

Primer set ID	Animal ID	Primer name	Primer sequence	Reference	MHC gene
A		5'LPH III 3'UT RI	5' GCAAGCTTATGACGGTTCATGGCTCCCCGAACC3' 5' GCCAATTCAGTCCCACACAAGGCAGCTG3'	10	A/B/C
B		HLA-B-5'UT HLA-B-3'UTB	5' GGGCGTCGACGGACTCAGAATCTCCCAGACGCCGAG3' 5' CTGGGGAGGAAACACAGGTTCAGCATGGGAAC3'	9	B (A)
C		HLA-C-CLSP-23 HLA-C-3UTC	5' CCGCGTCGACTCAGATTCTCCCAGACGCCGAGATG3' 5' CCGCAAGCTTTCGGGGAGGGAACACAGGTTCAGTGTGGGGAC3'	9	C (B/A)
D		5'Popy cl I 3'Popy cl I	5' ATGGCGGTCATGGCGCC3' 5' TCACACTTACAAGCTGTGAGAGAC3'	Designed for this project	A/B/C
E		Popy454cl1-ex2-Forward Popy454cl1-ex4-Reverse	5' CACCAGGAGCACGCAAGGACCAGTTCGTGYGGTTCGACAGCGAC3' 5' GAGGACCACGAGCGAACCAGCCGCTGCCAGGTTCAGTGTGATCTCC3'	Designed for this project	A/B/C
	PPY1	MID-145-Roche-454-A MID-145-Roche-454-B MID-123-Roche-454-A MID-123-Roche-454-B	5' CGTATCGCCTCCCTCGCGCCA TCAGTCTGTGACATGC ACCAGGAGCACGCAAGGAC3' 5' CTATGCGCCTTGCCAGCCCGC TCAGTCTGTGACATGC AGGAGACCACGAGCGAACCAG3' 5' CGTATCGCCTCCCTCGCGCCA TCAGATCGTAGCAG CACCAGGAGCACGCAAGGAC3' 5' CTATGCGCCTTGCCAGCCCGC TCAGATCGTAGCAG GAGGACCACGAGCGAACCAG3'		
	Jinjing	MID-146-Roche-454-A MID-146-Roche-454-B MID-124-Roche-454-A MID-124-Roche-454-B	5' CGTATCGCCTCCCTCGCGCCA TCAGTCTGTGACATGC ACCAGGAGCACGCAAGGAC3' 5' CTATGCGCCTTGCCAGCCCGC TCAGTCTGTGACATGC AGGAGACCACGAGCGAACCAG3' 5' CGTATCGCCTCCCTCGCGCCA TCAGATCGTAGCAG CACCAGGAGCACGCAAGGAC3' 5' CTATGCGCCTTGCCAGCCCGC TCAGATCGTAGCAG GAGGACCACGAGCGAACCAG3'		
	Guchi	MID-151-Roche-454-A MID-151-Roche-454-B MID-147-Roche-454-A MID-147-Roche-454-B	5' CGTATCGCCTCCCTCGCGCCA TCAGTCTGTGACATGC ACCAGGAGCACGCAAGGAC3' 5' CTATGCGCCTTGCCAGCCCGC TCAGTCTGTGACATGC AGGAGACCACGAGCGAACCAG3' 5' CGTATCGCCTCCCTCGCGCCA TCAGTGTGACATGC CACCAGGAGCACGCAAGGAC3' 5' CTATGCGCCTTGCCAGCCCGC TCAGTGTGACATGC CAGGAGACCACGAGCGAACCAG3'		
	Jago	MID-149-Roche-454-A MID-149-Roche-454-B MID-148-Roche-454-A MID-148-Roche-454-B	5' CGTATCGCCTCCCTCGCGCCA TCAGTGTGACATGC ACCAGGAGCACGCAAGGAC3' 5' CTATGCGCCTTGCCAGCCCGC TCAGTGTGACATGC AGGAGACCACGAGCGAACCAG3' 5' CGTATCGCCTCCCTCGCGCCA TCAGTGTGACATGC CACCAGGAGCACGCAAGGAC3' 5' CTATGCGCCTTGCCAGCCCGC TCAGTGTGACATGC CAGGAGACCACGAGCGAACCAG3'		
	Katja	MID-127-Roche-454-A MID-127-Roche-454-B MID-121-Roche-454-A MID-121-Roche-454-B	5' CGTATCGCCTCCCTCGCGCCA TCAGCAGACGATAG CACCAGGAGCACGCAAGGAC3' 5' CTATGCGCCTTGCCAGCCCGC TCAGCAGACGATAG GAGGACCACGAGCGAACCAG3' 5' CGTATCGCCTCCCTCGCGCCA TCAGATAGCTCTCG CACCAGGAGCACGCAAGGAC3' 5' CTATGCGCCTTGCCAGCCCGC TCAGATAGCTCTCG GAGGACCACGAGCGAACCAG3'		
F		PopyMHCgeneric-ex2-Forward PopyMHCgeneric-ex3-Reverse	5' CAGTTCGTGTGGTTCGACAGCGAC3' 5' CCCACTTGGCTGGGTGATCTGAG3'	Designed for this project	A/B/C

