

UK NEQAS for H&I Scheme 6 - HLA Antibody Detection

SAMPLES 6 11-20/2016

DISPATCHED ON 28TH JUNE 2016

COMPLEMENT DEPENDENT CYTOTOXICITY (CDC) TECHNIQUE INFORMATION

Lab No	Technique	Cell source	Frozen trays	In-house Commercial	Panel		Complement source	Volume added per well (µl)				Incubation (mins)		Dead cell visualisation	Analysis Procedures	Software Manufacturer	%PRA considered pos	Comments
					Composition	Size		Cells	Sera	Comp	DTT	Pre-C'	Post-C'					
20	NIH	B-cells	No		Random	40	Cedarlane	1	1	5	1	60	45	EB+AO	Manual/Computerised	In-House	>6%	
28	CDC		Yes	One Lambda	Selected	30	Cedarlane		1	5		30	60	Fluoroquen	Manual			
29	NIH	T-cells	Yes	One Lambda	Selected	58 (+2)	Cedarlane		1	5		30	60	Fluoroquen	Manual			CDC is used in conjunction with the Class I ID and SA kits to determine specificities
42	NIH	PBL	Yes	In-house	Mixture	40	TCS Biosciences	1	1	5	1	60	120	Fluoroquen	Manual			>10%
101		T-cells	Yes	GenTrak	Selected	30	Commercial		2	6	2	30	60	Fluoroquen	Manual			T-cell panel dependant
114	NIH	T and B-cells	No		Random	50	Commercial	1	1	5	0.1	30	60	Fluoroquen	Computerised	One Lambda		
120	Standard CDC	T and B-cells	Yes	In-house	Selected	36	Cedarlane	1	1	6	2	25	50	Fluoroquen	Computerised	Lambda scan		
136	NIH	PBL	No		Mixture	49	In-house	1	1	5		30	60	EB+AO	Computerised	One Lambda		20%
139	NIH	PBL	Yes	GenTrak	Selected	30	Cedarlane	1	2	5		30	60	Fluoroquen	Manual			>4%
154	NIH	T-cells	Yes	Servbio	Mixture	30	Servbio		2	5		30	60	Eosin	Manual			Any positive results
159	Classical CDC	PBL	Yes	In-house	Selected	30	In-house	2	1	5	10	30	60	EB+AO	Computerised	One Lambda		>5%
186	NIH	PBL	No	In-house	Selected	34	Invitrogen	1	1	6	1	30	60	EB+AO	Manual			N/A
194	CDC	PBL	No	In-house	Selected	42	In-house	1	1	5	1	30	60	Fluoroquen	Computerised	One Lambda		>5%
206	NIH	PBL	Yes	In-house	Selected		Cedarlane	1	1	5	1	30	90	Fluoroquen	Manual			10%. CDC is always used along with Luminex
223		T-cells	No		Random		Innotrain	0.5	0.5	2		45	60	EB+AO	Manual			
227	NIH	PBL	No		Random	30	Immucor	1	1	6	5	30	60	Fluoroquen	Manual			3%
238	NIH	PBL	No		Selected	40	Invitrogen	1	1	5		30	60	Fluoroquen	Computerised	Commercial		Any % PRA >0
245	NIH	T-cells	Yes	GenTrak	Selected	30	BAG		2	5	0.1	30	60	EB+AO	Manual			3 (1/30)
268	CDC AgH	T-cells	Yes		Random	60	GenTrak		2	5	1/10	30	60	Fluoroquen	Manual			5%
284	With and without DTT	PBL	No		Selected	30	Cedarlane	1	1	5	1	45	75	EB+AO	Manual/Computerised	Lymhoe		1
294	AHG	T-cells	Yes	GenTrak	Selected	60	GenTrak		2	5		30	60	Fluoroquen	Manual			
311	NIH	PBL	Yes	GenTrak	Selected		Servbio		2	5		45	90	Fluoroquen	Manual			3%
315		PBL	Yes	Servbio	Selected	30	Servbio	1	2	5	1/20	30	60	Fluoroquen	Manual/Computerised	SeraTrack		>10%

FLOW CYTOMETRY TECHNIQUE INFORMATION

Lab No	Flow Cytometer Model	Cell		Conjugate	Manufacturer	Conjugate	Manufacturer	Software	Manufacturer	Comments
		preparations	source							
115										
153	BD FACS Canto II			FITC anti-human IgG FAB2	One Lambda			DIVA	BD	
264	BD	T and B-cells	One Lambda	Anti-Human IgG-FITC	One Lambda			DIVA	BD	
335	FACSCanto II		One Lambda							
345			One Lambda FlowPRA™							

ELISA TECHNIQUE INFORMATION

Lab No	ELISA kit(s) used	Batch No.	Manufacturer	ELISA kit(s) used	Batch No.	Manufacturer	Comments
117	LATM10	011	One Lambda				

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LUMINEX TECHNIQUE INFORMATION

