

UK NEQAS for H&I Scheme 2B - Crossmatching by Flow Cytometry

T-CELL AND B-CELL FLOW CYTOMETRY RESULTS OF SAMPLE 2B03/2017 (COMPARED TO LOCAL NEGATIVE CONTROL)

DISPATCHED ON 14TH MARCH 2017

HLA PHENOTYPE OF BLOOD DONOR: HLA-A2, A3; B7, B57; Cw6, Cw7; DR4, DR15; DQ6, DQ8

Summary of Results												
T-cells					B-cells							
Total tested	42	52	44	52	42	48	47	43				
Positive	11	50	8	1	2	48	46	18				
Negative	31	2	36	51	40	0	1	25				
NT/Equivocal	19	9	17	9	13	7	8	12				
% Positive	26.2%	96.2%	18.2%	1.9%	4.8%	100.0%	97.9%	41.9%				
% Negative	73.8%	3.8%	81.8%	98.1%	95.2%	0.0%	2.1%	58.1%				
Consensus	Not Assessed	Positive	Negative	Negative	Negative	Positive	Positive	Not Assessed				
HLA Antibody Specificity (Defined by CDC)	A11	A2 A28 A9	DR4 DR9	B44	A11	A2 A28 A9	DR4 DR9	B44				
Lab No.	Serum 1	Serum 2	Serum 3	Serum 4	Serum 1	Serum 2	Serum 3	Serum 4	Date Received	Date Tested	Viability (%)	Comments
9	Equivocal	Positive	Negative	Negative	Equivocal	Positive	Positive	Positive	15-Mar	16-Mar	95	
11	Equivocal	Positive	Equivocal	Negative	Equivocal	Positive	Positive	Positive	15-Mar	16-Mar		
14	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Positive	15-Mar	15-Mar		
15	Positive	Positive	Positive	Negative	Negative	Positive	Positive	Positive	15-Mar	15-Mar		Sample amount very small, only able to test singly. This would not happen in a clinical setting
19	Positive	Positive	Negative	Negative	Negative	Positive	Positive	Positive	15-Mar	16-Mar		
20	Positive	Positive	Positive	Negative	Positive	Positive	Positive	Positive	15-Mar	16-Mar		
24	Equivocal	Positive	Negative	Negative	Equivocal	Positive	Positive	Positive	16-Mar	17-Mar	95	Serum 1 T/B cell result is reported as equivocal as the result falls within the uncertainty of measurement and we would await HLA antibody results if this was a patient sample.
25	Positive	Positive	Equivocal	Negative	Equivocal	Positive	Positive	Positive	15-Mar	15-Mar		
28	Negative	Equivocal	Negative	Negative	Negative	Positive	Positive	Positive	13-Mar	15-Mar		
35	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Positive	15-Mar	16-Mar	60	
38	Positive	Positive	Positive	Negative	Negative	Positive	Positive	Negative	15-Mar	15-Mar	90	
39	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Positive	15-Mar	15-Mar		
41	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Positive	15-Mar	15-Mar	96	
42	Positive	Positive	Equivocal	Negative	Positive	Positive	Positive	Positive	15-Mar	15-Mar	90	Insufficient sample received. Therefore, equivocal result reported for serum 3 as there was not enough cells for repeat testing.