Figure S1. Sets of primers used in the PCR amplifications for sequencing orangutan MHC class I using Sanger and 454-Roche platforms. Per animal, the Roche-454 specific primers are presented. Each primer pair, indicated by a three-digit number (A for the forward and B for the reverse), has its own specific multiplex identifier (MID) that is indicated in blue. The key sequence is indicated in red. The MHC genes indicated in brackets are genes that are also amplified with the respective set of primers although the primers were designed to be gene specific.

A

Species	Orangutan	Total # reads	# Reads FW	# Reads RV	Stringent filtering		Less stringent filtering	
					# Reads obtained	# Reads unassigned	# Reads obtained	# Reads unassigned
Poab	PPY1	9541	4190	5351	220	42	2990	261
	Jinjing	9765	4366	5399	205	21	2896	356
	Guchi	11983	5365	6618	206	4	3158	342
Popy	Jago	10344	4389	5955	205	11	2768	431
	Katja	8863	3840	5023	178	8	2640	303
	Total # reads	50496	22150	28346	1014	86	14452	1693

B

Species	Orangutan	Allele (C/T)	Stringent filtering data					Less stringent filtering data				
			# Reads		Transcription (%)		Average (%)	# Reads		Transcription (%)		Average (%)
			1st Amplicon	2nd Amplicon	1st Amplicon	2nd Amplicon		1st Amplicon	2nd Amplicon	1st Amplicon	2nd Amplicon	
Poab	PPY1	A*08:01 (C)	31	38	37.4	40	38.7	685	532	51.50	38.03	44.77
		A*06:01 (C)	28	27	33.7	28.4	31.1	448	465	33.68	33.24	33.46
		B*04:01:02 (T)	14	14	16.9	14.7	15.8	107	159	8.05	11.37	9.71
		B*10:01 (C)	10	14	12.1	14.7	13.4	67	170	5.04	12.15	8.59
		B*05:01 (T)	0	2	0	2.1	1.1	6	41	0.45	2.93	1.69
		B*08:04 (C)	0	0	0	0	0	10	17	0.75	1.22	0.98
		B*08:01 (C)	0	0	0	0	0	7	15	0.53	1.07	0.80