Lab No	Positive control serum for Luminex testing?	Source	Every run?	If no:	Positive control serum Same serum?	If no:	Samples stored prior to testing?	Conditions?	Storage time? (hours)	Methods used?	Dilution/ Other	Criteria?	Serum treatment
9	No						Sometimes	-20	>48		Dilution	High background	
11	Yes	Pooled sensitised patient sera	Yes		No	Different pos controls used for SA beads, not used for Scheme 6	Yes	-40	>48	Adsorption beads		High background	
12	No						Sometimes	-10	24-48	Adsorption beads		High background	
14	Yes	Sensitised patient sera	Yes		No	Sensitised patient control sera for each kit chosen by Ab profile	Always	-20	>48	Heat inactivation, EDTA		Borderline by screening	
15	Yes	Pooled HSP sera	Yes		Yes		Yes	5	>48	EDTA, Adsorption beads			
19	Yes	Sensitised patient sera	Yes		Yes		Yes	RT	24-48				
20	Yes	Sensitised patient sera	Yes		Yes		Always	-80	>48	EDTA			
23	Yes	Positive patient sera diluted 1:2	Yes		Yes		Always	-35	>48				
24	Yes	Sensitised patient sera	Yes		No	For mixed and ID runs using One Lambda we have an in house pos control which is a pool of highly sensitised patients	Yes	-20	>48	N/A			
25	Yes	Sensitised patient sera	No		No		Yes	Frozen	<24	EDTA, Adsorption beads			
26	Yes	Sensitised patient sera	Yes		Yes		Sometimes	4	24-48	EDTA		Low PC NC ratio or High background	
28	Yes	Sensitised patient sera	Yes		Yes		Yes	-20	>48				
29	Yes	Manufacturer	Yes		No	Kit Dependent	Sometimes	4	>48	Adsorption beads		High background	
34	Yes	Sensitised patient sera	Yes		Yes		Sometimes	-40	24-48				
38	Yes	In house	Yes		No	Lifecodes Neg for Lifecodes kits	Frozen	-40	24-48	EDTA		Added to sera, 6% solution, 1µl EDTA in 9µl sera/wash buffer, no incubation time, not stored	
39	Yes	Sensitised patient sera	Yes		Yes		Always	-40	>48	EDTA, Adsorption beads		High background	
41	Yes	Sensitised patient sera	Yes		Yes		Yes	-40	>48	EDTA, Adsorption beads	Dilution		
42	Yes	Sensitised patient sera	Yes		Yes		Always	-40	24-48	EDTA, Adsorption beads			
45	Yes	Pooled sensitised patient sera	Yes		Yes		Always	Frozen	>48	EDTA, Adsorption beads		EDTA: Selected samples, added to sera, 6% solution, 5µl EDTA in 85µl sera/wash buffer Adsorption beads: Selected samples, Adsorb out, ratio 1:1, no incubation time, not stored	
46	Yes	Pooled sensitised patient sera	Yes		No	Kit Dependent	Sometimes	-20	>48	Heat inactivation, Adsorption beads			
51	Yes	Pooled sensitised patient sera	Yes		Yes		Always	-40	>48	EDTA, Adsorption beads		EDTA used for SA tests	
54	Yes	Pooled sensitised patient sera	Yes		Yes		Always	-20	>48	Adsorption beads		NC <500	
58	Yes	Sensitised patient sera, Manufacturer	Yes		No	Kit Dependent	Always	-20	>48		FCS 5%		
62	Yes	Manufacturer	Yes		Yes		Always	-70	>48				
100	Yes	Manufacturer	Yes		No	Pos control is assay specific	Always	-20	>48				
101	Yes	Sensitised patient sera	Yes		Yes		Always	-20	>48	Adsorption beads	DTT	High background	
112	Yes	Sensitised patient sera, Manufacturer	Yes		Yes		Always	-20	>48				
114	Yes	Sensitised patient sera	Yes		No	One for kit	Sometimes	Frozen	24-48	N/A		Selected samples, Adsorb Out, ratio 1/10	
115	Yes												
117	No						Always	-65	>48	Adsorption beads			
120	Yes	Pooled positive sera from plasma donors	Yes		No	Used only for LSM12 kits	Always	-20	24-48	EDTA, Adsorption beads	Dilution	NC baseline >500 and/or PC baseline <4000	
127	Yes	Pooled patients sera	Yes		Yes		Always	-30	>48	Adsorption beads			
128	Yes	Sensitised patient sera	Yes		Yes		Yes	-22	>48				
133	Yes	Manufacturer	Yes		Yes		Always	-20	>48	Adsorption beads		High background	
136	Yes	Sensitised patient sera	Yes		No		Always	Frozen	>48	Adsorption beads		All samples, Adsorb Out, ratio 1/10	
139	Yes	Sensitised patient sera	Yes		No	Different PC for detection and identification Class I and II	Always	-30	>48	Adsorption beads			
142	No						Sometimes	-20	24-48	Adsorption beads		High background	
143	No						Always	-20	>48				
147	Yes	Sensitised patient sera	Yes		Yes	For each series HLA Antibody detection	Yes	4	>48	Adsorption beads			
154	Yes	Sensitised patient sera	Yes		No	Screening = Pos Class I and II serum	Yes	4	>48				
159	Yes	Sensitised femal donors	Yes		Yes		Always	-25	>48				
160	No				No		Always	4-8	>48	Adsorption beads			
162	No			Not requested	No	Not requested	Always	-20	24-48	EDTA			
163	Yes	Sensitised patient sera	Yes		Yes	Sensitised patient sera for the labscreen mixed, another sensitised patient sera different for labscreen SA	Always	-30	>48	EDTA, Adsorption beads		EDTA: All samples, added to sera, 0.1M solution, 2µl EDTA in 18µl sera/wash buffer, minimum incubation 10 mins, not stored Adsorption beads: Selected samples, Adsorb Out, ratio 500	
