

Developing an immune signature for triple-negative breast cancer to predict prognosis and immune checkpoint inhibitor

Supplementary Material

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Table S1 R packages in the study

Purpose	R packages
Data preprocessing for GSE58812	"affy" and "simpleaffy" packages
Calculate enrichment score for each sample using ssGSEA	"GSVA" package
KEGG and Go enrichment analyses	"clusterProfiler" package
Mutation profiles and calculate tumor mutation burden	"maftools" package
Estimate of immune score and stromal score using ESTIMATE algorithm	"Estimate" package

Table S2 66 prognostic IRGs of IPRS

Num	Genlist	Name	Category
1	AP3B1	adaptor related protein complex 3 subunit beta 1	Antigen_Processing_and_Presentation
2	APOD	apolipoprotein D	Antimicrobials
3	C5AR1	complement C5a receptor 1	Chemokine_Receptors,Cytokine_Receptors
4	CCL13	C-C motif chemokine ligand 13	Antimicrobials,Chemokines,Cytokines
5	CCL17	C-C motif chemokine ligand 17	Antimicrobials,Chemokines,Cytokines
6	CCL25	C-C motif chemokine ligand 25	Antimicrobials,Chemokines,Cytokines
7	CCL28	C-C motif chemokine ligand 28	Antimicrobials,Chemokines,Chemokines,Cytokines
8	CD19	CD19 molecule	BCRSignalingPathway
9	CD1B	CD1b molecule	Antigen_Processing_and_Presentation
10	CD79A	CD79a molecule	BCRSignalingPathway
11	CHP1	calcineurin like EF-hand protein 1	BCRSignalingPathway,NaturalKiller_Cell_Cytotoxicity,TCRsignalingPathway
12	CMTM4	CKLF like MARVEL transmembrane domain containing 4	Cytokines
13	CTSL	cathepsin L	Antimicrobials
14	CXCL13	C-X-C motif chemokine ligand 13	Antimicrobials,Chemokines,Cytokines
15	DCD	dermcidin	Antimicrobials
16	DKK1	dickkopf WNT signaling pathway inhibitor 1	Cytokines
17	FAM3B	FAM3 metabolism regulating signaling molecule B	Cytokines
18	FAM3C	FAM3 metabolism regulating signaling molecule C	Cytokines
19	FGF12	fibroblast growth factor 12	Cytokines
20	FGF6	fibroblast growth factor 6	Cytokines
21	FGF8	fibroblast growth factor 8	Cytokines
22	GAL	galanin and GMAP prepropeptide	Cytokines
23	GREM2	gremlin 2, DAN family BMP antagonist	Cytokines
24	HAMP	hepcidin antimicrobial peptide	Antimicrobials,Cytokines
25	HMOX1	heme oxygenase 1	Antimicrobials
26	HSPA2	heat shock protein family A (Hsp70) member 2	Antigen_Processing_and_Presentation

