

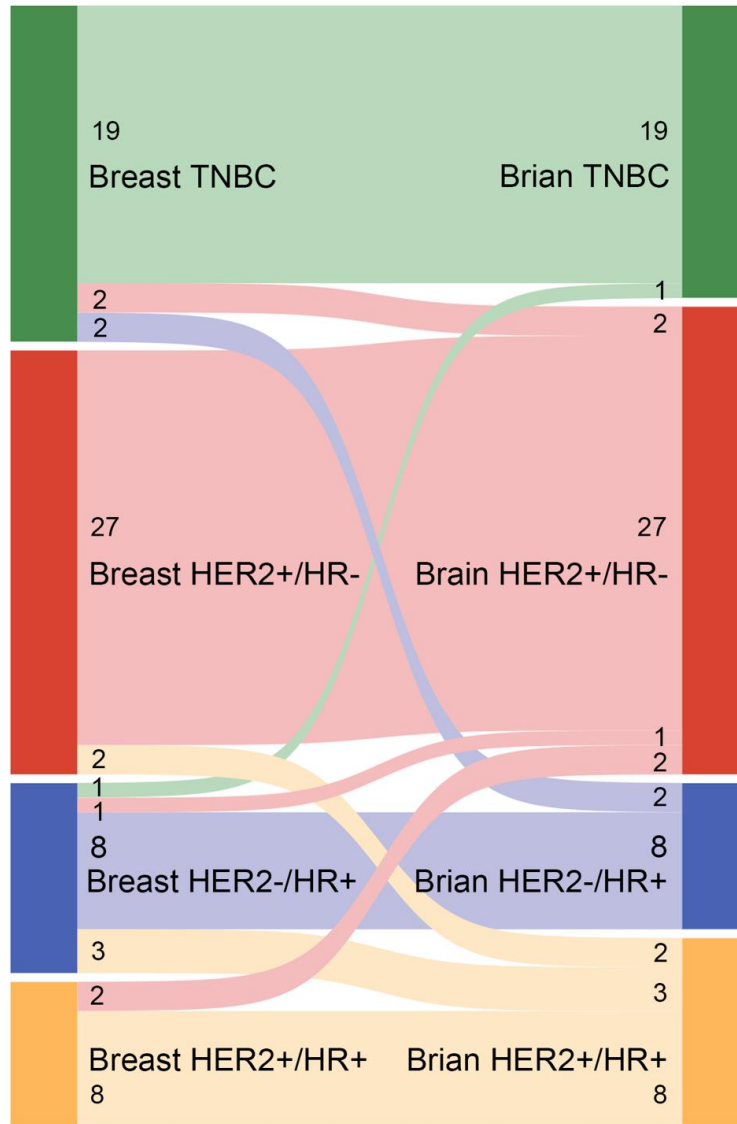
Supplementary Table S1

Antibodies used in panel 1 of Figure 5 and panel 2 of Supplementary Figure S3

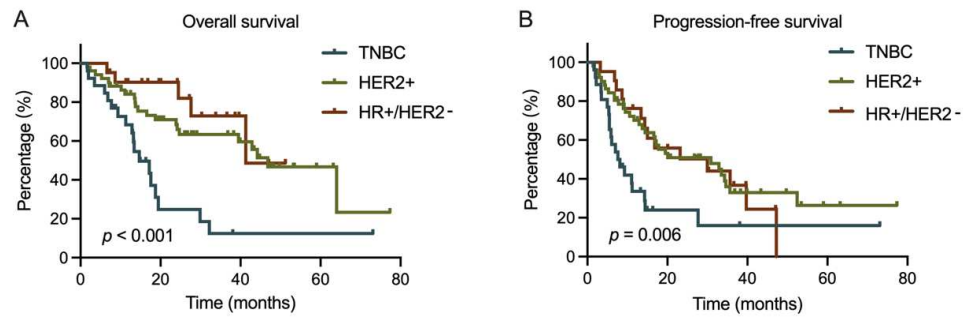
Panel	Cycle	Marker	Conjugate	Vendor	Catalog	Dilution
1	1	Autofluorescence	AF488	None	None	None
1	1	Autofluorescence	AF555	None	None	None
1	1	TIM-3	None	Abcam	ab241332	1/100
1	1	Goat anti-rabbit IgG	AF647	Abcam	ab150083	1/200
1	2	CD163	AF488	Abcam	ab218293	1/100
1	2	CD68	AF555	CST	79594	1/100
1	2	CD20	AF647	eBioscience	50-0202-80	1/100
1	3	CD3D	AF555	Abcam	ab208514	1/100
1	3	CD8a	AF647	eBioscience	50-0008-82	1/100
1	4	PD-L1	AF555	Abcam	ab213358	1/50
1	4	PD-1	AF647	Abcam	ab201825	1/100
1	5	pan-CK	AF555	CST	3478	1/200
1	5	Foxp3	AF647	BioLegend	320113	1/50
2	1	Cleaved Caspase-3	None	CST	9661	1/100
2	1	BCL-6	None	GeneAb	IHC029-100	1/100
2	1	Goat anti-mouse IgG	AF555	Abcam	ab150118	1/200
2	1	Goat anti-rabbit IgG	AF647	Abcam	ab150083	1/200
2	2	CD4	AF488	Abcam	ab196372	1/100
2	2	TCF1/TCF7	AF555	CST	17404	1/100
2	2	CXCR5	AF647	Abcam	ab316179	1/100
2	3	HLA-DR	AF488	BioLegend	307619	1/100
2	3	CD8a	AF647	eBioscience	50-0008-82	1/100
2	4	Ki-67	AF488	CST	11882	1/100
2	4	pan-CK	AF555	CST	3478	1/200
2	4	HLA-A	AF647	Abcam	ab199837	1/200
2	5	CD20	AF647	eBioscience	50-0202-80	1/100
2	5	CD57	AF555	BioLegend	393307	1/100

