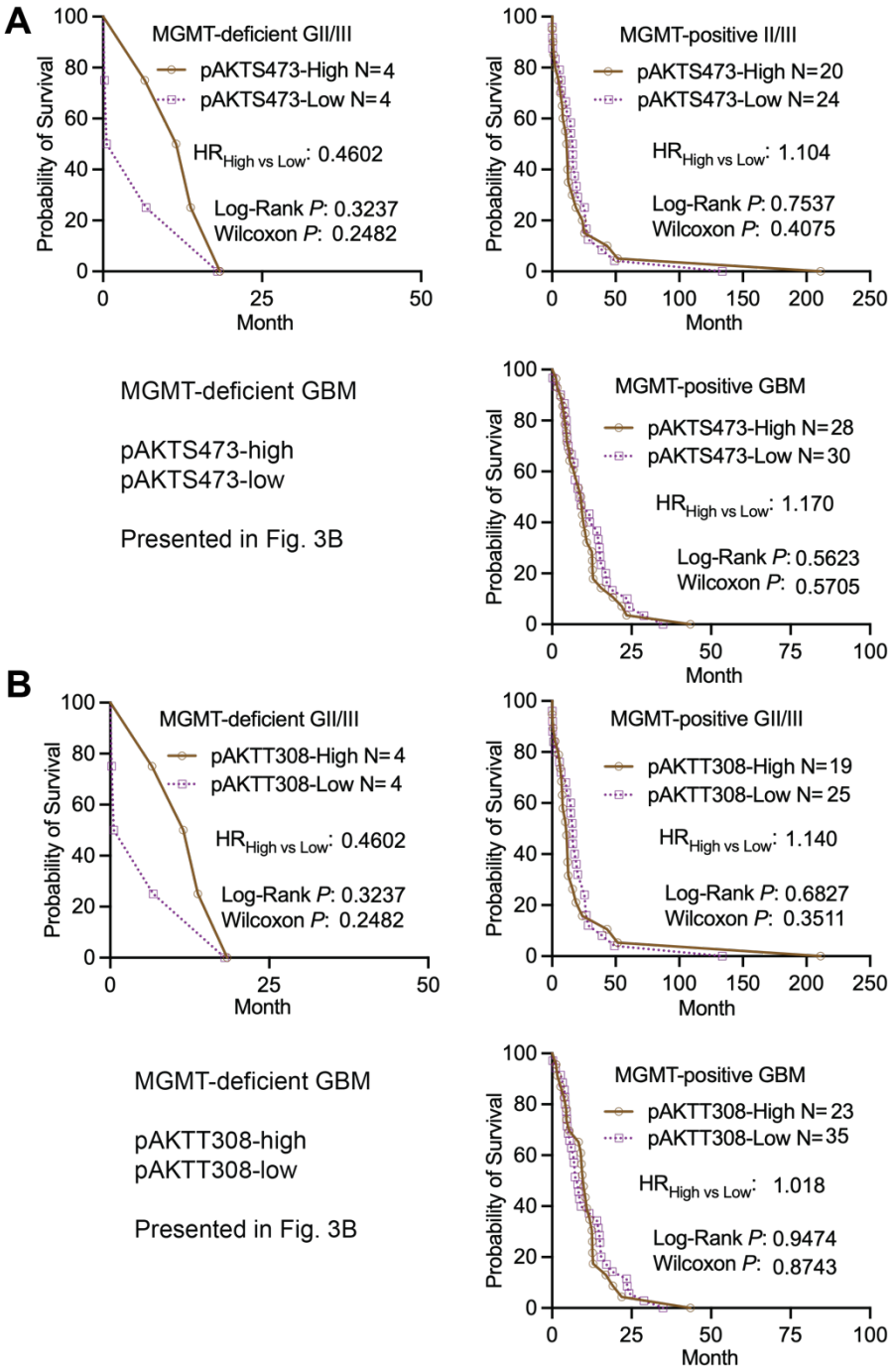

Figure S13. Survival analyses of pAKTS473 and pAKTT308 in IDH-wt gliomas, related to Figure 3. RPPA and associated clinical data were retrieved from TCGA and CGGA. Primary gliomas with IDH-wt were included in the analysis. Recurrent gliomas or gliomas with IDH-mut were excluded. Kaplan-Meier analysis was performed in MGMT-deficient or MGMT-positive grade II/III (GII/III) glioma and GBM by comparing the survival of patients with high or low levels of pAKTS473 (**A**) or pAKTT308 (**B**). Hazard ratios (HRs) of patients with high levels of pAKTS473/T308 over those with low levels of pAKTS473/T308 (high vs low), case numbers (N), Log-Rank *P* values, and Wilcoxon *P* values are shown. Results in MGMT-deficient GBMs are shown in Figure 3B.