27	HSPA6	heat shock protein family A (Hsp70) member 6	Antigen_Processing_and_Presentation
28	IFNW1	interferon omega 1	Cytokines,Interferons
29	IGF2R	insulin like growth factor 2 receptor	Cytokine_Receptors
30	IKBKB	inhibitor of nuclear factor kappa B kinase subunit beta	BCRSignalingPathway,TCRsignalingPathway
31	IL11RA	interleukin 11 receptor subunit alpha	Cytokine_Receptors,Interleukins_Receptor
32	IL26	interleukin 26	Cytokines,Interleukins
33	INHBA	inhibin subunit beta A	Cytokines,TGFb_Family_Member
34	ITGAV	integrin subunit alpha V	Antimicrobials
35	KRAS	KRAS proto-oncogene, GTPase	BCRSignalingPathway,NaturalKiller_Cell_Cytotoxicity,TCRsignalingPathway
36	LPA	lipoprotein(a)	Antimicrobials
37	MASP1	mannan binding lectin serine peptidase 1	Antimicrobials
38	MC3R	melanocortin 3 receptor	Cytokine_Receptors
39	MSR1	macrophage scavenger receptor 1	Antimicrobials
40	MUC4	mucin 4, cell surface associated	Antimicrobials
41	NR2E1	nuclear receptor subfamily 2 group E member 1	Cytokine_Receptors
42	NR3C2	nuclear receptor subfamily 3 group C member 2	Cytokine_Receptors
43	NRTN	neurturin	Cytokines
44	PAK2	p21 (RAC1) activated kinase 2	TCRsignalingPathway
45	PGRMC2	progesterone receptor membrane component 2	Cytokine_Receptors
46	PIK3CA	phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha	BCRSignalingPathway,NaturalKiller_Cell_Cytotoxicity,TCRsignalingPathway
47	PLAU	plasminogen activator, urokinase	Antimicrobials,Chemokines,Cytokines
48	PLXNA1	plexin A1	Chemokine_Receptors,Cytokine_Receptors
49	PRLHR	prolactin releasing hormone receptor	Cytokine_Receptors
50	PSME3	proteasome activator subunit 3	Antigen_Processing_and_Presentation
51	PTGDS	prostaglandin D2 synthase	Antimicrobials,Cytokine_Receptors
52	PTGFR	prostaglandin F receptor	Cytokine_Receptors

53	RASGR p1	RAS guanyl releasing protein 1	TCRsignalingPathway
54	RFXAP	regulatory factor X associated protein	Antigen_Processing_and_Presentation
55	RLN1	relaxin 1	Cytokines
56	SDC3	syndecan 3	Cytokine_Receptors
57	SEMA4 A	semaphorin 4A	Chemokines,Cytokines
58	SPP1	secreted phosphoprotein 1	Cytokines
59	TANK	TRAF family member associated NFKB activator	Antimicrobials
60	TGFA	transforming growth factor alpha	Cytokines
61	TGFBR1	transforming growth factor beta receptor 1	Cytokine_Receptors,TGFb_Family_Member_Receptor
62	THBS1	thrombospondin 1	Antigen_Processing_and_Presentation
63	TNFRSF 13C	TNF receptor superfamily member 13C	Cytokine_Receptors,TNF_Family_Members_Receptors
64	TNFRSF 8	TNF receptor superfamily member 8	Cytokine_Receptors,TNF_Family_Members_Receptors
65	TSHR	thyroid stimulating hormone receptor	Cytokine_Receptors
66	TSLP	thymic stromal lymphopoietin	Cytokines

Table S3 Multivariate Cox regression for IPRS

Variable	TCGA set			
	Multivariate (OS)		Multivariate (RFS)	
	HR (95% CI)	P	HR (95% CI)	P
IPRS	2.837(1.872-4.299)	<0.001	2.044(1.396-2.991)	<0.001
Age	0.983(0.949-1.018)	0.458	0.988(0.945-1.034)	0.614
Stage				
I/II	reference			
III/IV	11.509(3.170-41.785)	<0.001	15.742(4.427-55.980)	<0.001

(Continue Table S4)

Variable	GSE58812 set			
	Multivariate (OS)		Multivariate (RFS)	
	HR (95% CI)	P	HR (95% CI)	P
score	1.605(1.158-2.225)	0.005	1.746(1.238-2.377)	<0.001
Age	1.069(1.029-1.110)	<0.001	1.046(1.010-1.083)	0.011
Stage	-		-	
I/II	-		-	
III/IV	-		-	

