

Supplementary Table 1. Fluorochrome-conjugated antibodies source.

Antibody Specificity	Format	Clone	Source	Catalog Number
Roswell Park Comprehensive Cancer Center				
CD138	BV421	MI15	BD Biosciences	562935
CD27	BV510	O323	BioLegend	302835
CD38	FITC	T16	Beckman Coulter	IM0775U
CD56	PE	C5.9	Cytognos	CYT-56PE
CD45	PerCP-Cy5.5	H130	BioLegend	304028
CD19	PECy7	J3-119	Beckman Coulter	IM3628U
CD117	APC	104D2	BD Biosciences	341106
CD81	APCH7	JS-81	BD Biosciences	656154
cKappa	APC	Polyclonal	Dako	C0222
cLambda	APC-C750	Polyclonal	Cytognos	CYT-LAC750
Mayo Clinic				
CD138	BV421	MI15	BD Biosciences	630005 ^a
CD27	BV510	O323	BioLegend	302835
CD38 (Multiepitope)	FITC	Multi-epitope	Cytognos	CYT-MM-MRD ^b
CD56	PE	C5.9	Cytognos	CYT-MM-MRD ^b
CD45	PerCP-Cy5.5	EO1	Cytognos	CYT-MM-MRD ^b
CD19	PECy7	SA287	Cytognos	CYT-MM-MRD ^b
CD117	APC	104D2	Cytognos	CYT-MM-MRD ^b
CD81	APC-C750	M38	Cytognos	CYT-MM-MRD ^b
cKappa	APC	Polyclonal	Cytognos	CYT-MM-MRD ^b
cLambda	APC-C750	Polyclonal	Cytognos	CYT-MM-MRD ^b

^a Customized reagent identical to item #562935 except for larger reagent size; may not be found on manufacturer's website

^b Items sold as lyophilized product in a kit

Supplementary Table 2. Interpretation of patient samples before and after adopting consensus gating and MRD levels.

Sample	Stage 1		Stage 2	
	Agreeability	MRD Level, % (Mean \pm SD)	Agreeability	MRD Level, % (Mean \pm SD)
MRD+				
#1	100	0.0063 \pm 0.0026	100	0.0066 \pm 0.0023
#2	100	0.0096 \pm 0.0033	100	0.0084 \pm 0.0028
#4	88.24	0.0019 \pm 0.0010	70.59	0.0015 \pm 0.0007
#5	64.71	0.0011 \pm 0.0007	64.71	0.0010 \pm 0.0006
#6	94.12	0.0660 \pm 0.0247	94.12	0.0559 \pm 0.0239
#7	88.24	0.1117 \pm 0.0548	94.12	
Mean, %	89.22		87.25	
SD, %	13.11		15.53	
MRD-				
#3	70.59	0.0034 \pm 0.0077	82.35	0.0027 \pm 0.0075
#8	64.71	0.0019 \pm 0.0031	76.47	0.0007 \pm 0.0013
Mean, %	67.65		79.41	
SD, %	4.16		4.16	

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Supplementary Table 3: Immunophenotypic profile of myeloma cells.

Samples	CD38	CD138	CD19	CD45	CD56	CD117	CD81	CD27	cKappa	cLambda
Stages 1 and 2^a										
#1	+	+	-	-	+	dim	-	dim	+	-
#2	+	+	-	dim	+	het	-	dim	+	-
#4	+	+	-	-	het	-	-	dim	-	-
#5	dim	+	-	-	dim	-	-	dim	+	-
#6	dim	+	-	dim	-	+	dim	dim	+	-
#7	dim	+	-	dim	+	-	-	het	+	-
Stage 3^b										
#1	dim	+	-	dim	br	+	-	dim	+	-
#3	+	+	-	dim	+	+	het	dim	+	-
#6	+	+	-	+	+	+	-	dim	-	+
#7	dim	+	-	dim	het	+	+	dim	+	-
#10	dim	+	-	dim	het	het	dim	dim	+	-

^a Samples 3 and 8 were considered as MM MRD negative

^b Samples 2, 4, 5, 8 and 9 were considered as MM MRD negative

Supplementary Table 4. Variation in the measurement of MM MRD related parameters between participants using consensus analysis method (Stage 2).