164	Yes	Sensitised patient sera	Yes		Yes		Sometimes	5	>48				
169	No						Always	-20	>48		Dilution		
181	Yes	Sensitised patients pool	Yes		No	One for Class I and one for Class II	Always	Frozen	>48	Adsorption beads	Dilution		
186	Yes	Manufacturer	Yes		No		Always	Frozen	24-48	Adsorption beads		Selected samples, Sera Clean, ratio 4/20	
189	Yes	Manufacturer	Yes		No	Kit Dependent	Sometimes	2-8	24-48				
190	Yes	Manufacturer	Yes		No	Kit Dependent	Yes	-40	>48		Dilution		
191	Yes	Pooled sensitised patient sera	Yes		Yes		Always	-20	<24	Adsorption beads			
193	Yes	Sensitised patient sera	Yes		Yes		Always	-35	24-48	EDTA	DTT	All samples, added to sera, 0.1mol/L (0.2N), 3µl EDTA in 27µl sera/wash buffer, minimum incubation 10 mins, not stored	
194	Yes	Sensitised donor sera	Yes		Yes		Always	Frozen	>48			All samples	
195	Yes	Sensitised patient sera	Yes		No	Not the same for identification	Yes	Frozen	>48	N/A			
204	Yes	Pooled sensitised patient	Yes		Yes		Always	-20	>48	EDTA, Adsorption beads		High background	
205	Yes		Yes		Yes		Sometimes	Fridge	>48		Dilution		
206	Yes	Sensitised patient sera, Manufacturer	Yes		No	Kit Dependent	Yes	-30	<24	Adsorption beads			
209	Yes	Sensitised patient sera, NIBSC	Yes		No	Strong PC used for all Luminex, low pos for antibody detection	Always	-30	>48				
212	Yes	Kit Dependent	Yes		Yes		Always	4	24-48		Dilution	High background	
214	Yes	Manufacturer	Yes		Yes		Always	Frozen	>48		Dilution	Selected samples (high CONs), distilled water, 1/5 or 1/10	
227	Yes	Sensitised patient sera	Yes		Yes		Always	-40	>48	EDTA	Dilution		
229	Yes	Manufacturer	Yes		No	Kit Dependent	Always	-80	>48	Adsorption beads			
230	Yes	Kit Dependent	Yes		Yes		Always	-20	24-48				
231	No						No						
232	Yes	Manufacturer	Yes		Yes	Kit Dependent	Yes	-60	>48				
238	Yes	Pool of immunised patient sera	Yes		Yes		Always	2-5	<24	N/A			
239	Yes	Manufacturer	Yes		Yes		Always	4	>48				
240	Yes		Yes		Yes		Yes	-20	>48	N/A			
242	No						Always	Frozen	>48	Adsorption beads			
245	Yes	Pooled sensitised patient sera	Yes		No	Screening only	Always	-25	24-48	EDTA, Adsorption beads		High background	
252	Yes	Manufacturer	Yes		No	We use the positive control present in the kit for each test	Yes	-20	>48	N/A			
254	Yes												
259	Yes	Manufacturer	Yes		Yes		Sometimes	-24	24-48		Dilution	High background	
262	Yes	Immunized platelet donor	Yes		Yes		No		24-48				
264	Yes		Yes		Yes		Always	20	>48				
268	Yes	Sensitised patient sera, Manufacturer	Yes		Yes		Yes	-20	>48	N/A			
273	Yes	Sensitised patients	Yes		Yes		Always	Fridge	24-48	Adsorption beads		Selected samples, Adsorb Out, if bead negative control >500	
277	Yes	UK NEQAS samples tested last year	No	Once for lot control (each new lot)	Yes		Yes	≤30	24-48	EDTA, Adsorption beads		EDTA: All samples, added to sera, 6% solution, 5µl EDTA in 95µl sera/wash buffer, no minimum incubation, not stored Adsorption beads: Selected samples, Adsorb Out, ratio >1500	
284	Yes	Sensitised patient sera	Yes		Yes		Always	-30	>48	EDTA, Adsorption beads	Another frozen cycle	NC >500 = adsorb out, PC <5000 = frozen cycle	
292	Yes	Sensitised patient sera	Yes		No	Kit Dependent	Always	-30	>48	Adsorption beads		High background	
293	No						Yes	-70	>48	Heat inactivation, Adsorption beads		High MFI value	
294	Yes	Manufacturer	Yes		Yes		Always	-20	24-48	Adsorption beads			
297	Yes	Sensitised patient	Yes		No		Yes	-20	>48	Adsorption beads			
302	Yes		Yes		No		Always	-20	>48				
303	Yes	Sensitised patient	Yes		Yes		Always	-20	>48	Heat inactivation, EDTA, Adsorption beads		NC >500MFI = adsorb out, PC <5000MFI = Heat inactivation	
309	Yes	Manufacturer	Yes		No	Kit Dependent	Always	-80	>48	N/A		EDTA: All samples, added to sera, 0.1M solution, 2µl EDTA in 18µl sera/wash buffer, minimum incubation 10 mins, not stored	
311	Yes	Patient sera	Yes		Yes		Always	-80	>48	EDTA, Adsorption beads		Adsorption beads: Selected samples	
315	Yes	Sensitised patient sera	Yes		Yes		Yes	2-4	24-48	EDTA, Adsorption beads		EDTA: All samples, added to sera, 0.1M solution, 10µl EDTA in 90µl sera/wash buffer, minimum incubation 15 mins, not stored	
318	Yes	Manufacturer	Yes		Yes			2-8	>48	N/A		Adsorption beads: Selected samples, Adsorb Out, ratio 1/10	
322	Yes												
324	Yes	Sensitised patient sera	Yes		Yes		Sometimes	-80	>48	EDTA	Dilution		
351	Yes		Yes		Yes		Sometimes	2-8	24-48	N/A			