Table S4 Pathway enrichment analysis for 66 IRGs including KEGG and GO database

Database	ID	Pathway	GeneRatio	pvalue	p.adjust
KEGG	hsa04060	Cytokine-cytokine receptor interaction	13/55	5.58E-08	9.15E-06
KEGG	hsa04010	MAPK signaling pathway	10/55	2.40E-05	1.97E-03
KEGG	hsa04062	Chemokine signaling pathway	8/55	4.13E-05	2.26E-03
KEGG	hsa04151	PI3K-Akt signaling pathway	10/55	1.16E-04	3.62E-03
KEGG	hsa05340	Primary immunodeficiency	4/55	1.20E-04	3.62E-03
KEGG	hsa05212	Pancreatic cancer	5/55	1.56E-04	3.62E-03
KEGG	hsa04014	Ras signaling pathway	8/55	1.56E-04	3.62E-03
KEGG	hsa04612	Antigen processing and presentation	5/55	1.76E-04	3.62E-03
KEGG	hsa04662	B cell receptor signaling pathway	5/55	2.23E-04	4.07E-03
KEGG	hsa04350	TGF-beta signaling pathway	5/55	4.22E-04	6.93E-03
KEGG	hsa05215	Prostate cancer	5/55	4.88E-04	7.28E-03
KEGG	hsa04061	Viral protein interaction with cytokine and cytokine receptor	5/55	5.62E-04	7.68E-03
KEGG	hsa04660	T cell receptor signaling pathway	5/55	6.72E-04	8.48E-03
KEGG	hsa04213	Longevity regulating pathway - multiple species	4/55	8.03E-04	9.40E-03
KEGG	hsa05211	Renal cell carcinoma	4/55	1.20E-03	1.31E-02
KEGG	hsa05218	Melanoma	4/55	1.41E-03	1.44E-02
KEGG	hsa05220	Chronic myeloid leukemia	4/55	1.72E-03	1.66E-02
KEGG	hsa04960	Aldosterone-regulated sodium reabsorption	3/55	1.97E-03	1.80E-02
KEGG	hsa04915	Estrogen signaling pathway	5/55	2.38E-03	1.93E-02
KEGG	hsa05418	Fluid shear stress and atherosclerosis	5/55	2.45E-03	1.93E-02
KEGG	hsa05205	Proteoglycans in cancer	6/55	2.56E-03	1.93E-02
KEGG	hsa04012	ErbB signaling pathway	4/55	2.59E-03	1.93E-02
KEGG	hsa05210	Colorectal cancer	4/55	2.71E-03	1.93E-02
KEGG	hsa05235	PD-L1 expression and PD-1 checkpoint pathway in cancer	4/55	3.07E-03	2.08E-02
KEGG	hsa04810	Regulation of actin cytoskeleton	6/55	3.17E-03	2.08E-02
KEGG	hsa05226	Gastric cancer	5/55	3.32E-03	2.09E-02
KEGG	hsa04672	Intestinal immune network for IgA production	3/55	4.42E-03	2.68E-02
KEGG	hsa04064	NF-kappa B signaling pathway	4/55	5.36E-03	3.02E-02
KEGG	hsa04625	C-type lectin receptor signaling pathway	4/55	5.36E-03	3.02E-02
KEGG	hsa05225	Hepatocellular carcinoma	5/55	5.52E-03	3.02E-02
KEGG	hsa04360	Axon guidance	5/55	7.54E-03	3.99E-02
KEGG	hsa04080	Neuroactive ligand-receptor interaction	7/55	7.99E-03	4.09E-02
KEGG	hsa04929	GnRH secretion	3/55	9.30E-03	4.62E-02

