Single-cell analysis of the T-cell receptor repertoire in untreated myeloma patients suggests potential myeloma-reactive CD8+ T cells are shared between blood and marrow

Authors

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Supp. Table 1 Summary of recovered Chromium™ 10x data following QC

Sample ID	Sample	Number of cell barcodes recovered	Ave. number of detected genes per cell	Ave. number of recovered UMIs per cell	Mito genes (%)	Total TCR Clonotypes observed (n/%)	Cells with matching TCR data (n/%)
NDMM #13	[!]BM	7871	732	1625	1.9	4,455 (100)	6551 (83.2)
	PB	7822	1267	3966	4.1	3,876 (100)	7395 (94.5)
NDMM #31	[!]BM	2429	375	256	3.22	3,103 (100)	2317 (95.4)
	PB	7558	1098	3218	5.29	4,838 (100)	6559 (86.8)
NDMM #43	BM	7120	1005	3317	6.51	5,063 (100)	6869 (96.5)
	PB	6497	1097	3690	7.13	2,975 (100)	6207 (95.5)
NDMM #63	BM	7824	1251	3572	5.23	3,259 (100)	7472 (95.5)
	PB	6275	1374	3933	4.85	1,239 (100)	6147 (98.0)

[!] Gene expression library failed QC, excluded from further analysis.

Key: BM, Bone Marrow; PB, Peripheral Blood; NDMM, Newly Diagnosed Multiple Myeloma; ID, Identification; Mito, Mitochondrial; n, number; TCR, T-cell Receptor; UMIs, Unique Molecular Identifiers.

Supp. Table 2 Top 10 clones ranked by size of expansion (raw data)

BM13					
Rank	**Clones**	**Proportion**	**Variable genes**	**CDR3 (aa)**	**Joining genes**
1	182	2.25%	*TRAV12-2;TRBV7-8*	CAVKGNAGKSTF;CASSLESGGTEAFF	*TRAJ27;TRBJ1-1*
2	109	1.35%	*TRAV14/DV4;TRBV6-6*	CAMREGAEGAQKLVF;CASSYPSSGSLYNEQFF	*TRAJ54;TRBJ2-1*
3	99	1.22%	*TRAV29/DV5;TRBV7-9*	CAYGAGSYQLTF;CASSLIERETDTQYF	*TRAJ28;TRBJ2-3*
4	88	1.09%	*TRAV38-2/DV8;TRBV7-9*	CAYRRLGANNLFF;CASSPLDGRGTDTQYF	*TRAJ36;TRBJ2-3*
5	85	1.05%	*TRAV38-2/DV8;TRAV23/DV6;TRBV4-1*	CAYRGLFGNEKLTF;CAAKDPLMNRDDKIIF;CASSQDGTGEAKNIQYF	*TRAJ48;TRAJ30;TRBJ2-4*
6	63	0.78%	*TRAV12-2;TRBV2*	CAVMRDGGSQGNLIF;CASNRPQSGGSSYNSPLHF	*TRAJ42;TRBJ1-6*
7	49	0.61%	*TRBV10-3*	CAISEFGGTYEQYF	*TRBJ2-7*
8	48	0.59%	*TRAV38-2/DV8;TRBV4-1*	CAYRGLFGNEKLTF;CASSQDGTGEAKNIQYF	*TRAJ48;TRBJ2-4*
9	43	0.53%	*TRAV14/DV4;TRBV19*	CAMRDPSGGSYIPTF;CASSIDRAGAYNEQFF	*TRAJ6;TRBJ2-1*
10	39	0.48%	*TRAV13-1;TRBV3-1*	CAASPAQLTF;CASSQEGATGNEKLFF	*TRAJ22;TRBJ1-4*
PB13					
Rank	**Clones**	**Proportion**	**Variable genes**	**CDR3 (aa)**	**Joining genes**
1	209	2.50%	*TRAV38-2/DV8;TRBV7-9*	CAYRRLGANNLFF;CASSPLDGRGTDTQYF	*TRAJ36;TRBJ2-3*
2	141	1.69%	*TRAV13-1;TRBV3-1*	CAASPAQLTF;CASSQEGATGNEKLFF	*TRAJ22;TRBJ1-4*
3	111	1.33%	*TRAV24;TRBV7-6*	CAFIPNTNAGKSTF;CASSFLLAGADTQYF	*TRAJ27;TRBJ2-3*
4	110	1.32%	*TRAV29/DV5;TRBV2*	CAGSGGGNKLTF;CASSVGTDEQYF	*TRAJ10;TRBJ2-7*
5	101	1.21%	*TRAV5;TRBV6-5*	CAEMGNYGQNFVF;CASSPGALNYGYTF	*TRAJ26;TRBJ1-2*
6	101	1.21%	*TRAV12-1;TRBV5-1*	CVVRTDKLIF;CASSPDSQSSGNTIYF	*TRAJ34;TRBJ1-3*
7	93	1.11%	*TRAV12-2;TRBV7-8*	CAVKGNAGKSTF;CASSLESGGTEAFF	*TRAJ27;TRBJ1-1*
8	85	1.02%	*TRAV19;TRBV6-2*	CALSEADSGYSTLTF;CASSYWDGRVNTEAFF	*TRAJ11;TRBJ1-1*
9	71	0.85%	*TRAV12-2;TRBV2*	CAVMRDGGSQGNLIF;CASNRPQSGGSSYNSPLHF	*TRAJ42;TRBJ1-6*
10	69	0.83%	*TRAV20;TRBV9*	CAVRVGLTGGGNKLTF;CASSSLGDSNQPQHF	*TRAJ10;TRBJ1-5*