UK NEQAS for H&I Scheme 6 - HLA Antibody Detection

SAMPLES 6 01-10/2016 DISPATCHED ON 26TH JANUARY 2016

LUMINEX TECHNIQUE INFORMATION - Ctd

Lab No	Incubation times? Manufacturers instruction	Other	Wash step details Wash medium Anything added	Washes	First wash step Volume (ul)	Second wash step Volume (ul)	Buffer removed by	Software	Min bead count	Min pos control bead MFI	High background criteria	Data analysis
9	Yes		Supplied	5	200	5	200	Vacuum Manifold	Fusion	50	500	NC >500
11	Yes		Supplied	3	200	2	200	Centrifuge	Fusion	50	1000	>1000MFI
12	Yes		Supplied	5	175	5	175	Vacuum Manifold	Fusion	50		NC >500
14	Yes		Supplied	3	200	2	200	Centrifuge	Fusion	50	5000	Neg MFI >500, Pos/Neg ratio <2
15	Yes		Supplied	3	200	5	200	Vacuum Manifold	Fusion	50	3000	NC >1500
19	Yes		Supplied	5	200	5	200	Vacuum Manifold	Fusion	50	5000	1000-1500MFI
20	Yes		Supplied	2	200	2	200	Vacuum Manifold	Fusion	50	2000	NC >500MFI
23	Yes		Supplied	2	200	2	200	Lab Xpress	Fusion	50	500	>1500
24	Yes		Supplied	5	200	5	200	Vacuum Manifold	Fusion	50	3000	NC >500
25	Yes		Supplied	5	220	5	220	Vacuum Manifold	Fusion	150	>500	NC >500
26	Yes		Supplied	5	200	5	200	Vacuum Manifold	Fusion	50	5000	NC >750
28	Yes		Supplied	4	100	4	100	Vacuum Manifold	Fusion	30	500	NC >500
29	Yes		Supplied	1	100	3	250	Vacuum Manifold	MatchIT	100	≥10000	Manufacturer
34	Yes		Supplied	5	200	5	200	Vacuum Manifold	Fusion	50	>500 and >×2 NC	NC >500
38	Yes		Supplied	3	200	2	200	Centrifuge	Fusion	60	1500	500
39	Yes		Supplied	5	200	5	200	Vacuum Manifold	Fusion	50	5000	NC >500
41	Yes		Supplied	4	200	4	200	Vacuum Manifold	Fusion	50	1500	
42	Yes		Supplied	3	200	3	200	Vacuum Manifold	Fusion	50	2000	NC >1500
45	Yes		Supplied	4	200	4	200	Vacuum Manifold	Fusion and in house	50	5000	>500
48	Yes		Supplied	3	250	3	150, 250	Vacuum Manifold	MatchIT, Fusion	50	1000, 10000	Kit Dependent
51	Yes		Supplied	5	250	5	250	Vacuum Manifold	MatchIT, Fusion	50	5000	>500
54	Yes		Supplied	3	150	3	150	Centrifuge	Fusion	40	1000	MC >500
58	Yes		Supplied	5	150	5	150	Vacuum Manifold	MatchIT, Fusion	40	Kit Dependent	Kit Dependent
62	Yes		Supplied	5	200	5	200	Vacuum Manifold	Vacuum Manifold	50	500	>500
100	Yes		Supplied	3	250			Vacuum Manifold	MatchIT	100	1000MFI control serum	3 control beads above normal range
101	Yes		Supplied					Centrifuge	Fusion	100	4000	>500
112	Yes		Supplied	1	100	3	250	Vacuum Manifold	MatchIT	60	12000	Values of negative control beads are over the acceptable range (Manufacturer kit/kit). Irregular or atypical graphs or reactivity profiles in analysis software.
114	Yes		Supplied	3	160	2	160	Centrifuge	Fusion	50	PC >1500	NC >500
115	Yes		Supplied	3	200	3	200	Centrifuge	Fusion	50	500	raw data of NC 300-400
120	Yes		Supplied	3	150	2	200	Centrifuge	Fusion	50	4000	NC >500MFI
127	Yes		Supplied	3	200	2	200	Centrifuge	Fusion	50	NC <500 and PC/NC >×2	NC <2
128	Yes		Supplied	5	175	5	175	Vacuum Manifold	Fusion	50	500	PC/NC <2
133	Yes		Supplied	3	250	2	150	Vacuum Manifold	MatchIT	60	10000	>1000
136	Yes		Supplied	3	150	2	150	Centrifuge	Fusion	50	MFI >1000	
139	Yes		Supplied	5	175	5	175	Vacuum Manifold	Fusion	50		NC >500
142	Yes		Supplied	5	175	5	175	Vacuum Manifold	Fusion	50	5000	NC >1500MFI
143	Yes		Supplied	3	150	2	200	Centrifuge	Fusion	100	2000	NC >1500MFI
147	Yes		Supplied	4	250	4	250	Vacuum Manifold	Fusion	50	2000	>500
154	Yes		Supplied	5	175	5	175	Vacuum Manifold	Fusion	50	1500	NC >500MFI
159	Yes		Supplied	5	175	5	175	Vacuum Manifold	Fusion	100	1500	>500
160	Yes		Supplied	3	150	2	200	Centrifuge	Fusion	50	5000	
162	Yes		Supplied	3	200	2	200	Centrifuge	Fusion	50	1500	NC >500MFI
163	Yes		Supplied	4	175	4	175	Vacuum Manifold	Fusion	50	4000	500