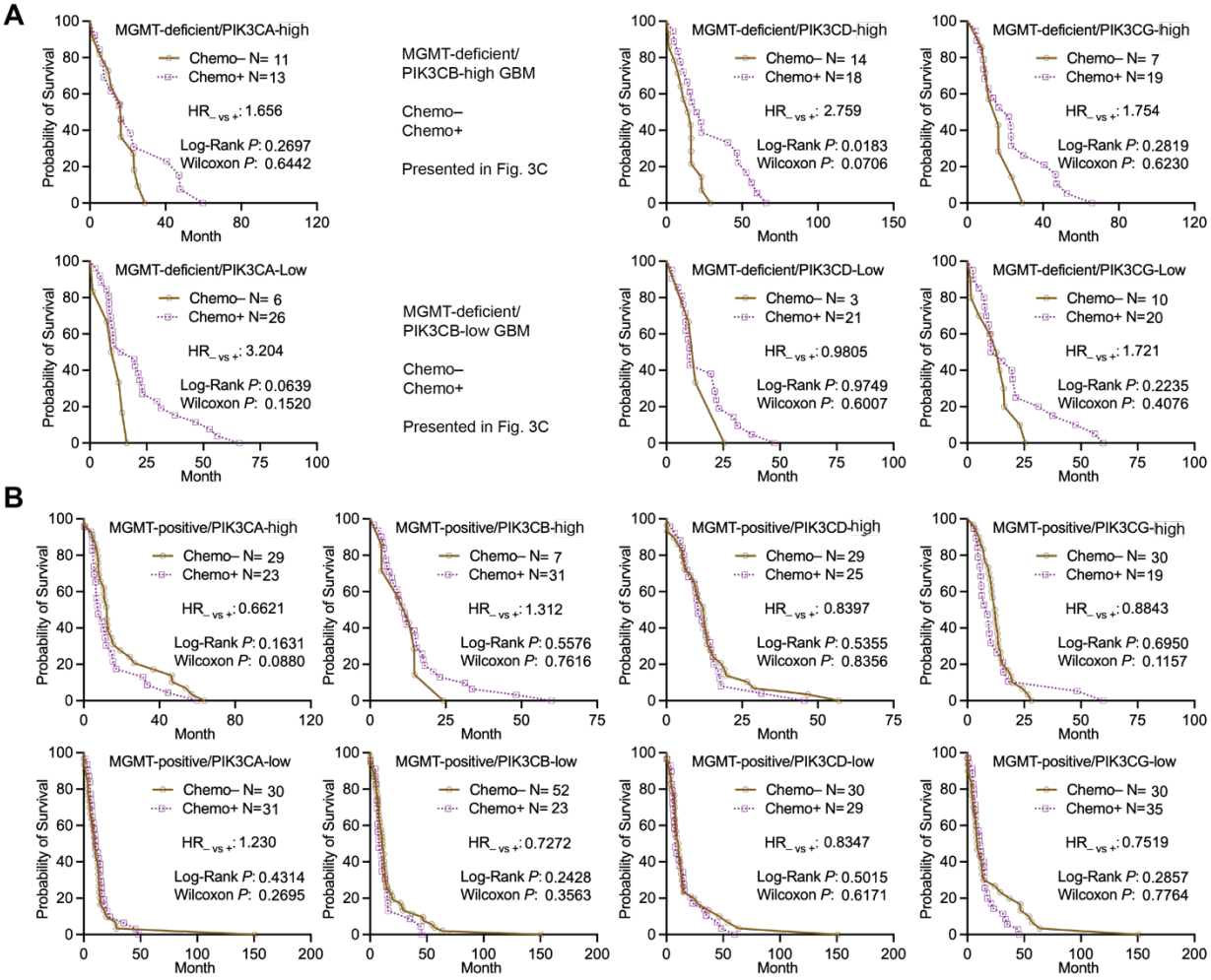

Figure S14. Survival analyses of gliomas treated with or without chemotherapy, related to Figure 3. RNAseq and associated clinical data were retrieved from TCGA and CGGA. IDH-wt primary GBMs treated with or without chemotherapy were included in the analysis. Kaplan-Meier analysis was performed in MGMT-deficient (**A**) or MGMT-positive (**B**) GBMs by comparing the survival of patients treated with or without chemo. Hazard ratios (HRs) of patients receiving no treatment (Chemo–) or treated with chemotherapy (Chemo+) (– vs +), case numbers (N), Log-Rank *P* values, and Wilcoxon *P* values are shown. Results in MGMT-deficient GBMs are shown in Figure 3C.