Sample No.	#1	#2	#3*	#4	#5	#6	#7	#8*	Mean CV
Total Cells Analyzed									
Mean	8,149,491	8,157,372	2,628,127	8,892,494	8,826,923	9,420,484	9,320,259	5,425,946	
SD	531,537	634,388	75,603	205,399	549,253	662,481	229,831	185,864	
CV, %	6.52	7.78	2.88	2.31	6.22	7.03	2.47	3.43	4.83
Total Normal PCs									
Mean	2,433	367	1,172	304	3,213	7,813	660	436	
SD	907	171	393	151	1491	2294	1,032	98	
CV, %	37.3	46.53	33.5	49.65	46.41	29.36	156.42	22.38	52.694
Total Abnormal PCs									
Mean	536	681	70	130	91	5,215	7,086	39	
SD	180	219	195	62	53	2,207	5,299	72	
CV, %	33.5	32.12	279.09	47.52	57.77	42.33	74.77	186.74	94.23
MRD Level, %									
Mean	0.00662	0.0084	0.00266	0.00146	0.00104	0.05589	0.0769	0.00071	
SD	0.00226	0.00279	0.00748	0.0007	0.00061	0.02385	0.05766	0.00134	
CV, %	34.14	33.22	280.6	47.48	58.01	42.67	74.99	187.7	94.851
Limit of Detection, %									
Mean	0.00025	0.00025	0.00076	0.00023	0.00023	0.00022	0.00022	0.00037	
SD	0.00002	0.00002	0.00002	0.00001	0.00001	0.00002	0.00001	0.00001	
CV, %	6.6	8.01	3.05	2.77	6.12	7.72	3.4	3.38	5.1313
Lower Limit of Quantification, %									
Mean	0.00123	0.00119	0.00358	0	0.00113	0.00112	0.00099	0.00109	
SD	0.00008	0.0001	0.00012	0	0.00003	0.00009	0.0001	0.00004	
CV, %	6.64	8.81	3.47	2.51	8.33	9.92	3.26	4.01	5.8688
Total Mast Cells									
Mean	79	87	18	63	199	252	172	38	
SD	22	45	11	34	37	51	104	35	
CV, %	27.51	52.04	63.68	54.02	18.5	20.26	60.44	92.04	48.561
Total Hematogones									
Mean	263	784	8,918	4,448	168	253,159	1,447	16,173	
SD	51	121	3,339	916	310	69,565	1,803	2,688	
CV, %	19.35	15.37	37.44	20.59	184.44	27.48	124.64	16.62	55.741

*Samples considered to be MRD-negative

Bold values represent maximum, minimum, and mean CVs

Supplementary Table 5. Variation in the measurement of MM MRD related parameters between participants using consensus analysis method (Stage 3)

Sample No.	#1*	#2*	#3*	#4*	#5*	#6	#7	#8	#9	#10	Mean CV
Total Cells Analyzed											
Mean	8,673,870	6,924,800	8,669,318	8,217,839	8,508,059	7,883,203	8,694,735	9,210,348	8,605,036	7597720	
SD	470,247	480,566	159,688	761,248	261,122	596,793	282,205	283,418	389,262	368,096	
CV, %	5.42	6.94	1.84	9.26	3.07	7.57	3.25	3.08	4.52	4.84	4.979
Total Normal PCs											
Mean	6,399	18779	1,060	3473	5,180	66,028	2125	2189	1560	638	
SD	1,301	3811	228	1159	1305	15981	283	415	249	175	
CV, %	20.33	20.29	21.49	33.37	25.19	24.2	13.34	18.95	15.94	27.44	22.05
Total Abnormal PCs											
Mean	4	780	22	5	222	111	169	184	208	66	
SD	7	1918	41	9	552	67	96	64	67	34	
CV, %	179.67	245.85	185.17	166.01	249.07	60.33	56.78	35.03	32.09	52.16	126.22
MRD Level, %											
Mean	0.00002	0.00485	0.00023	0.00005	0.00024	0.00130	0.00179	0.00203	0.00251	0.00086	
SD	0.00006	0.01268	0.00048	0.00008	0.00082	0.00072	0.00091	0.0007	0.00058	0.00045	
CV, %	280.31	261.35	211.23	178.67	339.44	55.7	50.52	34.8	22.94	52.69	148.77
Limit of Detection, %											
Mean	0.00023	0.00029	0.00023	0.00025	0.00024	0.00025	0.00023	0.00022	0.00023	0.00026	
SD	0.00001	0.00002	0	0.00002	0.00001	0.00002	0.00001	0.00001	0.00001	0.00001	
CV, %	5.9	7.52	1.86	8.54	2.62	8.23	3.26	3.46	4.44	4.45	5.028
Lower Limit of Quantification, %											
Mean	0.00058	0.00073	0.00073	0.00058	0.00061	0.00064	0.00057	0.00058	0.00054	0.00059	
SD	0.00003	0.00005	0.00005	0.00003	0.00005	0.00005	0.00002	0.00001	0.00002	0.00002	

CV, %	5.12	7.31	1.71	8.63	3.08	8.1	3.49	3.11	4.32	4.52	4.939
Total Mast Cells											
Mean	99	584	1062	37	171	962	121	415	150	171	
SD	29	137	52	25	37	103	80	70	34	102	
CV, %	29.51	23.39	4.88	67.07	21.72	10.69	66.48	16.89	22.86	59.61	32.31
Total Hematogones											
Mean	126,475	88,101	197,742	2,142	53,248	9,102	53,524	75,366	109,679	3,026	
SD	56,642	30,287	39,680	799	11,844	2,627	16,507	8,467	25,735	852	
CV, %	44.79	34.38	20.07	37.3	22.24	28.87	30.84	11.23	23.46	28.15	28.13

*Samples considered to be MRD-negative

Bold values represent maximum, minimum, and mean CVs

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