

Title: Annexin A2 Mediates Bevacizumab Resistance and Infiltrative Escape in Glioblastoma

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Supplementary TableS1: Clinical Data

Naïve-Refractory (n=15)

Case	age	sex	recurrent pattern of MRI	TMZ cycles	Bev cycles	PFS (months)	OS (months)	EOR
1	65	M	T2 circumscribed	15	19	7.5	31.9	STR
2	48	F	cT1-flare up	18	22	7.5	30.4	STR
3	50	F	cT1-flare up	14	20	4.4	19.0	GTR
4	52	F	T2 diffuse	9	16	6.5	NA	GTR
5	73	M	T2 diffuse	13	26	9.6	18.5	GTR
6	52	M	T2 diffuse	20	41	9.8	23.3	PR
7	41	M	T2 diffuse	26	26	6.5	NA	GTR
8	51	M	T2 circumscribed	9	11	11.7	NA	biopsy
9	56	M	cT1-flare up	27	21	4.7	31.1	GTR
10	65	F	T2 diffuse	10	26	10.3	16.9	GTR
11	54	M	cT1-flare up	7	15	6.3	11.0	STR
12	51	M	cT1-flare up	8	11	6.5	14.6	STR
13	64	M	cT1-flare up	9	11	4.0	12.8	GTR
14	68	M	cT1-flare up	10	9	6.0	20.3	GTR
15	29	F	cT1-flare up	22	3	21.0	27.5	GTR

NeoBev-Refractory (n=18)

Case	age	sex	recurrent pattern of MRI	TMZ cycles	Bev cycles	PFS (months)	OS (months)	EOR	volume reduction rate on T1 Gd after NeoBev. (%)	volume reduction rate on FLAIR after NeoBev. (%)
1	77	F	T2 circumscribed	10	17	8.6	13.3	GTR	-14	-33
2	55	M	T2 diffuse	18	38	21.2	27.8	STR	-61	-27
3	68	M	cT1-flare up	10	24	10.3	16.4	GTR	-38	-26
4	50	M	cT1-flare up	9	12	10.0	14.5	GTR	-26	-54
5	78	M	cT1-flare up	3	8	9.8	17.4	GTR	-66	-71
6	76	M	T2 circumscribed	2	10	5.4	10.4	STR	-15	-60
7	63	M	cT1-flare up	10	13	6.9	16.1	GTR	-56	-63
8	70	M	cT1-flare up	6	7	7.1	12.5	GTR	-42	-64
9	68	F	cT1-flare up	15	16	17	19.3	PR	-42	-33
10	70	M	cT1-flare up	3	4	6.1	7.5	GTR	-35	-64
11	80	M	cT1-flare up	5	1	5.2	9.3	GTR	-18	-63
12	53	M	cT1-flare up	13	16	9.1	20.4	STR	30	-24
13	69	M	T2 circumscribed	9	10	9.5	38.3	GTR	-58	-38
14	51	F	cT1-flare up	3	3	6.3	9.8	GTR	-14	-75
15	74	F	cT1-flare up	7	3	11.0	45.2	STR	-37	-85
16	45	M	non-responder	18	21	18.1	25.7	GTR	-52	-14
17	65	F	cT1-flare up	3	8	11.4	21.6	STR	-33	-65
18	78	M	cT1-flare up	7	5	8.5	11.3	GTR	-59	-68

Bev, bevacizumab; EOR, extent of resection; F, female; GTR, gross total resection; M, male; N, Not Available ; OS, overall survival; PFS, progression free survival; PR, partial resection; STR, subtotal resection; TMZ, temozolomide

Supplementary TableS2: The primer sequences used for qPCR

Name	primer sequences
GAPDH-F	GTCCACTGGCGTCTTCACCA
GAPDH-R	GTGGCAGTGATGGCATGGAC
ACTβ-F	GATCAAGATCATTGCTCCTCCT
ACTβ-R	GGGTGTAACGCAACTAAGTCA
ANXA2-F	CCCAAGTGCCTACGGGTCAG
ANXA2-R	GCTGTCTCAATGTTTCAGAGCA

ACTβ: Actin beta, ANXA2: Annexin A2, F: forward,
GAPDH: Glyceraldehyde-3-phosphate dehydrogenase, R: reverse