BM31					
Rank	**Clones**	**Proportion**	**Variable genes**	**CDR3 (aa)**	**Joining genes**
1	214	3.42%	*TRAV38-2/DV8;TRBV5-6*	CAYRRSNNDMRF;CASSLGAFTIYF	*TRAJ43;TRBJ1-3*
2	132	2.11%	*TRAV35;TRBV19*	CAGPVGDMRF;CASSVAGVRETQYF	*TRAJ43;TRBJ2-5*
3	130	2.08%	*TRAV12-1;TRBV9*	CVVNVLDMRF;CASSVGINEKLFF	*TRAJ43;TRBJ1-4*
4	72	1.15%	*TRAV20;TRBV2*	CAVQLDGQKLLF;CASRSRANVLTF	*TRAJ16;TRBJ2-6*
5	64	1.02%	*TRAV12-3;TRBV11-3*	CASWNSGYSTLTF;CASSLDGVTQFF	*TRAJ11;TRBJ2-1*
6	53	0.85%	*TRAV12-1;TRAV16;TRBV9*	CVVNVLDMRF;CSHNNAGNMLTF;CASSVGINEKLFF	*TRAJ43;TRAJ39;TRBJ1-4*
7	48	0.77%	*TRAV20;TRBV6-5*	CAVDMDSNYQLIW;CASSYGRAKGHFF	*TRAJ33;TRBJ2-1*
8	42	0.67%	*TRAV21;TRBV21-1;TRBV25-1*	CAAPNSGGGADGLTF;CASSKAAGQGSETQYF;CASSDSGANVLTF	*TRAJ45;TRBJ2-5;TRBJ2-6*
9	37	0.59%	*TRAV26-1;TRAV35;TRBV28*	CIVRGHGSSNTGKLIF;CAAPGTLTGNQFYF;CASSLGGPPAYGYTF	*TRAJ37;TRAJ49;TRBJ1-2*
10	35	0.56%	*TRAV14/DV4;TRBV19*	CAIENTDKLIF;CASSTRRAVGEKLFF	*TRAJ34;TRBJ1-4*