164	Yes		Supplied	5	175	5	175	Vacuum Manifold	Fusion	50	>500	NC >1500
169	Yes		Supplied	2	150	1	150	Centrifuge	Fusion	50	500	NC >500
181	Yes		Supplied	5	175	5	175	Vacuum Manifold	Fusion	50	5000	High NC
186	Yes		Supplied	3	250	3	250	Vacuum Manifold	MatchIT	50	10000	NC criteria
189	Yes		Supplied	1	100	3	250	Vacuum Manifold	MatchIT			r=2 or NC <1500
190	Yes		Supplied	3	200	2	200	Centrifuge	Fusion	100	3000	NC <500
191	Yes		Supplied	3	150	3	150	Centrifuge	Fusion	100	raw data PC >1000	raw data NC >500
193	Yes		Supplied	3	200	2	200	Centrifuge	Fusion	100		
194	Yes		Supplied	5	175	5	175	Vacuum Manifold	Fusion	100		MFI ≤500
195	Yes		Supplied	4	175	4	175	Vacuum Manifold	Fusion	50	PC >5000	NC >500
204	Yes		Supplied	1	150	4	1x100, 3x250	Vacuum Manifold	MatchIT	60	PC ≥10000	>1000
205	Yes		Supplied	1	100	3	250	Vacuum Manifold	MatchIT	100	Sera >3500, PC >10000	Kit Dependent
206	Yes		Supplied	5	175	5	175	Vacuum Manifold	Fusion	50	3000	NC >500MFI
209	Yes		Supplied	1	100	2	250	Vacuum Manifold	MatchIT	60	≥10000	>1000MFI
212	Yes		Supplied	1	100	3	250	Vacuum Manifold	MatchIT	100	5000	BCM ≥300
214	Yes		Supplied	3	250	5	175	Vacuum Manifold	MatchIT, Fusion	50	1500 raw	≥500 raw
227	Yes		Supplied	5	175	5	175	Vacuum Manifold	MatchIT, Fusion	50	>10000	
229	Yes		Supplied	1	150	3	250	Vacuum Manifold	MatchIT	>300		>1000MFI
230	Yes		Supplied	1				Vacuum Manifold	MatchIT		1000	
231	Yes		Supplied	1	100	3	250	Vacuum Manifold	Luminex	500	1500	Internal control values
232	Yes		Supplied	4	250			Vacuum Manifold	MatchIT	1000	5000	NC out of range
238	Yes		Supplied	4	100	3	250	Vacuum Manifold	MatchIT	60	3500	NC out of range
239	Yes		Supplied	4	1x100			Vacuum Manifold	MatchIT	60	≥10000	>1000MFI
240	Yes		Supplied	3	250			Vacuum Manifold	MatchIT	60	10000	1000
242	Yes		Supplied	1	100	3	750	Vacuum Manifold	MatchIT	100	3132	
245	Yes		Supplied	5	175	5	175	Vacuum Manifold	Fusion	50	500	NC >1500
252	Yes		Supplied	4	first 100 (250 later)			Vacuum Manifold	MatchIT			
254	Yes		Supplied	1	150	3	250	Vacuum Manifold	MatchIT	50	Kit Dependent	Kit Dependent
259	Yes		Supplied	3	250	3	250	Vacuum Manifold	MatchIT	50	3500	Kit Dependent
262	Yes		Supplied	3	150	2	200	Centrifuge	Fusion	100	Apr-00	
264	Yes		Supplied	4	1x100			Vacuum Manifold	MatchIT	60	3500	values for the 3 negative beads outside the expected ranges (manufacturer)
268	Yes		Supplied	4	3x250			Vacuum Manifold	MatchIT			
273	Yes		Supplied	5	175	5	175	Vacuum Manifold	Fusion	50	>500	NC >500
277	Yes		Supplied	3	1st 150 2nd 200 3rd 200	2	200	Centrifuge	Fusion	100	500, PC/NC ratio ≥2.0	NC ≥1500
284	Yes		Supplied	4	175	4	175	Vacuum Manifold	Fusion	50	5000	500
292	Yes		Supplied	3	200	3	200	Centrifuge	Fusion	50	5000	NC >500
293	Yes		Supplied	3	1000	2	1000	Centrifuge	Fusion	100	PC >500 and x2 NC	NC >500
294	Yes		Supplied	3	250	4	175	Vacuum Manifold	MatchIT	60	>10000 PC and NC, >3500 sample patient	Con >range
297	Yes		Supplied	4	175			Vacuum Manifold	Fusion	50	1500	NC >500
302	Yes		Supplied	1	250	3	250	Vacuum Manifold	MatchIT	10000	5000	
303	Yes		Supplied	3	200	3	200	Centrifuge	Fusion	50	5000	NC >500MFI
309	Yes		Supplied	3	250			Vacuum Manifold	Fusion	50		NC >500MFI
311	Yes		Supplied	5	175	5	175	Vacuum Manifold	Fusion	50	4000	NC >500MFI
315	Yes		Supplied	4	175	4	175	Vacuum Manifold	Fusion	50	>500	<1500
318	Yes		Supplied	1	100	3	250	Vacuum Manifold	MatchIT	100	1000	
322	Yes		Supplied	5	175			Vacuum Manifold	Fusion	50	500	Known PC and NC MFI
351	Yes		Supplied	2				Vacuum Manifold	MatchIT	100	8000	All samples screened by LMX and then tested for single antigen Class I and II. High background values are monitored for single antigen testing only.