Supplementary TableS3: classification of modified response assessment in neuro-oncology 2.0 (mRANO 2.0)

condition	Classification (RANO response)	Number of Patients	mPFS (months)	mOS (months)
≥50% reduction in T1-Gd and ≤30% reduction in FLAIR	PR	4	9.0	16.8
other cases	SD	13	10.0	14.5
≥25% increase in T1-Gd or ≥25% increase in FLAIR	PD	1	9.1	20.4

PR: Partial Response, PD: Progressive Disease, SD: Stable Disease
PFS: Progression-Free Survival, OS: Overall Survival

Supplementary TableS4: Multivariate Cox Proportional Hazards Analysis for Survival

		Progression-Free Survival (PFS)			Overall Survival (OS)		
Variable		Hazard Ratio	95% CI	P value	Hazard Ratio	95% CI	P value
ANXA2 expression	High vs. Low	2.85	1.12 – 7.24	0.028	3.12	1.25 – 7.80	0.015
Treatment group	NeoBev vs. Naïve	0.33	0.14 – 0.77	0.010	0.95	0.42 – 2.16	0.912
Age	≥65 vs. <65	1.42	0.65 – 3.10	0.385	1.58	0.72 – 3.45	0.254
Sex	Male vs. Female	1.15	0.52 – 2.54	0.728	3.06	1.15 – 8.13	0.025
EOR	Non-GTR vs. GTR	1.28	0.58 – 2.82	0.540	1.45	0.62 – 3.38	0.392
TMZ cycles	Continuous	0.96	0.89 – 1.03	0.251	0.89	0.80 – 0.99	0.031
Bev cycles	Continuous	1.02	0.98 – 1.06	0.342	1.01	0.97 – 1.05	0.655

ANXA2, annexin A2; Bev, bevacizumab; CI, confidence interval; EOR, extent of resection; GTR, gross total resection; TMZ, temozolomide

Supplementary TableS5: Comparison of ANXA2 Expression Levels and Survival Data

condition	N	mPFS (months)	N	mOS (months)
naïve low expression	7	7.5	6	21.8
naïve high expression	8	6.5	6	18.0
neoBev low expression	10	9.9	10	19.9
neoBev high expression	8	6.7	8	10.9

mPFS: median Progression-free Survival

mPFS: median Overall Survival

Supplementary TableS6: Multivariate Cox Proportional Hazards Analysis for Survival

condition of treatment and ANXA2 expression level	Progression-Free Survival (PFS)			Overall Survival (OS)		
	Hazard Ratio	95% CI	<i>P</i> value	Hazard Ratio	95% CI	<i>P</i> value
neoBev/low vs. neoBev/high	0.43	0.16 - 1.13	0.066	0.37	0.13 - 1.04	0.051
neoBev/low vs. naïve/low	0.78	0.28 - 2.15	0.635	1.21	0.42 - 3.53	0.733
neoBev/low vs. naïve/high	0.58	0.22 - 1.51	0.264	0.74	0.26 - 2.09	0.565
neoBev/high vs. naïve/low	1.81	0.65 - 5.06	0.258	3.24	1.12 - 9.38	0.030
neoBev/high vs. naïve/high	1.35	0.53 - 3.44	0.534	1.99	0.76 - 1.78	0.366

CI; confidence interval
PFS: Progression-free Survival
OS: Overall Survival

Supplementary TableS7: Cox proportional hazards analysis across treatment and ANXA2 expression subgroups

Subgroup	Progression-Free Survival (PFS)			Overall Survival (OS)		
	Hazard Ratio	95% CI	<i>P</i> value	Hazard Ratio	95% CI	<i>P</i> value
neoBev / low	1.00	Reference	-	1.00	Reference	-
neoBev / high	2.34	0.88 - 6.18	0.086	2.70	0.95 - 7.66	0.061
naïve / low	1.23	0.46 - 3.57	0.627	0.83	0.28 - 2.41	0.725
naïve / high	1.73	0.66 - 4.51	0.264	1.35	0.48 - 3.80	0.565
Overall p-value			0.287			0.088

CI: confidence interval
PFS: Progression-free Survival
OS: Overall Survival