		B*03:01 (C)	0	0	0	0	0	0	0	0	0	0
	Jinjing	A*09:01 (C)	30	33	33.7	34.7	34.2	418	496	32.76	39.24	36.00
		A*04:01 (C)	26	26	29.2	27.4	28.3	325	389	25.47	30.78	28.12
		B*01:01 (T)	13	12	14.6	12.6	13.6	207	166	16.22	13.13	14.68
		B*02:01 (T)	8	12	9.0	12.6	10.8	201	127	15.75	10.05	12.90
		B*08:02 (C)	12	12	13.5	12.6	13.1	99	70	7.76	5.54	6.65
		B*03:02 (C)	0	0	0	0	0	7	4	0.55	0.32	0.43
		C*02:01 (C)	0	0	0	0	0	19	12	1.49	0.95	1.22
		Guchi	A*03:01 (C)	32	25	32.0	24.5	28.3	680	701	45.95	52.47
	B*06:01 (C)		27	30	27.0	29.4	28.2	295	256	19.93	19.16	19.55
	B*04:01:01 (T)		15	19	15.0	18.6	16.8	198	152	13.38	11.38	12.38
	B*02:01 (T)		9	14	9.0	13.7	11.4	149	132	10.07	9.88	9.97
	B*08:03 (C)		10	10	10.0	9.8	9.9	80	58	5.41	4.34	4.87
	B*08:01 (C)		7	4	7.0	3.9	5.5	49	35	3.31	2.62	2.97
	B*03:03 (C)		0	0	0	0	0	5	0	0.34	0	0.17
	C*02:01 (C)		0	0	0	0	0	24	2	1.62	0.15	0.89
	Popy	Jago	A*01:02 (C)	23	25	23.7	25.8	24.7	333	377	26.41	35.04
A*05:01 (C)			22	20	22.7	20.6	21.7	326	284	25.85	26.39	26.12
B*13:01 (T)			20	16	20.6	16.5	18.6	299	229	23.71	21.28	22.50
B*11:01 (C)			13	20	13.4	20.6	17.0	188	115	14.91	10.69	12.80
B*08:07 (C)			11	11	11.3	11.3	11.3	53	45	4.20	4.18	4.19
B*08:02 (C)			8	5	8.3	5.2	6.7	59	26	4.68	2.42	3.55
B*03:07 (C)			0	0	0	0	0	3	0	0.24	0	0.12
Katja			A*06:01 (C)	33	33	41.3	36.7	39.0	387	515	42.86	35.91
		A*05:02 (C)	24	25	30.0	27.8	28.9	320	404	35.44	28.17	31.81
		B*04:02 (T)	14	18	17.5	20.0	18.8	152	338	16.83	23.57	20.20
		B*05:02 (T)	4	6	5.0	6.7	5.8	25	65	2.77	4.53	3.65
		B*08:02 (C)	5	8	6.3	8.9	7.6	17	54	1.88	3.77	2.82
		B*03:03 (C)	0	0	0	0	0	2	22	0.22	1.53	0.88
		C*02:03 (C)	0	0	0	0	0	0	36	0	2.51	1.26

