

Fluorophore	NK	Monocyte	T	B	Treg (Fix/Perm)	Perforin/ Granzyme (Fix/Perm)
FITC or Alexa Fluor 488	CD56 NCAM16.2 (BD)	PD-1 MIH4 (EB)	PD-1 MIH4 (EB)	Lambda MHL-38 (BL)	CD25 2A3 (BD)	Perforin dG9 (BL)
PE	NKp44 P44-8 (BL)	CD14 HCD14 (BL)	CD62L DREG-56 (BL)	Kappa MHK-49 (BL)		Granzyme B GB11 (BL)
APC	NKG2D 1D11 (BL)	CD56 NCAM16.2 (BD)	CD56 NCAM16.2 (BD)	CD5 UCHT2 (BL)	FoxP3 206D (EB)	CD56 NCAM16.2 (BD)
PerCP/Cy5.5	CD45 2D1 (BD)	CD45 2D1 (BD)	CD45 2D1 (BD)	CD45 2D1 (BD)	CD45 2D1 (BD)	CD45 2D1 (BD)
Cy7/APC or APCH7	CD3 SK7 (BD)	CD3 SK7 (BD)	CD3 SK7 (BD)	CD10 HI10a (BL)	CD3 SK7 (BD)	CD3 SK7 (BD)
AlexaFluor 700		CD16 3G8 (BL)	CD8 HIT8a (BL)	CD27 O323 (BL)	CD8 HIT8a (BL)	CD8 HIT8a (BL)
Cy7/PE		CD19 HIB19 (EB)	CD45RA L48 (BD)	CD19 HIB19 (EB)		
PerCP	Propidium Iodide 100 ng/ml	Propidium Iodide 100 ng/ml	Propidium Iodide 100 ng/ml	Propidium Iodide 100 ng/ml		
Pacific Blue	CD69 FN50 (BL)	CD69 FN50 (BL)	CD4 OKT4 (BL)	CD20 L27 (BD)	CD4 OKT4 (BL)	CD4 OKT4 (BL)

Supplementary Table S1 - Antibody Staining Panel for Flow Cytometry. Each column represents a pool of antibodies used to stain an aliquot of PBMC with the cell type focus listed in the header. Rows designate the fluorophore label of individual antibodies as listed in the first column. The target antigens are listed in bold, followed by hybridoma clone and source (BD = BD Biosciences, San Jose, CA; EB = eBiosciences/Affymetrix, San Diego, CA; BL = Biolegend, San Diego, CA).

Myeloid Cell Parameters	Lymphocyte Counts	T cell Parameters
Classical Monocyte %	Total Lymphocytes per μ l	CD56+ % of T
Intermediate Monocyte %	CD3+ T cells per μ l	CD4+ Central Memory
Non-Classical Monocytes %	CD8+ T cells per μ l	CD4+ Effector
Classical Monocyte PD-1 GMFI	CD4+ cells per μ l	CD4+ Effector Memory
Intermediate Monocyte PD-1 GMFI	CD4+ CD8+ cells per μ l	CD4+ Naïve
Non-Classical Monocytes PD-1 GMFI	NK cells per μ l	CD8+ Central Memory
Classical Monocyte CD69 GMFI	B cells per μ l	CD8+ Effector
Intermediate Monocyte CD69 GMFI	CD4/CD8 ratio	CD8+ Effector Memory
Non-Classical Monocyte CD69 GMFI	CD4+ Central Memory Count	CD8+ Naïve
Granulocytes	CD4+ Effector Count	NKG2D %+
Granulocytes PD1 GMFI	CD4+ Effector Memory Count	NKG2D+ GMFI
Granulocytes CD69 GMFI	CD4+ Naïve Count	PD-1 GMFI
NK Cell Parameters	CD8+ Central Memory Count	CD4+ PD-1 GMFI
% CD56 ^{bright}	CD8+ Effector Count	CD8+ PD-1 GMFI
CD56 ^{dim} NKG2D GMFI	CD8+ Effector Memory Count	CD56+ T PD-1 GMFI
CD56 ^{bright} NKG2D GMFI	CD8+ Naïve Count	CD4+ Central Memory PD-1 GMFI
CD56 ^{dim} NKp44 %+	B Cell Parameters	CD4+ Effector PD-1 GMFI
CD56 ^{bright} NKp44 %+	CD20+ CD10+ Immature B cells	CD4+ Naïve PD-1 GMFI
CD56 ^{dim} CD69 GMFI	CD20+ CD27- Naïve B cells	CD8+ Central memory PD-1 GMFI
CD56 ^{bright} CD69 GMFI	CD20+ CD27+ Memory B cells	CD8+ Effector PD-1 GMFI
CD56 ^{dim} Perforin GMFI	CD20- CD27+ Plasma cells	CD8+ Effector Memory PD-1 GMFI
CD56 ^{bright} Perforin GMFI		CD8+ Naïve PD-1 GMFI
CD56 ^{dim} Granzyme B GMFI		CD4+ Treg (FoxP3+)
CD56 ^{bright} Granzyme B GMFI		
CD56 ^{dim} PD-1 GMFI		
CD56 ^{bright} PD-1 GMFI		
CD56 ^{dim} CD16 GMFI		
CD56 ^{bright} CD16 GMFI		
CD56 ^{dim} NKp44 GMFI		
CD56 ^{bright} NKp44 GMFI		