45	Positive	Positive	Negative	Negative	Negative	Positive	Positive	Positive	15-Mar	15-Mar		
48	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	15-Mar	15-Mar		Half volume of blood sent - <10ml EDTA. Lower than ideal cell counts as a result
51	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	15-Mar	16-Mar		
112	NT	NT	NT	NT	NT	NT	NT	NT	17-Mar	17-Mar	0	
115	NT	Positive	Negative	Negative	NT	Positive	Positive	Negative	15-Mar	17-Mar		Technical failure (serum 1)
117	NT	NT	NT	NT	NT	NT	NT	NT	16-Mar	16-Mar		Very poor viability and quality of cells
118	Negative	Positive	Negative	Negative	NT	NT	NT	NT	15-Mar	15-Mar	90	
119	NT	NT	NT	NT	NT	NT	NT	NT	15-Mar	15-Mar	50	2B03 not tested due to poor cell suspension and >90% apoptotic cells
120	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	15-Mar	15-Mar	100	
122	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	15-Mar	16-Mar	50	Cells were clumped. Insufficient B cells
126	Positive	Positive	Negative	Negative	Negative	Positive	Positive	Negative	15-Mar	15-Mar	>90	
130	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	15-Mar	15-Mar	96	
138	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	15-Mar	15-Mar	90	
139	Positive	Positive	Positive	Negative	Negative	Positive	Positive	Negative	17-Mar	17-Mar		
143	NT	NT	NT	NT	Negative	Positive	Positive	Negative	16-Mar	17-Mar	69.4	Not tested due to poor cell viability and low cell count
144	Negative	Positive	Positive	Negative	Negative	Positive	Positive	Negative	15-Mar	16-Mar		Second time in a row that cells were clumped on arrival.
145	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	22-Mar	22-Mar	92	
147	Equivocal	Positive	Negative	Negative	Negative	Positive	Positive	Positive	16-Mar	16-Mar	100	Serum S1 2B03 2017 shows a very weak positive reaction on T cells but being negative on B cells, the result is given as equivocal (no duplicate tests performed, see email)
149	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	16-Mar	16-Mar	81	
154	Equivocal	Positive	Negative	Negative	Negative	Positive	Positive	Positive	16-Mar	16-Mar		Clot present in sample
157	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	16-Mar	16-Mar	80	Cell sample in poor conditions of viability
159	Equivocal	Positive	Equivocal	Negative	Negative	Positive	Positive	Positive	15-Mar	16-Mar	99	T cell results are equivocal as within measurement of uncertainty
160	Negative	Positive	Negative	Equivocal	Negative	Positive	Positive	Positive	15-Mar	16-Mar		
167	Positive	Positive	Positive	Negative	Negative	Positive	Positive	Equivocal	15-Mar	16-Mar	93	
169	Negative	Negative	Negative	Negative	Negative	Positive	Positive	Negative	15-Mar	16-Mar	97	Cell suspension clotted. Low number of LyT and LyB
176	Negative	Positive	Negative	Negative	Negative	Positive	NT	Negative	15-Mar	16-Mar		Poor sample quality, low concentration of cells, B cells in particular
190	Negative	Positive	Positive	Negative	Negative	Positive	Positive	Negative	16-Mar	16-Mar	85	
191	Positive	Positive	Positive	Negative	Negative	Positive	Positive	Positive	16-Mar	17-Mar	80	
193	Equivocal	Positive	Equivocal	Negative	Negative	Positive	Positive	Negative	15-Mar	15-Mar		Requested number of cells according to our procedures has not been obtained because of the presence of aggregates.
194	Negative	Positive	Equivocal	Positive	Negative	Positive	Positive	Equivocal	15-Mar	16-Mar	98	
195	NT	NT	NT	NT	NT	NT	NT	NT	17-Mar	17-Mar	98	Not enough live cells for testing
201	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Positive	15-Mar	16-Mar	98	
209	Equivocal	Positive	Equivocal	Negative	Negative	Positive	Positive	Negative	14-Mar	16-Mar	90	
218	NT	NT	NT	NT	NT	NT	NT	NT				Unable to resuspend cell pellet. Attempted agitation and warming. It is unclear why trial failed.
220	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	15-Mar	16-Mar	95	
235	Negative	Positive	Negative	Negative	Negative	Positive	Positive	NT	16-Mar	16-Mar	80	
238	NT	NT	NT	NT	NT	NT	NT	NT	15-Mar	16-Mar		Cell suspension received with clusters, couldn't perform crossmatches
240	Negative	Positive	Equivocal	Negative	Negative	Positive	Positive	Negative	17-Mar	17-Mar	50	
246	NT	NT	NT	NT	NT	NT	NT	NT	15-Mar	15-Mar		Donor cell sample contained only cell clumps which is a sign of apoptosis i.e. bad material quality of the donor cell sample. Therefore, it was not possible to acquire enough cells and analyze them.