PB31					
Rank	**Clones**	**Proportion**	**Variable genes**	**CDR3 (aa)**	**Joining genes**
1	340	4.70%	*TRAV35;TRBV19*	CAGPVGDMRF;CASSVAGVRETQYF	*TRAJ43;TRBJ2-5*
2	95	1.31%	*TRBV19*	CASSVAGVRETQYF	*TRBJ2-5*
3	87	1.20%	*TRAV9-2;TRBV18*	CALSEIATGRRALTF;CAGSHPTGVLNGYTF	*TRAJ5;TRBJ1-2*
4	67	0.93%	*TRAV21;TRBV28*	CAAAGTGGFKTIF;CASSSSTSGSGLNEQFF	*TRAJ9;TRBJ2-1*
5	48	0.66%	*TRAV14/DV4;TRBV12-4*	CAMRERQAGTALIF;CASSGTANYGYTF	*TRAJ15;TRBJ1-2*
6	48	0.66%	*TRAV38-1;TRBV10-3*	CAFMKHSGFGNVLHC;CAISGGNTGELFF	*TRAJ35;TRBJ2-2*
7	45	0.62%	*TRAV13-2;TRBV7-9*	CAENTNAGKSTF;CASSLGGFEQYF	*TRAJ27;TRBJ2-7*
8	38	0.53%	*TRAV38-2/DV8;TRBV25-1*	CAYGLGGGADGLTF;CASTFGTGTDTQYF	*TRAJ45;TRBJ2-3*
9	33	0.46%	*TRAV12-2;TRBV4-3*	CAVNTGGFKTIF;CASSQDVASGAGELFF	*TRAJ9;TRBJ2-2*
10	33	0.46%	*TRAV38-2/DV8;TRBV7-9*	CAWWDNYGQNFVF;CASSHPDRVEKLFF	*TRAJ26;TRBJ1-4*

BM43

Rank	Clones	Proportion	Variable genes	CDR3 (aa)	Joining genes
1	345	4.32%	*TRAV26-2;TRBV19*	CILRDNGGKSTF;CASSIWGTSNQPQHF	*TRAJ27;TRBJ1-5*
2	192	2.40%	*TRAV8-6;TRBV7-3*	CAVRVDYKLSF;CASSPNPGDNYGYTF	*TRAJ20;TRBJ1-2*
3	164	2.05%	*TRAV21;TRBV6-5*	CAEGTGNQFYF;CASRGTGPPYEQYF	*TRAJ49;TRBJ2-7*
4	100	1.25%	*TRAV13-1;TRBV19*	CAAYDYKLSF;CASTGGRTDTQYF	*TRAJ20;TRBJ2-3*
5	79	0.99%	*TRAV17;TRBV4-1*	CATDAGSSGNTPLVF;CASSQAGRGTTYNEQFF	*TRAJ29;TRBJ2-1*
6	76	0.95%	*TRAV38-2/DV8;TRBV2*	CAYRSADSGYALNF;CASSQAVSGGNSPLHF	*TRAJ41;TRBJ1-6*
7	69	0.86%	*TRBV6-5*	CASRGTGPPYEQYF	*TRBJ2-7*
8	67	0.84%	*TRAV26-2;TRAV29/DV5;TRBV19*	CILRDNGGKSTF;CTSVQRPGNTGKLIF;CASSIWGTSNQPQHF	*TRAJ27;TRAJ37;TRBJ1-5*
9	60	0.75%	*TRBV19*	CASSIWGTSNQPQHF	*TRBJ1-5*
10	52	0.65%	*TRAV12-2;TRAV9-2;TRBV28*	CAALSWGKLQF;CAPYNTNAGKSTF;CASRVDRMSNQPQHF	*TRAJ24;TRAJ27;TRBJ1-5*