Supplementary Table S2. Specific immune parameters analyzed in peripheral blood by flow cytometry.

Patient #	Sample collection	Blood collection	Time after blood	PD-1 score	PD-L1 score	Description	Pathology report
2	Month 22	Month 12	10 months	0	0	skin, punch biopsy, left breast	carcinoma infiltrating dermis and dermal lymphatics consistent with breast primary
2	Month 12	Month 12	same day*	0	0	Skin, right scapula, punch biopsy	Poorly differentiated adenocarcinoma in dermal lymphatics
3	Month 34	Month 20	15 months	1	0	Right inframammary crease, punch biopsy	Dermis and dermal lymphatics with high grade carcinoma, consistent with breast origin
6	Month 8	Month 8	same day	1	2	skin, right upper chest, punch biopsy	carcinoma in dermis
7	Month 21	Month 21	8 days	0	0	skin, left posterior shoulder, medial	High grade carcinoma in dermis and dermal lymphatics
9	Month 4	Month 4	1 day	3	3	skin, right lateral breast, punch biopsy	Ductal carcinoma within dermal lymphatics and invading dermis
9	Month 4	Month 4	20 days	1	4	4R lymph nodes, excision	metastatic carcinoma in 6/6 lymph nodes
10	Month 32	Month 32	same day	1	2	skin, left chest wall, punch biopsy	carcinoma within dermal lymphatics, consistent with the patient's history of ductal carcinoma
11	Month 17	Month 13	4 months	3	4	skin, right chest wall, punch biopsy	Carcinoma with dermal lymphatics consistent with breast primary (ductal carcinoma)
11	Month 20	Month 13	5 months	2	4	tumor- right chest wall, punch biopsy	high grade carcinoma in dermal lymphatics consistent with breast primary (ductal carcinoma)
13	Month 1	Month 1	3 days	0	1	left breast core biopsy	invasive duct carcinoma grade III
13	Month 1	Month 1	3 days	2	2	left axillary mass, biopsy	Invasive ductal carcinoma grade III
14	Month 20	Month 13	7 months	0	4	liver, core needle biopsy	Metastatic carcinoma consistent with breast origin ER-PR-
15	Month 22	Month 22	4 weeks	0	1	left breast mass, core biopsy	Invasive ductal carcinoma, nuclear grade 3
16	Month 27	Month 26	3 days	2	4	skin, right breast, punch biopsy	High grade ductal carcinoma, involves the entire dermis
16	Month 32	Month 26	5 months	1	2	skin, right breast in area of mastectomy, punch biopsy	High grade ductal carcinoma, involves the dermis, with lymphatic invasion

*yellow highlighting designates IHC samples obtained within one month of blood draw

Supplementary Table S3. Pathological characteristics of 16 tumor samples from 11 of the metastatic IBC patients analyzed for PD-1 and PD-L1 by immunohistochemistry. Yellow highlights are IHC samples obtained within one month of blood that were used for correlation analysis shown in Figure 5.