C

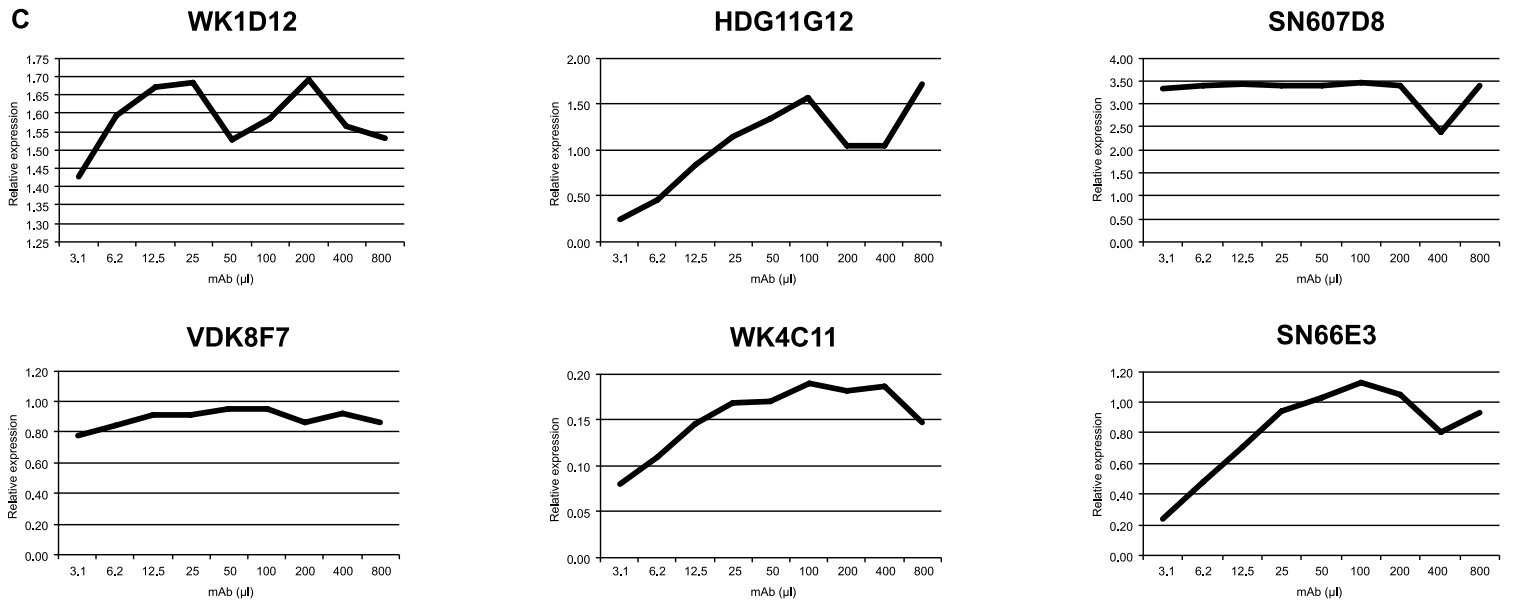


Figure S2. Next generation sequencing data and saturation measurements for the six monoclonal antibodies. (A) The total number of reads obtained for the orangutans, and the numbers analyzed for the stringent and the less stringent filtering's. Poab and Popy signify *Pongo abelii* and *Pongo pygmaeus*, respectively. # Indicates number; FW indicates forward; RV indicates reverse. (B) Distribution of the number of reads detected per MHC allele in each orangutan for the stringent and the less stringent filtering. From these data the percentage transcription is calculated, and the average percentage transcription is provided (Fig. 3). In the third column the distribution of the bases C and T at position 175 in the orangutan MHC class I sequences is indicated. (C) Overview of the titration experiments for the six monoclonal antibodies (mAbs) determining the concentration of mAb that gives saturation or near saturation. Each monoclonal is tested against its preferred allele expressed by a human B-cell-line (WK1D12: HLA-B*27:05 expressed by the cell-line BTB; HDG11G12: HLA-B*51:01, cell-line Tubo; SN607D8, HLA-A*02, cell-line JY; VDK8F7: HLA-B*27:05, cell-line BTB; WK4C11: HLA-C*01:02, cell-line BTB; SN66E3: HLA-A*02, cell-line JY). Note that the reactivity of WK4C11 with HLA-C*01:02 in BTB is higher than with HLA-C*07:02 in JY, which is in accordance with the reactivities observed with these alleles in the OneLambda single antigen bead test (Hiby S.E. et al. 2010. J Clin Invest 120: 4102-10).