252	Negative	Negative	Negative	Negative	Negative	Positive	Positive	Negative			70	Not assessed
260	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	17-Mar	17-Mar		
262	Equivocal	Positive	Negative	Negative	Equivocal	Positive	Positive	Positive	15-Mar	15-Mar	77	Viability of cells was correct, but we have low number of cells due to cell clustering in the vial
271	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	21-Mar	21-Mar	93	Samples received 7 days later than the shipping date and were clumped
293	Negative	Positive	Negative	Negative	Negative	Positive	Negative	Negative	16-Mar	16-Mar		
297	Equivocal	Positive	Negative	Negative	Equivocal	Positive	Positive	Equivocal	15-Mar	16-Mar		S1 'Equivocal' for T and B cells because the T cell X-c is weakly positive and the B cell XM is weakly negative. S4 'Equivocal' on B cells because it is weakly positive but with poor B cell concentration.
341	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	15-Mar	16-Mar	98	
351	Negative	Positive	Negative	Negative	Negative	Positive	Positive	Negative	18-Mar	18-Mar	99.9	

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Lab No.	Serum 1 cytometer reading	Serum 2 cytometer reading	Serum 3 cytometer reading	Serum 4 cytometer reading	Negative control (local)	Positive control (local)	Strong positive (local)	Weak positive (local)	Positive result value	Cytometer reading units
9	2.5	5.22	0.7	0.47	2.34	42.4			>1.5 x Trimmed mean	Median x (log)
11	184212	921539	196185	160174	141, 144, 164, 174	801, 2739			186.9 1.2RMF	Median log channel
14	661.5	1495	610	486	443.5	17783.5			1.5 x mean NEG	Median log channel
15	1.6	6.1	1.52	0.92	213	23740			RMF >1.5	Median
19	0.374	1.757	0.278	0.235		21.218			1.48 x average negative control	Log Median X
20	0.782	4.507	0.643	0.468	0.433	2.595			40 Linear Channel Shifts	Linear Channel Shift
24	216	841	202	162	146	4303, 625			1.5 x negative	MFCN
25	117	477	91	62	76	5940			RMF >1.3	Median log channel
28	73	145	90	64	51	821			≥1.6-3 Equivocal, ≥3.1 Positive	MFI
35	2.04	6.66	2.03	1.46	1.69	21.86			mean of negative control +3SD	Median log channel
38	45.5	231	154.4	1.3	0	493.1			33	Linear Channel Shift
39	0.31	1.01	0.26	0.23	0.24	6.43			>1.5 RMF	Median Log Fluorescence
41	312	980	247	206	237	9115			RMF >1.6	RMF
42	1.66	4.39	1.41	1	1	130.44			>1.3	RMF
45	236.5	354	211.5	182.5	190	625				Median Channel Value
48	0.67	1.8	0.68	0.59	0.6	8.57			>1.3 RMF	Median Log Channel
51	6.04	17.32	5.14	4.39	4.75	128.6				
112	NT	NT	NT	NT	NT	NT	NT	NT		Median Log Channel
115	Tech Failure	513	300	267	290	488			>40	Median Linear Channel Shifts
117	NT	NT	NT	NT	NT	NT	NT	NT		NT
118	204	329	221	178	173	796			239	Median Linear Channel
119	NT	NT	NT	NT	NT	NT	NT	NT		NT
120	1.28	3.83	1.28	1.03	72	8421.5			Ratio >1.6	Median
122	11	42	12	7	9		109	35	3SD	Geo mean linear values
126	296	472	202	173	162	742			>80 Linear Channel Shift	Median Linear Channel
130	191	300	186	180	186	343			40	Mean Channel Shift
138	0.05	0.29	0.16	0.04	0	0.86			>0.2	D Value Kolmogorov-Smirnov Statistic