UK NEQAS for H&I Scheme 6 - HLA Antibody Detection

SAMPLES 6 11-20/2016 DISPATCHED ON 28TH JUNE 2016

IgG Results - Additional Information

Lab No.	Methods used to test these samples:			Have the samples been treated?	Apply to all?	If No, details	Please list any samples that gave "high background" in Luminex assay		Comments
	C = CDC, F = Flow, E = ELISA, LSA = Luminex Single Antigen, LM = Luminex Mixed/Multi	Apply to all?	If No, details				Further details		
9									
11	LSA, LM	No	All samples were tested by Luminex Mixed Screen. Samples 615, 616, 618 and 619 were tested in addition by LSA	No treatment	Yes				
12	LM	Yes		No treatment	Yes				
14	LM	Yes		No treatment	Yes				
15	LSA, LM	No	The following samples were positive with LM and confirmed by LSA: 613, 614, 615, 616, 618, 619 + 620	Heat inactivation, EDTA	No		613 + 620		Heat inactivation and EDTA used to run the following samples confirmed by LSA: 613, 614, 615, 616, 618, 619
19	LM	Yes		EDTA	Yes				
20	LM	Yes		No treatment	Yes				
23	C, LSA, LM	No	LSA performed for samples 613, 614, 616, 618, 619 + 620 only (when LM result indicated undetermined results based on LM cut-off ration)	EDTA	Yes				
24	LM	Yes		EDTA	Yes				
25	LM	Yes		No treatment	Yes				
26	LSA, LM	No	All samples were tested using LM. 616 also tested using LSA.	EDTA	Yes				
28	LM	Yes		EDTA	Yes				
29	LM	Yes		No treatment	Yes				
34	C, LSA, LM	No	611, 612, 615, 616 + 617 tested by LM (LifeCodes Lifescreen Deluxe Kit), 613, 614, 618, 619 + 620 tested by C, LSA and LMA (LifeCodes Lifescreen Deluxe kit and Class I ID kit)	Adsorb Out/SeraClean beads	No		617 + 618 had high background and were treated with SeraClean beads.	617 + 618	High background was removed with SeraClean beads
38	LSA, LM	Yes		EDTA	Yes				
39	LM	Yes		EDTA	Yes				
41	LM	Yes		No treatment	Yes				
42	LM	Yes		No treatment	Yes				
45	LM	Yes		EDTA	Yes				
48	LM	Yes		EDTA	Yes				
51	LM	Yes	If there were discrepancies between results, LSA were used to try and resolve these.	Adsorb Out/SeraClean beads	Yes				
54	LSA, LM	No	LM on all. LSA on 614, 618 + 620	EDTA	No				EDTA treatment only on samples tested on LSA (614, 618 + 620)
58	LM	Yes		No treatment	Yes				
62	LSA, LM	No	LSA Class I - 613-620	No treatment	Yes				
100	LM	Yes		No treatment	Yes				
101	LM	Yes		No treatment	Yes				
112	LSA, LM	Yes	LSA Class I and II: 613 + 620	No treatment	Yes				
114	LM	Yes		No treatment	Yes				
115	F, LM	Yes		No treatment	Yes				
117	E, LM	Yes		No treatment	Yes				
120	C, LM	Yes		EDTA	Yes				
127	LM	Yes		No treatment	Yes				
128	LM	Yes		No treatment	Yes				
133	LM	Yes		No treatment	Yes				The sample 619 is equivocal. The normalized/NBG ratio is in the "grey zone".
136	C, LM	Yes		Adsorb Out/SeraClean Beads	No	613, 617 + 618	613 (Class II), 617 (Class I) + 618 (Class II)		Sera Clean eliminated the high background
139	LM	Yes		No treatment	Yes				
142	LM	Yes		No treatment	Yes				
143	LM	Yes		No treatment	Yes				
147	LSA, LM	No	618: LSM12 - LSZA01, others LSM12 only	EDTA	No				618 serum gave a positive screening in Class I (one bead with NBG ratio >5) but "Single Antigen" result is negative with EDTA
153	F	Yes		No treatment	Yes				
154	C, LM	Yes		No treatment	Yes				
159	C, LM	Yes		No treatment	Yes				
160	LM	Yes		Adsorb Out/SeraClean Beads	Yes				
162	LM	Yes		EDTA	Yes				
163	LM	Yes		EDTA	Yes				
164	LSA	Yes		No treatment	Yes				613: grey zone (borderline to the limit of threshold detection): Negative
169	LM	Yes		No treatment	Yes				
181	LM	Yes		No treatment	Yes				
186	LSA, LM	No	613 + 615 were tested using LSA because they were, according to our rules, weakly positive for screening. 613 shows MFI >2000 with SA test, it's why we answer positive. In SA, 615 shows values below 2000, we report it as negative	No treatment	Yes				