GO(BP)	GO:0010469	regulation of signaling receptor activity	20/65	1.51E-13	3.15E-10
GO(BP)	GO:0001525	angiogenesis	12/65	5.18E-06	4.41E-03
GO(BP)	GO:0050900	leukocyte migration	11/65	7.23E-06	4.41E-03
GO(BP)	GO:0043410	positive regulation of MAPK cascade	12/65	9.13E-06	4.41E-03
GO(BP)	GO:2001236	regulation of extrinsic apoptotic signaling pathway	7/65	1.06E-05	4.41E-03
GO(BP)	GO:0051897	positive regulation of protein kinase B signaling	7/65	1.28E-05	4.45E-03
GO(BP)	GO:0071621	granulocyte chemotaxis	6/65	2.72E-05	8.11E-03
GO(BP)	GO:0097530	granulocyte migration	6/65	4.78E-05	1.13E-02
GO(BP)	GO:0071674	mononuclear cell migration	5/65	4.88E-05	1.13E-02
GO(BP)	GO:0043542	endothelial cell migration	7/65	5.65E-05	1.18E-02
GO(BP)	GO:0051896	regulation of protein kinase B signaling	7/65	6.99E-05	1.24E-02
GO(BP)	GO:0090132	epithelium migration	8/65	7.44E-05	1.24E-02
GO(BP)	GO:0072676	lymphocyte migration	5/65	8.16E-05	1.24E-02
GO(BP)	GO:0090130	tissue migration	8/65	8.79E-05	1.24E-02
GO(BP)	GO:0071356	cellular response to tumor necrosis factor	7/65	9.80E-05	1.24E-02
GO(BP)	GO:0001667	ameboidal-type cell migration	9/65	1.08E-04	1.24E-02
GO(BP)	GO:0002429	immune response-activating cell surface receptor signaling pathway	8/65	1.18E-04	1.24E-02
GO(BP)	GO:0048247	lymphocyte chemotaxis	4/65	1.28E-04	1.24E-02
GO(BP)	GO:0030593	neutrophil chemotaxis	5/65	1.29E-04	1.24E-02
GO(BP)	GO:2001237	negative regulation of extrinsic apoptotic signaling pathway	5/65	1.29E-04	1.24E-02
GO(BP)	GO:0097191	extrinsic apoptotic signaling pathway	7/65	1.31E-04	1.24E-02
GO(BP)	GO:0006869	lipid transport	8/65	1.31E-04	1.24E-02
GO(BP)	GO:0070374	positive regulation of ERK1 and ERK2 cascade	7/65	1.39E-04	1.24E-02
GO(BP)	GO:0043491	protein kinase B signaling	7/65	1.48E-04	1.24E-02
GO(BP)	GO:0070371	ERK1 and ERK2 cascade	8/65	1.49E-04	1.24E-02
GO(BP)	GO:0034612	response to tumor necrosis factor	7/65	1.57E-04	1.26E-02
GO(BP)	GO:1901889	negative regulation of cell junction assembly	3/65	1.85E-04	1.41E-02
GO(BP)	GO:0002223	stimulatory C-type lectin receptor signaling pathway	4/65	1.92E-04	1.41E-02
GO(BP)	GO:1990266	neutrophil migration	5/65	1.96E-04	1.41E-02
GO(BP)	GO:0002253	activation of immune response	10/65	2.09E-04	1.46E-02
GO(BP)	GO:0002768	immune response-regulating cell surface receptor signaling pathway	8/65	2.25E-04	1.51E-02
GO(BP)	GO:0002220	innate immune response activating cell surface receptor signaling pathway	4/65	2.40E-04	1.51E-02

