

PBL/T-CELL AND B-CELL CROSSMATCHING METHOD OF SAMPLE 2A05+2A06/2018

**UK NEQAS for H&I Scheme 2A - Cytotoxic Crossmatching**

DESPATCHED ON 11TH SEPTEMBER 2018

Lab No.	Cells used			Immunobeaded cells used?		Complement Source	DTT addition		DTT Concentration	Volumes				Incubation Times				Visualisation of dead cells	Modifications to the standard NIH CDCXM method	Comments		
	PBL	T-cells	B-cells	T-cells	B-cells		Sera	Cells		DTT	Sera	Cells	Comp	DTT	B-cell	Pre Comp T-cell	B-cell				Post Comp T-cell	B-cell
9	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.1M	5	95	1	5	30	30	30	30	30	Fluoroquench				
11	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.008M	1	1	1	5	30	30	45	45	90	90	Fluoroquench			
12	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.01M	1	1	1	5	30	30	30	30	90	60	EB/AO			
15	Yes	Yes	Yes	Yes	Yes	TCS Biosciences	Yes	0.02M	1	2	1	5	30	30	60	60	60	60	Fluoroquench			
20	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.01M	1	1	1	5	30	30	30	30	60	60	EB/AO			
23	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.01M	1	1	1	5	30	30	40	40	40	40	EB/AO			
24	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.05M	4	6	1	5	30	30	45	45	90	90	Fluoroquench			
25	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.01M	9:81	1	1	5	30	30	30	30	45	45	Fluoroquench			
28	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.01M	1	1	1	5	30	30	30	30	45	45	EB/AO			
34	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.01M	1	1	1	5	30	30	30	30	90	90	EB/AO			
38	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.01M	1	1	1	5	60	60	60	60	60	60	EB/AO			
39	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.1M	0.1	1	1	5	10	10	45	45	90	90	Fluoroquench			
41	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.1M	1	1	1	5	30	30	45	45	45	45	EB/AO			
42	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	12mg/ml	1	1	1	5	60	60	60	120	60	60	Fluoroquench			
45	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.005M	1	1	1	5	30	30	60	30	90	75	EB/AO			
51	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.01M	1	1	1	5	30	30	60	60	60	60	Fluoroquench			
54	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.01M	1	1	1	5	30	30	30	30	60	60	PI/AO			
58	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.05M	1	2	1	5	45	45	60	60	90	90	Fluoroquench			
101	Yes	Yes	Yes	Yes	Yes	BAG	Yes	0.05M					45	45					Fluoroquench			
112	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	1M	0.1	0.9	1	5	30	30	60	120			Stain Fix			
114	Yes	Yes	Yes	Yes	Yes	One Lambda	Yes	1/4	1	1	1	5	30	30	35	45	75	90	Fluoroquench			
116	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.05	1-5	1	2-4	30	30	20	20	30	30	30	EB/AO	AMOS modification	DTT treatment of the sera is done in a separate tube 4µl DTT and 36µl sera. Cells extracted with Rosettesep	
117	Yes	Yes	Yes	Yes	Yes	Life Technology	Yes	1%	1	1	1	5	30	30	30	30	60	60	Fluoroquench			
119	Yes	Yes	Yes	Yes	Yes	Biorad	Yes	0.005M	1	1	1	6	30	30	30	30	60	60	Fluoroquench			
120	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.005M	1	1	1	5	30	30	45	45	90	90	Eosin			
128	Yes	Yes	Yes	Yes	Yes	One Lambda	Yes	0.01M	1	1	1	5	90	90	30	30	60	60	Fluoroquench			