PB43

Rank	Clones	Proportion	Variable genes	CDR3 (aa)	Joining genes
1	1206	17.46%	*TRAV26-2;TRBV19*	CILRDNGGKSTF;CASSIWGTSNQPQHF	*TRAJ27;TRBJ1-5*
2	625	9.05%	*TRAV21;TRBV6-5*	CAEGTGNQFYF;CASRGTGPPYEQYF	*TRAJ49;TRBJ2-7*
3	253	3.66%	*TRAV26-2;TRAV29/DV5;TRBV19*	CILRDNGGKSTF;CTSVQRPGNTGKLIF;CASSIWGTSNQPQHF	*TRAJ27;TRAJ37;TRBJ1-5*
4	209	3.03%	*TRAV17;TRBV4-1*	CATDAGSSGNTPLVF;CASSQAGRGTTYNEQFF	*TRAJ29;TRBJ2-1*
5	151	2.19%	*TRBV19*	CASSIWGTSNQPQHF	*TRBJ1-5*
6	150	2.17%	*TRAV1-2;TRBV24-1*	CALRGDYKLSF;CATSDSPRTSGNNEQFF	*TRAJ20;TRBJ2-1*
7	141	2.04%	*TRDV1;TRAV21;TRBV6-5*	CSWGGQVMNYGGSQGNLIF;CAEGTGNQFYF;CASRGTGPPYEQYF	*TRAJ42;TRAJ49;TRBJ2-7*
8	110	1.59%	*TRBV6-5*	CASRGTGPPYEQYF	*TRBJ2-7*
9	104	1.51%	*TRAV1-2;TRAV10;TRBV24-1*	CALRGDYKLSF;CGEREGNARLMF;CATSDSPRTSGNNEQFF	*TRAJ20;TRAJ31;TRBJ2-1*
10	99	1.43%	*TRAV38-2/DV8;TRBV10-3*	CAYRSADSNYQLIW;CAISESVSGAGNTIYF	*TRAJ33;TRBJ1-3*

BM63

Rank	Clones	Proportion	Variable genes	CDR3 (aa)	Joining genes
1	372	4.04%	*TRAV19;TRBV20-1*	CALRTPTNSNSGYALNF;CSASGPAEINEQFF	*TRAJ41;TRBJ2-1*
2	311	3.38%	*TRAV12-1;TRBV28*	CVVNWRSNDYKLSF;CASSFPSGGVSTDTQYF	*TRAJ20;TRBJ2-3*
3	219	2.38%	*TRAV12-1;TRBV20-1*	CVVGDWFGDMRF;CSALKPGTSSYNEQFF	*TRAJ43;TRBJ2-1*
4	152	1.65%	*TRAV14/DV4;TRBV7-6*	CAMREVESNMLTF;CASSTFSYEQYF	*TRAJ39;TRBJ2-7*
5	148	1.61%	*TRAV8-1;TRBV11-2*	CAVNHYNTDKLIF;CASSLDYFSGNTIYF	*TRAJ34;TRBJ1-3*
[!]6	129	1.40%	*TRAV23/DV6;TRBV4-3*	CAASIGNFGNEKLTF;CASSPSRNTEAFF	*TRAJ48;TRBJ1-1*
7	107	1.16%	*TRAV14/DV4;TRBV12-4*	CAMREAITQGGSEKLVF;CASRGGWGSPLHF	*TRAJ57;TRBJ1-6*
8	101	1.10%	*TRAV8-6;TRBV27*	CAVSDHNNARLMF;CASSWVRGDTGELFF	*TRAJ31;TRBJ2-2*
9	98	1.06%	*TRBV20-1*	CSASGPAEINEQFF	*TRBJ2-1*
10	92	1.00%	*TRAV13-1;TRBV28*	CAAPVDFGNEKLTF;CASSQISGGNTIYF	*TRAJ48;TRBJ1-3*

PB63

Rank	Clones	Proportion	Variable genes	CDR3 (aa)	Joining genes
[!]1	359	5.32%	*TRAV23/DV6;TRBV4-3*	CAASIGNFGNEKLTF;CASSPSRNTEAFF	*TRAJ48;TRBJ1-1*
[@]2	224	3.32%	*TRAV8-6;TRAV8-6;TRBV28*	CAVPPTGGGNKLTF;CAVSDRSGGGADGLTF;CASSLGLHYEQYV	*TRAJ10;TRAJ45;TRBJ2-7*
[@]3	223	3.31%	*TRAV8-6;TRAV8-6;TRBV28*	CAVSDRSGGGADGLTF;CAVPPTGGGNKLTF;CASSLGLHYEQYV	*TRAJ45;TRAJ10;TRBJ2-7*
4	209	3.10%	*TRAV19;TRBV20-1*	CALWYGRDDKIIF;CSAKVNTEAFF	*TRAJ30;TRBJ1-1*
5	198	2.93%	*TRAV12-1;TRBV28*	CVVNWRSNDYKLSF;CASSFPSGGVSTDTQYF	*TRAJ20;TRBJ2-3*
6	189	2.80%	*TRAV13-1;TRBV28*	CAAPVDFGNEKLTF;CASSQISGGNTIYF	*TRAJ48;TRBJ1-3*
7	173	2.56%	*TRAV29/DV5;TRBV6-2*	CAARNTGNQFYF;CASSSRPGPSGYF	*TRAJ49;TRBJ2-7*
8	171	2.53%	*TRAV14/DV4;TRBV12-4*	CAMREAITQGGSEKLVF;CASRGGWGSPLHF	*TRAJ57;TRBJ1-6*
[@]9	156	2.31%	*TRAV8-6;TRAV8-6;TRBV28;TRBV3-1*	CAVPPTGGGNKLTF;CAVSDRSGGGADGLTF; CASSLGLHYEQYV;CISVPAAKSFPGTTSSYEQYF	*TRAJ10;TRAJ45;TRBJ2-7;TRBJ2-7*
[@]10	145	2.15%	*TRAV8-6;TRAV8-6;TRBV28;TRBV3-1*	CAVSDRSGGGADGLTF;CAVPPTGGGNKLTF; CASSLGLHYEQYV;CISVPAAKSFPGTTSSYEQYF	*TRAJ45;TRAJ10;TRBJ2-7;TRBJ2-7*