The code used for the analyses of the nomogram in Figure 4 is shared in the link below.

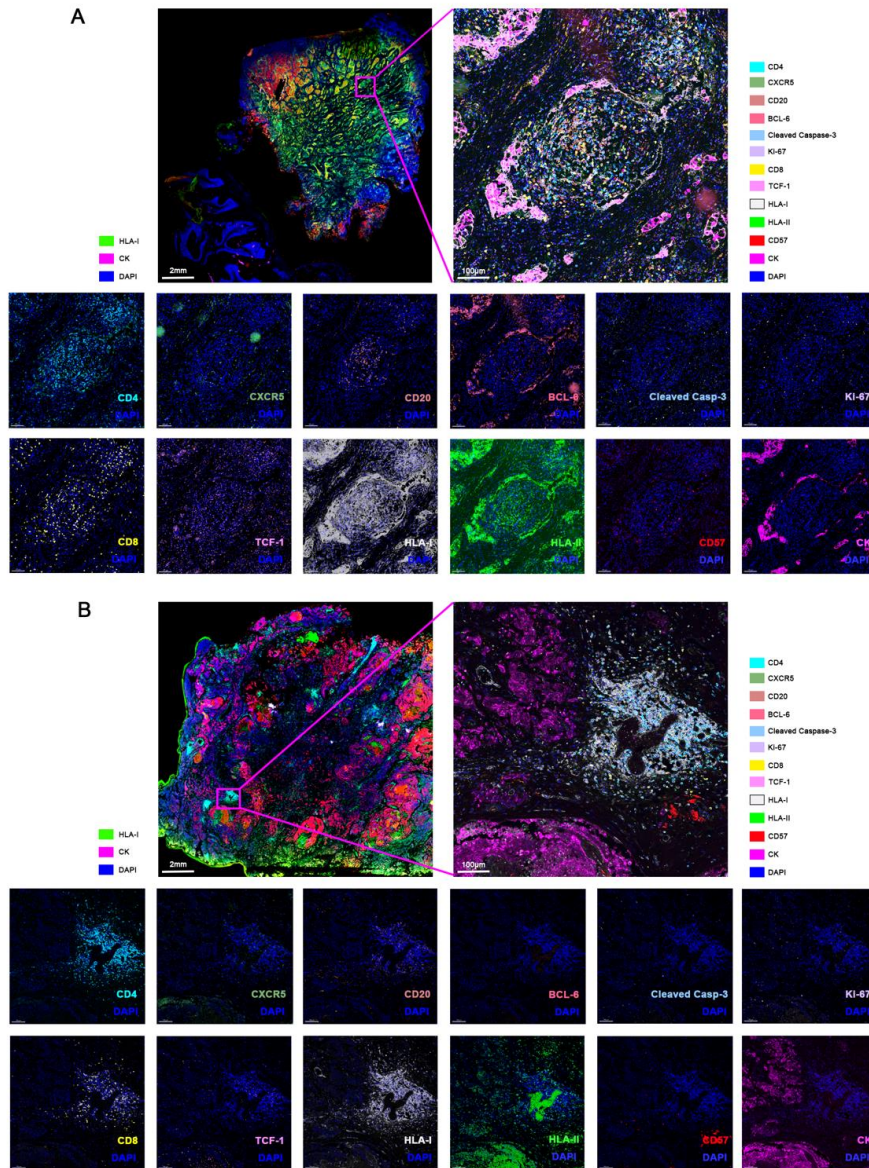
<https://gitee.com/tao-taos-warehouse/nomo.-r-score-sur.-r.git>



Supplementary Figure S1: Subtype switching. Width of the bands indicates number of patients crossing over. + = positive; - = negative; HR = hormonal receptor; HER2 = human epidermal growth factor receptor 2.



Supplementary Figure S2: Kaplan-Meier curves for overall survival (A) and progression-free survival (B) according to breast subtype. + = positive; - = negative; HR = hormonal receptor; HER2 = human epidermal growth factor receptor 2.