GO(BP)	GO:0019882	antigen processing and presentation	6/65	2.45E-04	1.51E-02
GO(BP)	GO:0010876	lipid localization	8/65	2.46E-04	1.51E-02
GO(BP)	GO:0002685	regulation of leukocyte migration	6/65	2.54E-04	1.51E-02
GO(BP)	GO:0043406	positive regulation of MAP kinase activity	7/65	2.70E-04	1.56E-02
GO(BP)	GO:0031099	regeneration	6/65	2.81E-04	1.59E-02
GO(BP)	GO:0002757	immune response-activating signal transduction	9/65	3.10E-04	1.70E-02
GO(BP)	GO:0090288	negative regulation of cellular response to growth factor stimulus	5/65	3.71E-04	1.99E-02
GO(BP)	GO:1905952	regulation of lipid localization	5/65	4.21E-04	2.15E-02
GO(BP)	GO:0097529	myeloid leukocyte migration	6/65	4.27E-04	2.15E-02
GO(BP)	GO:0071347	cellular response to interleukin-1	5/65	4.38E-04	2.15E-02
GO(BP)	GO:0044344	cellular response to fibroblast growth factor stimulus	5/65	4.57E-04	2.15E-02
GO(BP)	GO:0007492	endoderm development	4/65	4.63E-04	2.15E-02
GO(BP)	GO:0010631	epithelial cell migration	7/65	4.63E-04	2.15E-02
GO(BP)	GO:0002764	immune response-regulating signaling pathway	9/65	4.90E-04	2.22E-02
GO(BP)	GO:0019884	antigen processing and presentation of exogenous antigen	5/65	5.56E-04	2.47E-02
GO(BP)	GO:0071774	response to fibroblast growth factor	5/65	5.77E-04	2.48E-02
GO(BP)	GO:0055024	regulation of cardiac muscle tissue development	4/65	5.83E-04	2.48E-02
GO(BP)	GO:1901214	regulation of neuron death	7/65	6.15E-04	2.57E-02
GO(BP)	GO:0070372	regulation of ERK1 and ERK2 cascade	7/65	6.29E-04	2.58E-02
GO(BP)	GO:0001889	liver development	5/65	6.71E-04	2.69E-02
GO(BP)	GO:0030595	leukocyte chemotaxis	6/65	6.82E-04	2.69E-02
GO(BP)	GO:0043524	negative regulation of neuron apoptotic process	5/65	7.21E-04	2.75E-02
GO(BP)	GO:0007160	cell-matrix adhesion	6/65	7.41E-04	2.75E-02
GO(BP)	GO:0061008	hepaticobiliary system development	5/65	7.47E-04	2.75E-02
GO(BP)	GO:0045860	positive regulation of protein kinase activity	9/65	7.62E-04	2.75E-02
GO(BP)	GO:0031100	animal organ regeneration	4/65	7.63E-04	2.75E-02
GO(BP)	GO:0048638	regulation of developmental growth	7/65	8.23E-04	2.91E-02
GO(BP)	GO:0071902	positive regulation of protein serine/threonine kinase activity	7/65	8.77E-04	3.05E-02
GO(BP)	GO:0070555	response to interleukin-1	5/65	9.20E-04	3.15E-02
GO(BP)	GO:0010594	regulation of endothelial cell migration	5/65	9.52E-04	3.20E-02
GO(BP)	GO:0090049	regulation of cell migration involved in sprouting angiogenesis	3/65	9.99E-04	3.31E-02
GO(BP)	GO:0070098	chemokine-mediated signaling pathway	4/65	1.03E-03	3.35E-02
GO(BP)	GO:0051051	negative regulation of transport	8/65	1.12E-03	3.60E-02