139	1.48	5.71	0.97	0.43	0.57	63			>1.5 of negative mean log channel	Mean log channel
143	NT	NT	NT	NT	NT	NT	NT	NT		Mean Channel Shift
144	1.03	3.18	1.98	0.82	1516	10.05			1.7	Median - Ration
145	0.541	2.56	0.432	0.402	0.531	68.3			2SD of ratio	Median log channel
147	187	227	150	129	131	665			>40 linear channel shift	Linear channel
149	0.512	3.71	0.588	0.485	0.346	148			2SD	Median log channel
154	176	657	129	93	87	3615			Median >150% of negative control and shape of curve	MFI
157	128	177	139	128	122	623			152 (2SD)	Median Linear Channel
159	192	368	192	146	152	520			40 Linear Channel Shift	Mean Fluorescence
160	19.1	55.5	11.5	23.1	13.4	503			>28 (NEGATIVE + 2SD)	Mean linear channel
167	296	1797	254	197	168	23828			2SD	Median log channel
169	292	337	296	288	293	567			>=50 Channel	Median Channel
176	0.27	1	0.12	-0.02	0	1.27			0.4	MFI Shift
190	84	339	177	69	68.7	943			Ratio sample/negative >1.5	Median log channel
191	44	123	74	18	0	330			>30	Delta MFI
193	15.97	101.98	12.08	5.46	5.92	388.61			Negative control x 2.4	Geo Mean Linear Channel
194	1.2	3.5	1.51	0.6	0.7	13.3			ratio >1.2	Median Channel Log
195	NT	NT	NT	NT	NT	NT	NT	NT		NT
201	0.804	2.33	1.09	0.59	0.529	52.1			S/NK ≥2.0	X-MEAN
209		1104		97	103	961			1.5 (ratio)	Geometric Mean Log Channel (Ratio)
218	NT	NT	NT	NT	NT	NT	NT	NT		NT
220	3275	24604.5	2018.5	1514.5	2093.5	261348			6000 above the mean of negative controls	Linear Acquisition, Linear Values
235	2.23	10.84	1.95	1.46	1.2	152.6			>2 of negative mean log channel	Median Log Channel
238	NT	NT	NT	NT	NT	NT	NT	NT		NT
240	73	92	88	77	71	703			1.2* NC	MFI
246	NT	NT	NT	NT	NT	NT	NT	NT		NT
252	0.86	0.97	0.71	1.32	186.01	1207.9			Median ratio ± 2SD	
260	371.31	1491.80	624.36	342.86	306.08	2439.77			2 X negative control MFI	MFI
262	604	2051	434	464	506	19675			>2 NEG	Median Channel Log
271	188	364	139	129	145	356			64	Median Log Channel
293	1.07	4.73	0.769	0.75	0.715	57			10 Channel Shift	Channel Shift
297	127	682	59	41	51	473			1.3x MFI Tneg	MFI
341	4.37	9.45	4.75	4.25	4.25	31.65			3SD	Geo mean linear values
351	270	522	266	250	242	26920			Neg control + 500	MFI

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Lab No.	Serum 1 cytometer reading	Serum 2 cytometer reading	Serum 3 cytometer reading	Serum 4 cytometer reading	Negative control (local)	Positive control (local)	Strong positive (local)	Weak positive (local)	Positive result value	Cytometer reading units
11	x.2492	19573.15730	31506.28934	14109.16430	2566, 2345, 1498, 1659	8237, 15810			2447.4 1.2RMF	Median log channel
14	2031	13091	18821	9866	1357	33803.5			1.5 x mean NEG	Median log channel
15	1.0	13.1	19.7	5.0	1081	45,247			RMF >1.5	Median
19	4.159	23.016	24.511	17.966		50.771			1.88 x average negative control	Log Median X
20	4.103	50.17	61.87	14.43	1.6675	5.935			40 Linear Channel Shift	Linear Channel Shift
24	381	9916	10067	3350	217	10633, 1051			2 x negative	MFCN
25	583	6465	9054	1084	488	14341			RMF >1.3	Median log channel