[!] Clone found to be specific (with 100% confidence) for the CMV derived epitope, RPHERNGFTVL by all tested methods. [@] Example of difficulties in reconstructing paired data; clone is identical differing only in order of the 2nd TCRα J gene and the inclusion of a second TCRβ.

Key: BM, Bone Marrow; PB, Peripheral Blood; CDR3, Complementarity-determining region 3

Supp. Table 3 Top 20 differentially expressed genes comparing the dominant clone in PT43 between the BM and PB

Gene	P value	Average Log$_2$FC	Proportion of cells expressing gene in the bone marrow (%)	Proportion of cells expressing gene in the peripheral blood (%)	Adjusted P value
HLA-A	4.41 X 10^{-59}	-0.70	99.1	100	7.31 X 10^{-55}
HLA-C	2.11 X 10^{-54}	-0.60	99.7	100	3.50 X 10^{-50}
PPDPF	4.11 X 10^{-54}	0.96	93.3	77.8	6.81 X 10^{-50}
SARAF	2.18 X 10^{-37}	1.01	72.9	47.4	3.6 X 10^{-33}
TRIR	2.19 X 10^{-37}	1.036	46.2	16.9	3.62 X 10^{-33}
HLA-B	4.06 X 10^{-35}	-0.40	100	100	6.72 X 10^{-31}
S100A10	1.06 X 10^{-34}	0.89	86.0	65.2	1.75 X 10^{-30}
C9orf16	4.91 X 10^{-34}	0.88	62.9	34.3	8.13 X 10^{-30}
VIM	3.19 X 10^{-32}	1.035	72.9	51.9	5.27 X 10^{-28}
NDUFB10	6.85 X 10^{-31}	0.87	51.4	24.9	1.13 X 10^{-26}
C12orf75	7.73 X 10^{-31}	0.921	59.6	33.3	1.28 X 10^{-26}
ISCU	4.40 X 10^{-30}	0.88	31.0	8.6	7.287 X 10^{-26}
PAXX	9.54 X 10^{-30}	0.90	62.0	35.6	1.58 X 10^{-25}
IL32	1.05 X 10^{-28}	-0.56	94.2	98.9	1.74 X 10^{-24}
CD52	7.25 X 10^{-25}	-0.466	97.6	99.5	1.20 X 10^{-20}
LIME1	1.27 X 10^{-24}	0.83	40.4	17.2	2.11 X 10^{-20}
CD8B	2.01 X 10^{-23}	0.68	73.3	54.3	3.33 X 10^{-19}
CALM1	9.24 X 10^{-21}	0.589	87.5	78.5	1.53 X 10^{-16}
GYPC	1.57 X 10^{-20}	0.71	26.4	8.8	2.60 X 10^{-16}

Average Log$_2$FC: log$_2$ fold-change of the average expression between the two groups. Positive values indicate that the feature is more highly expressed in the BM Adjusted P value is based on Bonferroni correction using all features in the dataset.

Key: PT, Patient; BM, Bone Marrow; PB, Peripheral Blood