189	C, LM	Yes		No treatment	Yes				
190	LM	Yes		No treatment	Yes				
191	LM	Yes		EDTA	Yes				
193	LM	Yes		EDTA	Yes				
194	C, LM	Yes		DTT	Yes				
195	LM	Yes		No treatment	Yes				
204	LM	Yes		EDTA	Yes				
205	LM	Yes		No treatment	Yes				
206	C, LM	Yes		No treatment	Yes				
209	LM	Yes		No treatment	Yes				
212	LM	Yes		Dilution	No		613 + 615 (1/3 dilution)		
214	LM	Yes		No treatment	Yes				
217	LM	Yes		No treatment	Yes				
223	C	Yes		No treatment	Yes				
227	C	Yes		No treatment	Yes				
229	C, LSA, LM	Yes		Dilution, EDTA	Yes				Results of HLA antibody detection were assessed according to laboratory's Luminex Mixed positive cut-off as for samples without sensitise events.
230	LM	Yes		No treatment	Yes				
231	L	Yes		No treatment	Yes				
232	LSA, LM	Yes		No treatment	Yes				
238	LSA, LM	Yes		No treatment	Yes				
239	C, LSA, LM	No	LSA made when "grey zone" is found for antibody detection	DTT (for CDC)	Yes				617: Luminex weak positive but anti A2 (SA), found negative by CDC
240	LM	Yes		No treatment	Yes				
242	LM	Yes		No treatment	Yes				
245	C, LM	Yes		EDTA	Yes				
252	LM	Yes		No treatment	Yes				
254	LM	Yes		No treatment	Yes				
259	LM	Yes		No treatment	Yes				
260	F	Yes		No treatment	Yes				
262	LM	Yes		No treatment	Yes				
264	LM	Yes		No treatment	Yes				
268	C, LM	Yes		DTT (for CDC)	Yes				CDC - AgH only on T cells
273	LM	Yes		No treatment	Yes				
277	LM	Yes		EDTA	Yes				
284	C, LM	Yes		EDTA	Yes				
292	LM	Yes		No treatment	Yes				
293	LM	Yes		No treatment	Yes				
294	C, LM	Yes		Adsorb Out/SeraClean beads	No	613, 616, 617 + 620			
297	LM	Yes		No treatment	Yes				
302	Im	Yes		No treatment	Yes				
303	LM	Yes		EDTA	Yes				
309	LM	Yes		No treatment	Yes				
311	LM	No	611-615	EDTA	No		611-615		
315	LM	Yes		EDTA	Yes				
318	LM	Yes		EDTA	Yes				
322	LM	Yes		No treatment	Yes				

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SAMPLES 6 11-15/2016

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Lab	611/2016				612/2016				613/2016				614/2016				615/2016			
	CI I	CI I	CI II	CI II	CI I	CI I	CI II	CI II	CI I	CI I	CI II	CI II	CI I	CI I	CI II	CI II	CI I	CI I	CI II	CI II
	IgM	Method	IgM	Method	IgM	Method	IgM	Method	IgM	Method	IgM	Method	IgM	Method	IgM	Method	IgM	Method	IgM	Method
38	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L	Positive	L	Negative	L	Negative	L	Negative	L
42	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Positive	C	NT	NT	Negative	C	NT	NT
54	Positive	L	Negative	L	Positive	L	Negative	L	Negative	L	Negative	L	Positive	L	Negative	L	Negative	L	Negative	L
101	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
120	Negative	C	Negative	C	Equivocal	C	Equivocal	C	Negative	C	Negative	C	Equivocal	C	Equivocal	C	Negative	C	Negative	C
139	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Positive	C	NT	NT	Negative	C	NT	NT
154	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Positive	C	NT	NT	Negative	C	NT	NT
159	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
162	Negative	L	Negative	L	Positive	L	Negative	L	Negative	L	Negative	L	Positive	L	Negative	L	Negative	L	Negative	L
194	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Positive	C	NT	NT	Negative	C	NT	NT
206	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Positive	C	NT	NT	Negative	C	NT	NT
227	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
238	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Positive	C	NT	NT	Negative	C	NT	NT
245	Positive	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
268	Negative	C	NT	NT	Positive	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
284	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Positive	C	NT	NT	Negative	C	NT	NT
294	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
315	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT