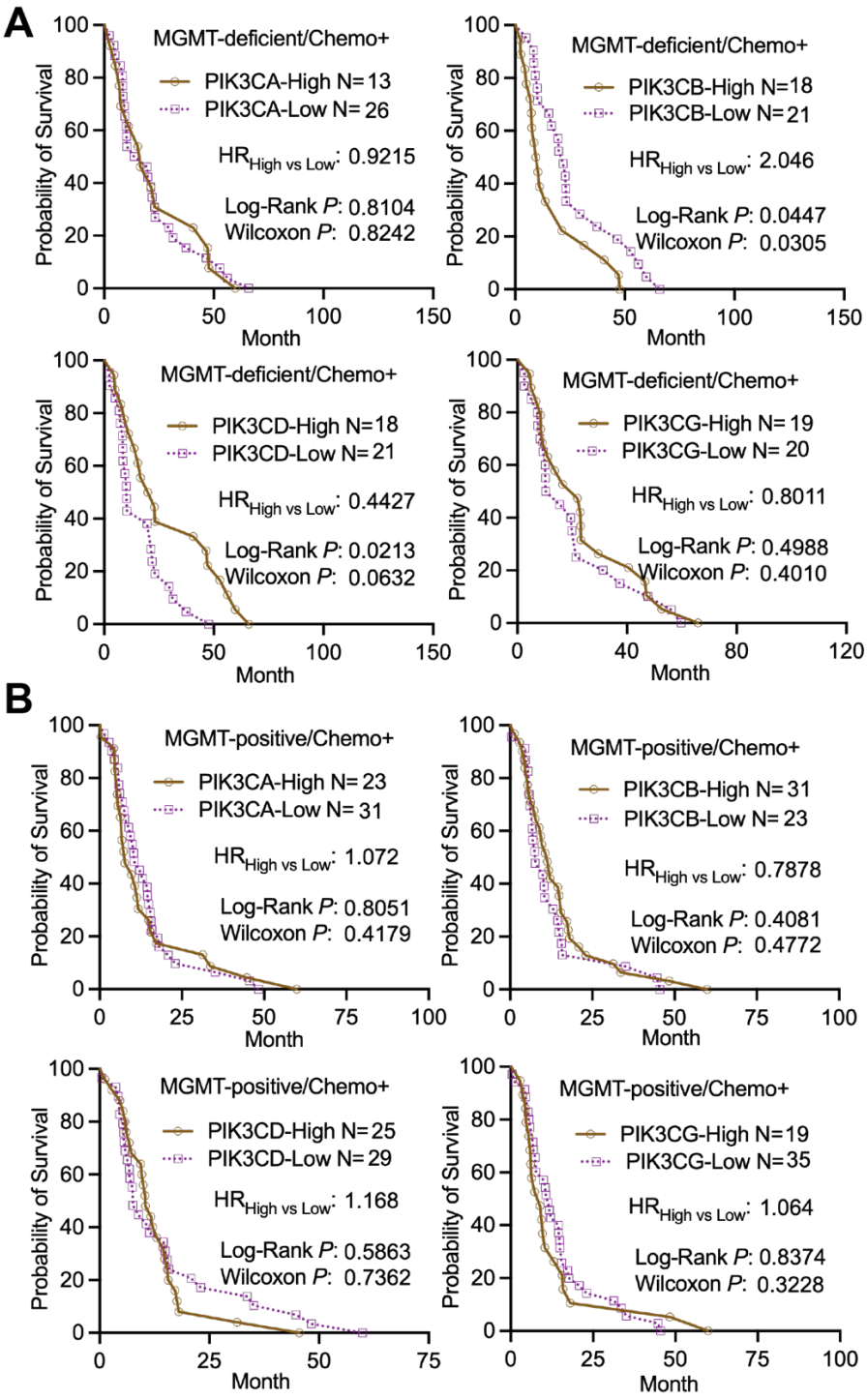

Figure S15. Survival analyses of chemotherapy-treated GBM patients expressing with high or low levels of PI3K kinases, related to Figure 3. RNAseq and associated clinical data were retrieved from TCGA and CGGA. IDH-wt primary GBMs treated with chemotherapy (Chemo+) were included in the analysis. Kaplan-Meier analysis was performed in MGMT-deficient (**A**) or MGMT-positive (**B**) GBMs by comparing the survival of patients with high or low levels of PI3K kinases. Hazard ratios (HRs) of patients with high levels to those with low levels of PI3K genes (high vs low), case numbers (N), Log-Rank *P* values, and Wilcoxon *P* values are shown.