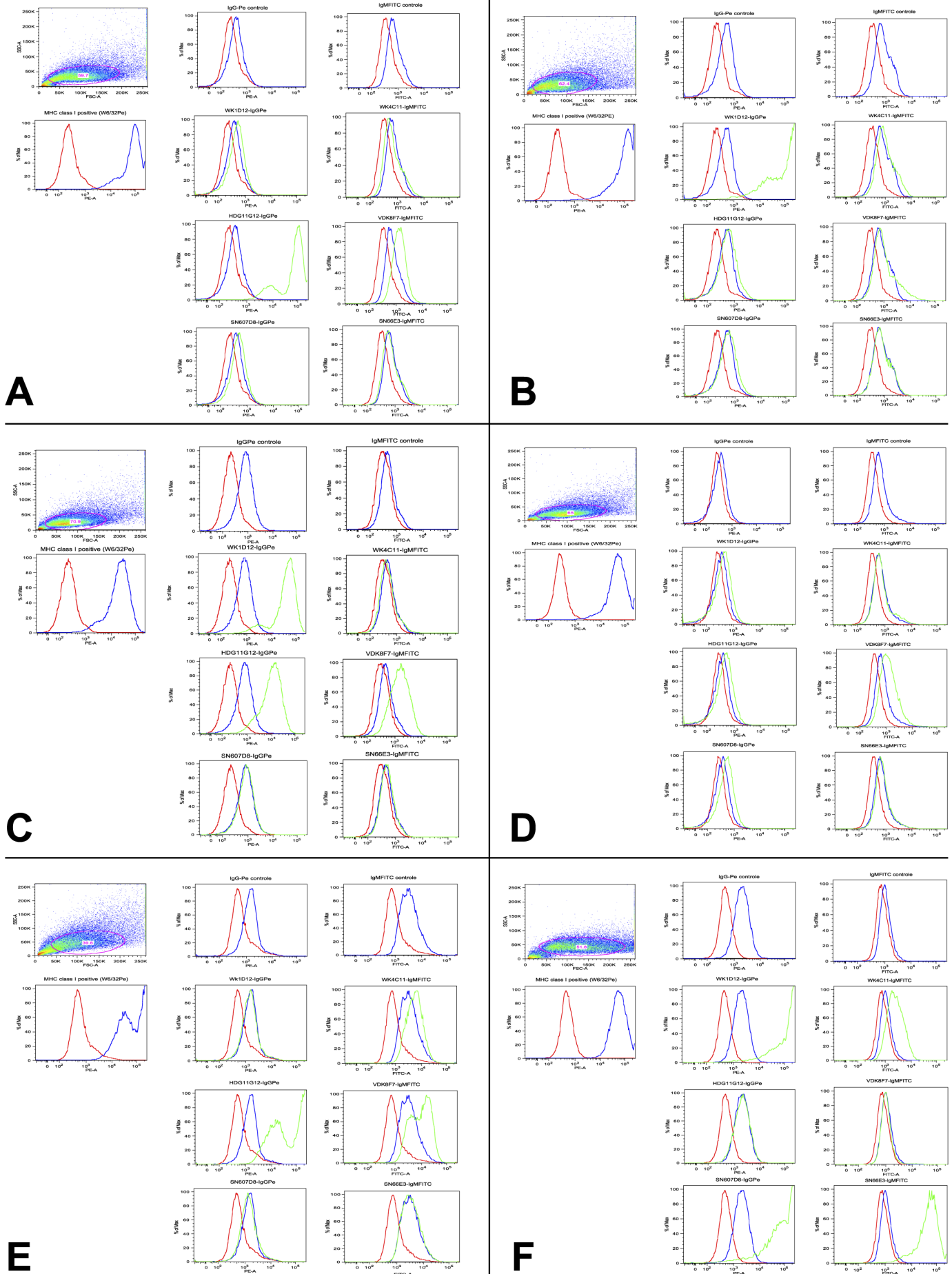


Figure S4. Representative plots of the flow cytometry results of the six human mAbs tested against the B-cell line of (A) PPY1, (B) Jinjing, (C) Guchi, (D) Jago, (E) Katja, and (F) JY. The total MHC class I expression is determined using the anti HLA-ABC/R-PE (first column). The second and third columns represent respectively the IgG (Pe labeled) and IgM (FITC labeled) based mAbs. The red line represents the auto-fluorescence, the blue line represents either the IgG or the IgM second step conjugate control, and the green line represents the indicated mAb specific expression.