UK NEQAS for H&I Scheme 6 - HLA Antibody Detection

SAMPLES 6 16-20/2016

DISPATCHED ON 28TH JUNE 2016

Lab	616/2016				617/2016				618/2016				619/2016				620/2016			
	CI I	CI I	CI II	CI II	CI I	CI I	CI II	CI II	CI I	CI I	CI II	CI II	CI I	CI I	CI II	CI II	CI I	CI I	CI II	CI II
	IgM	Method	IgM	Method	IgM	Method	IgM	Method	IgM	Method	IgM	Method	IgM	Method	IgM	Method	IgM	Method	IgM	Method
38	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L
42	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
54	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L	Negative	L
101	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
120	Negative	C	Negative	C	Negative	C	Negative	C	Negative	C	Negative	C	Negative	C	Negative	C	Negative	C	Negative	C
139	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
154	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
159	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
162	Negative	L	Negative	L	Negative	L	Negative	L	Positive	L	Negative	L	Negative	L	Negative	L	Positive	L	Negative	L
194	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
206	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
227	Negative		NT	NT	Negative		NT	NT	Negative		NT	NT	Negative		NT	NT	Negative		NT	NT
238	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
245	Negative		NT	NT	Negative		NT	NT	Negative		NT	NT	Negative		NT	NT	Negative		NT	NT
268	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
284	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
294	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT
315	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT	Negative	C	NT	NT

UK NEQAS for H&I Scheme 6 - HLA Antibody Detection

SAMPLES 6 11-20/2016

DISPATCHED ON 28TH JUNE 2016

IgG Results - Additional Information

Lab No.	Methods used to test these samples:			Have the samples been treated?	Apply to all?	If No, details	Please list any samples that gave "high background" in Luminex assay	
	C = CDC, F = Flow, E = ELISA, LSA = Luminex Single Antigen, LM = Luminex Mixed/Multi	Apply to all?	If No, details				Luminex assay	Further details
38	LM	Yes		EDTA	Yes		618	
42	C	Yes		No treatment	Yes			
	LM	Yes		Adsorb Out/SeraClean Beads	No	618 only, remaining had no treatment		High NC value, reduced with Adsorb Out treatment
54							618	
101	C	Yes		No treatment	Yes			Positive CDC screening for IgM Ab in samples 612 + 614 for B cells, so we can't identify if it's Class I or Class II Antibodies
	C	Yes		DTT	Yes			
120								
139	C			No treatment				
154	C	Yes		DTT	No	611 + 614		
159	C	Yes		No treatment	Yes			
	LSA, LM	Yes		EDTA	Yes		618	High background removed by Adsorb Out treatment
162								
194	C	Yes		DTT	Yes			
206	C	Yes		No treatment	Yes			
227	C	Yes		DTT	Yes			
238								
245	C	Yes						
268	C			DTT	Yes			
284	C	Yes		DTT	Yes			
294	C	Yes		No treatment	Yes			
315	C	Yes		No treatment	Yes			Treatment with DTT

UK NEQAS for H&I Scheme 6 - HLA Antibody Detection

CDC % PRA OF 6 11-20/2016

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Lab no.	T-cell CDC % PRA	B-cell CDC % PRA	PBL CDC % PRA	611/2016			612/2016			613/2016			614/2016			615/2016			616/2016			617/2016			618/2016			619/2016			620/2016									
				T-cell	B-cell	PBL	T-cell	B-cell	PBL	T-cell	B-cell	PBL	T-cell	B-cell	PBL	T-cell	B-cell	PBL	T-cell	B-cell	PBL	T-cell	B-cell	PBL	T-cell	B-cell	PBL	T-cell	B-cell	PBL	T-cell	B-cell	PBL							
20			40	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	10	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0				
42				0	0	NT	0	28	NT	0	0	NT	0	14	NT	0	0	NT	0	0	NT	0	0	NT	0	0	NT	0	0	NT	0	0	NT	0	0	NT	0			
120	35	35		NT	NT	16	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	8	NT	NT	14	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	
136			49	12	NT	NT	0	NT	NT	0	NT	NT	7	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0			
154	30			NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	3.33	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	
159			30	NT	NT	18	NT	NT	18	NT	NT	0	NT	NT	18	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	
186			34	NT	NT	5	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	
194			42	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	20	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	
206				NT	NT	16	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	
227			30	NT	NT	20	NT	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0
268	60			12	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0
284			30	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	23	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	NT	NT	0	
294				10	NT	NT	10	NT	NT	10	NT	NT	10	NT	NT	10	NT	NT	10	NT	NT	10	NT	NT	10	NT	NT	10	NT	NT	10	NT	NT	10	NT	NT	10	NT	NT	10
311																																								
315																																								