Supplementary Figure S3: Representative multiplex immunofluorescence images of case 1 (A) and case 2 (B) showing the positivity of CD4, CXCR5, CD20, BCL-6, Cleaved Caspase-3, Ki-67, CD8, TCF-1, HLA-I, HLA-II, CD57, and CK in intra-tumoral TLSs. Abbreviations: CD, cluster of differentiation; CXCR5, C-X-C motif chemokine receptor 5; BCL-6, BCL6 transcription repressor; HLA, histocompatibility antigen; TCF-1, T cell factor 1; CK, cytokeratin; DAPI, 4',6-diamidino-2-phenylindole.

Supplementary Table S2 The univariate analysis and multivariate analysis between clinicopathological characteristics and OS

Variables		Univariate analysis		Multivariate analysis	
		HR ^a (95% CI)	<i>p</i> value	HR (95% CI)	<i>p</i> value
age at time of BCBM (years)	<65	0.443 (0.183-1.072)	0.071	0.335 (0.121-0.930)	0.036
	≥65	Reference	-	Reference	-
Breast cancer subtype of BrM	TNBC	4.848 (1.781-13.193)	0.002	4.440 (1.505-13.100)	0.007
	HER2+	1.648 (0.621-4.374)	0.315	2.488 (0.872-7.101)	0.088
	HR ^b +/HER2-	Reference	-	Reference	-
TLS score	0	4.414 (1.510-12.902)	0.007	3.249 (1.050-10.055)	0.041
	1	2.195 (0.636-7.572)	0.213	1.072 (0.288-3.997)	0.917
	2	1.270 (0.357-4.522)	0.712	0.529 (0.136-2.062)	0.359
	3	Reference	-	Reference	-
KPS	90	0.342 (0.127-0.926)	0.035	0.226 (0.071-0.722)	0.012
	70-80	1.133 (0.495-2.595)	0.768	0.980 (0.380-2.529)	0.967
	60 or less	Reference	-	Reference	-
De novo Stage IV breast cancer	yes	0.301 (0.156-0.578)	<0.001	0.283 (0.129-0.620)	0.002
	no	Reference	-	Reference	-
Systemic therapy after BrM diagnosis	yes	1.994 (0.911-4.364)	0.084	2.366 (0.960-5.831)	0.061
	no	Reference	-	Reference	-
Radiotherapy after BrM diagnosis	yes	1.224 (0.597-2.512)	0.581	1.185 (0.490-2.867)	0.707
	no	Reference	-	Reference	-

OS = overall survival; HR^a = hazard ratio; CI = confidence interval; HR^b = hormone receptor; HER2 = epidermal growth factor receptor 2; TNBC: triple-negative breast cancer; KPS = Karnofsky performance status; TLS = tertiary lymphoid structure; BCBM = breast cancer brain metastasis; BrM = brain metastasis.

Supplementary Table S3 The univariate analysis and multivariate analysis between clinicopathological characteristics and PFS

Variables		Univariate analysis		Multivariate analysis	
		HR ^a (95% CI)	<i>p</i> value	HR (95% CI)	<i>p</i> value
age at time of BCBM (years)	<65	0.576 (0.271-1.225)	0.152	0.580 (0.250-1.345)	0.204
	≥65	Reference	-	Reference	-
Breast cancer subtype of BrM	TNBC	2.245 (1.121-4.495)	0.022	2.341 (1.079-5.080)	0.031
	HER2+	0.938 (0.498-1.766)	0.842	1.561 (0.766-3.182)	0.220
	HR ^b + / HER2-	Reference	-	Reference	-
TLS score	0	5.190 (2.010-13.397)	0.001	4.268 (1.595-11.417)	0.004
	1	3.594 (1.276-10.121)	0.015	2.517 (0.859-7.379)	0.093
	2	1.893 (0.631-5.678)	0.255	1.003 (0.317-3.177)	0.996
	3	Reference	-	Reference	-
KPS	90	0.324 (0.157-0.669)	0.002	0.233 (0.101-0.535)	0.001
	70-80	0.678 (0.363-1.265)	0.222	0.622 (0.321-1.204)	0.159
	60 or less	Reference	-	Reference	-
De novo Stage IV breast cancer	yes	0.312 (0.172-0.564)	<0.001	0.311 (0.156-0.619)	0.001
	no	Reference	-	Reference	-
Systemic therapy after BrM diagnosis	yes	1.625 (0.816-3.236)	0.167	1.981 (0.911-4.310)	0.085
	no	Reference	-	Reference	-
Radiotherapy after BrM diagnosis	yes	0.787 (0.416-1.486)	0.460	0.594 (0.277-1.274)	0.181
	no	Reference	-	Reference	-

PFS = progression-free survival; HR^a = hazard ratio; CI = confidence interval; HR^b = hormone receptor; HER2 = epidermal growth factor receptor 2; TNBC: triple-negative breast cancer; KPS = Karnofsky performance status; TLS = tertiary lymphoid structure; BCBM = breast cancer brain metastasis; BrM = brain metastasis.