28	58	849	698	557	48	750			≥2.0-3.5 Equivocal, ≥3.6 Positive	MFI
38	-37.8	220.5	240.2	-9.4	0	289.3			40	Linear Channel Shift
39	0.96	13.87	15.33	9.83	0.64	16.70			>2.0 RMF	Median Log Fluorescence
41	502	7073	6028	1396	530	13525			RMF >1.6	RMF
42	1.62	19.62	19.55	4.62	1	97.15			>1.5	RMF
48	1.49	16.71	21.41	5.43	2.1	15.04			>1.3 RMF	Median Log Channel
51	7.23	100.01	106.04	6.18	6.34	237				
112	NT	NT	NT	NT	NT	NT			NT	Median Log Channel
115	Technical failure	788	813	512	443	741			>60	Median Linear Channel Shifts
117	NT	NT	NT	NT	NT	NT			NT	NT
118	153	212	282	152	154	512			179.25	Median Linear Channel
119	NT	NT	NT	NT	NT	NT			NT	NT
120	0.97	8.05	20.26	0.89	357	13004			ratio >2	Median
122	57	707	749.5	161.5	79		287	124	3SD	Geo mean linear values
126	255	749	779	192	180	822			>120 linear channel shift	Median Linear Channel
130	222	351	505	272	244	538			80	Mean Channel Shift
138										
139	4.17	95.3	70.8	2.88	4.04	243			>2 of negative mean log channel	Mean log channel
143	NT	NT	NT	NT	NT	NT			NT	Mean Channel Shift
144	0.96	11.32	18.94	2.00	4413	5.39			2.50	Median - Ration
145	2.71	26.1	13.02	3.07	2.3	15.2			2SD of ratio	Median log channel
147	279	648	643	336	256	665			>60 linear channel shift	Linear channel
149	2.66	58.6	97	2.97	2.59	179			2SD	Median log channel
154	501	11561	26028	1432	461	14975			Median >250% of control and shape of curve	MFI
157	109	337	402	110	140	460			182 (2SD)	Median Linear Channel
159	288	621	599	418	240	626			100 linear channel shift	Mean Fluorescence
160	52	1505	1156	276	40.6	450			>91 (NEGATIVE +2SD)	Mean linear channel
167	1310	13365	20850	4196	2164	30811			2SD	Median log channel
169	304	497	614	331	344	571			≥=80	Median Channel
176	0.09	1.44	-0.07	-0.07	0	1.07			0.22	MFI Shift
190	170	771	700	169	150	762			Ratio sample/negative >2.5	Median log channel
191	89	285	397	106	0	331			>100	Delta MFI
193	47.51	1169.71	1503.54	53.83	37.74	708.23			Negative control mean x 1.9	Geo Mean Linear Channel
194	3.8	34	60	1.7	45	45			ratio >4	Median Channel Log
195	NT	NT	NT	NT	NT	NT			NT	NT
201	1.95	27.1	31.9	3.16	0,814	102			S/NK ≥2.5	X-MEAN
209	641	10944	16680	534	580	2049			1.7 (ratio)	Geometric Mean Log Channel (Ratio)
218	NT	NT	NT	NT	NT	NT			NT	NT
220	2513.5	112524	212978.5	1618	3016	261693			6000 above mean of negative controls	Linear Acquisition, Linear Values
235	12.52	38.39	29.43		5.33	3105.9			>18 of neg median log channel	Median Log Channel
238	NT	NT	NT	NT	NT	NT			NT	NT
240	1036	1436	2117	1089	788	3501			1.4*NC	MFI
246	NT	NT	NT	NT	NT	NT			NT	NT
252	0.75	7.97	9.06	1.10	134.56	2090.80			Median ratio ± 2SD	
260	742.83	5849.25	5584.57	2573.63	1476.90	4941.90			2 X Negative control MFI	MFI
262	732	16433	11143	1668	635	42485			>2 NEG	Median Channel Log
271	184	573	529	148	184	616			90	Median Log Channel
293	3.35	29	2.2	2.16	2.06	85.5			20 Channel shift	Channel Shift
297	593	7217	10182	1163	463	964			1.5xMFI Tneg	MFI
341	6.09	71.68	89.82	15.72	10.04	124.5			3SD	Geo mean linear values
351	420	4411	15837	408	448	51638			Neg control + 2500	MFI