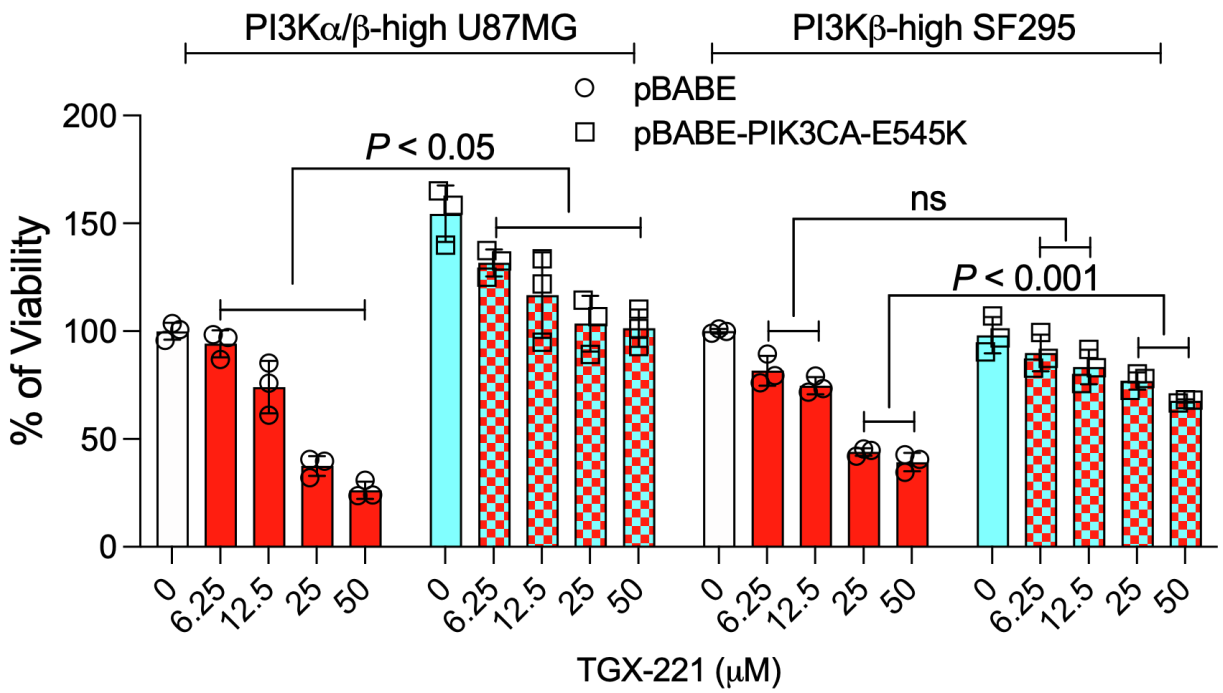

Figure S16. Overexpression of PIK3CA-E545K antagonizes growth inhibition caused by TGX-221, related to Figure 4. PI3Kα/β-high U87MG and PI3Kβ-high SF295 cells were transfected with the empty pBABE vector or a plasmid pBABE-PIK3CA-E545K that encodes a constitutively active PI3K mutant Pi3Kα-E545K. Cells were then treated with TGX-221 at different doses (0 to 50 μM). Cell viability was measured using the MTS viability assay. Student *t* test or One-way ANOVA was used to determine *P* values. ns: not significant.