GO(BP)	GO:0042035	regulation of cytokine biosynthetic process	4/65	1.18E-03	3.73E-02
GO(BP)	GO:0043405	regulation of MAP kinase activity	7/65	1.20E-03	3.73E-02
GO(BP)	GO:0034605	cellular response to heat	4/65	1.23E-03	3.77E-02
GO(BP)	GO:0070997	neuron death	7/65	1.27E-03	3.77E-02
GO(BP)	GO:0002686	negative regulation of leukocyte migration	3/65	1.27E-03	3.77E-02
GO(BP)	GO:0033674	positive regulation of kinase activity	9/65	1.29E-03	3.77E-02
GO(BP)	GO:0006959	humoral immune response	6/65	1.30E-03	3.77E-02
GO(BP)	GO:0035987	endodermal cell differentiation	3/65	1.37E-03	3.92E-02
GO(BP)	GO:1990868	response to chemokine	4/65	1.41E-03	3.92E-02
GO(BP)	GO:1990869	cellular response to chemokine	4/65	1.41E-03	3.92E-02
GO(BP)	GO:0032368	regulation of lipid transport	4/65	1.53E-03	4.21E-02
GO(BP)	GO:0048732	gland development	8/65	1.57E-03	4.23E-02
GO(BP)	GO:0001706	endoderm formation	3/65	1.59E-03	4.23E-02
GO(BP)	GO:0008543	fibroblast growth factor receptor signaling pathway	4/65	1.60E-03	4.23E-02
GO(BP)	GO:0042535	positive regulation of tumor necrosis factor biosynthetic process	2/65	1.68E-03	4.34E-02
GO(BP)	GO:1903587	regulation of blood vessel endothelial cell proliferation involved in sprouting angiogenesis	2/65	1.68E-03	4.34E-02
GO(BP)	GO:0042089	cytokine biosynthetic process	4/65	1.88E-03	4.73E-02
GO(BP)	GO:0046854	phosphatidylinositol phosphorylation	4/65	1.88E-03	4.73E-02
GO(BP)	GO:0002694	regulation of leukocyte activation	8/65	1.93E-03	4.78E-02
GO(BP)	GO:0042107	cytokine metabolic process	4/65	1.96E-03	4.80E-02
GO(MF)	GO:0048018	receptor ligand activity	20/65	6.95E-15	1.56E-12
GO(MF)	GO:0030545	receptor regulator activity	20/65	2.18E-14	2.46E-12
GO(MF)	GO:0005125	cytokine activity	10/65	4.30E-08	3.22E-06
GO(MF)	GO:0001968	fibronectin binding	4/65	6.34E-06	3.56E-04
GO(MF)	GO:0008083	growth factor activity	7/65	1.83E-05	7.49E-04
GO(MF)	GO:0048020	CCR chemokine receptor binding	4/65	2.00E-05	7.49E-04
GO(MF)	GO:0008009	chemokine activity	4/65	5.97E-05	1.77E-03
GO(MF)	GO:0019838	growth factor binding	6/65	6.30E-05	1.77E-03
GO(MF)	GO:0050431	transforming growth factor beta binding	3/65	1.11E-04	2.78E-03
GO(MF)	GO:0042379	chemokine receptor binding	4/65	1.64E-04	3.46E-03
GO(MF)	GO:0005179	hormone activity	5/65	1.76E-04	3.46E-03
GO(MF)	GO:0017134	fibroblast growth factor binding	3/65	1.84E-04	3.46E-03
GO(MF)	GO:0005126	cytokine receptor binding	7/65	3.67E-04	6.28E-03
GO(MF)	GO:0046934	phosphatidylinositol-4,5-bisphosphate 3-kinase activity	4/65	4.08E-04	6.28E-03

GO(MF)	GO:0019955	cytokine binding	5/65	4.19E-04	6.28E-03
GO(MF)	GO:0052813	phosphatidylinositol bisphosphate kinase activity	4/65	4.89E-04	6.87E-03
GO(MF)	GO:0035004	phosphatidylinositol 3-kinase activity	4/65	7.21E-04	9.54E-03
GO(MF)	GO:0016303	1-phosphatidylinositol-3-kinase activity	3/65	1.70E-03	2.12E-02
GO(MF)	GO:0052742	phosphatidylinositol kinase activity	3/65	2.35E-03	2.78E-02
GO(MF)	GO:0050840	extracellular matrix binding	3/65	2.50E-03	2.81E-02
GO(MF)	GO:0030169	low-density lipoprotein particle binding	2/65	2.75E-03	2.94E-02
GO(MF)	GO:0003707	steroid hormone receptor activity	3/65	3.32E-03	3.39E-02
GO(MF)	GO:0044183	protein binding involved in protein folding	2/65	3.60E-03	3.52E-02
GO(MF)	GO:0051787	misfolded protein binding	2/65	5.07E-03	4.75E-02