133	Yes	Yes	Yes	Yes	Yes	Innotrain	Yes	0.01M	1	1	1	5	35	35	75	75	75	75	EB/AO			
136	Yes	Yes	Yes	Yes	Yes	In-house	Yes	0.05M	1	1	1	5	20	20	30	30	60	60	Fluoroquench			
139	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	1:10	1	1	2	5	45	45	30	30	60	60	Fluoroquench	Washes		
143	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	1:10	1	1	2	5	45	45	30	30	60	60	Eosin			
145	Yes	Yes	Yes	Yes	Yes	Invitrogen	Yes	0.005M	1	19	1	5	30	30	30	30	45	45	45	EB/AO		
147	Yes	Yes	Yes	Yes	Yes	In-house	Yes	0.05M	0.5	0.5	1	5	60	60	30	30	60	60	Fluoroquench			
149	Yes	Yes	Yes	Yes	Yes	Biorad	Yes	1:4	1	1	1	5	180	180	60	60	120	120	Eosin			
154	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.05M	10	90	1	5	45	45	30	30	60	60	Fluoroquench			
157	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.01M	10	90	1	5	45	45	60	60	60	60	Fluoroquench	AHG incubation during 3 minutes		
159	Yes	Yes	Yes	Yes	Yes	In-house	Yes	0.01M	10	90	1	5	45	45	60	60	60	60	Fluoroquench			
162	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.1M	0.1	1	1	5	30	30	30	30	60	60	Fluoroquench			
163	Yes	Yes	Yes	Yes	Yes	In-house	Yes	50mM	0.1	1	1	5	30	30	30	30	60	60	Fluoroquench			
169	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.005M	1	1	1	5	30	30	30	30	45	45	Fluoroquench			
181	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	5mM	3	57	1	6	30	30	30	30	60	60	Fluoroquench			
186	Yes	Yes	Yes	Yes	Yes	Innotrain	Yes	0.1mg/l	2	2	2	5	30	30	60	60	30	30	EB/AO			
189	Yes	Yes	Yes	Yes	Yes	Servbio	Yes	0.1M	1	19	1	5	30	30	30	30	30	60	Fluoroquench			
190	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.05M	0.05	0.95	1	5	30	30	30	30	60	60	Stain Fix			
193	Yes	Yes	Yes	Yes	Yes	Biorad	Yes	5%	1	1	1	6	15	15	30	30	45	45	Fluoroquench			
194	Yes	Yes	Yes	Yes	Yes	In-house	Yes	0.1M	2	1,2	1	5	15	15	30	30	60	60	Fluoroquench			
195	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	1/20	1	1	1	5	15	15	45	45	90	90	Fluoroquench			
197	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.01M	0.5	0.5	0.5	3.3	30	30	30	45	45	45	Fluoroquench			
204	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.0015M	1	2	1	5	10	10	30	30	60	60	Fluoroquench			
206	Yes	Yes	Yes	Yes	Yes	One Lambda, Ingen	Yes	1/10	10	90	1	5	30	30	60	30	60	60	PI/AO			
209	Yes	Yes	Yes	Yes	Yes	Innotrain	Yes	0.1M	5	0.5	0.5	2	45	45	60	60	60	60	EB/AO			
216	Yes	Yes	Yes	Yes	Yes	Biorad	Yes	0.1M	5	1	1	6	30	30	30	30	60	60	Fluoroquench			
223	Yes	Yes	Yes	Yes	Yes	Innotrain	Yes	1/10	5	45	1	5	30	30	30	30	60	60	Fluoroquench			
227	Yes	Yes	Yes	Yes	Yes	Innotrain	Yes	1/10	1	0.5	0.5	2.5	30	30	45	60	120	120	Fluoroquench			
229	Yes	Yes	Yes	Yes	Yes	Biorad	Yes	0.001M	1	1	1	5	30	30	30	30	60	60	Eosin			
232	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	1/20	2	38	1	5	30	30	30	45	60	90	Fluoroquench			
235	Yes	Yes	Yes	Yes	Yes	Innotrain	Yes	0.1M%	5	95	1,2	5	30	30	30	30	60	120	Fluoroquench			
238	Yes	Yes	Yes	Yes	Yes	BAG	Yes	0.1M	0.1	1	1	5	30	30	30	30	60	45	EB/AO			
239	Yes	Yes	Yes	Yes	Yes	Innotrain	Yes	0.1M	1	1	1	5	30	30	30	30	60	60	Fluoroquench			
245	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.1M	1	1	1	5	30	30	30	30	60	60	Fluoroquench			
252	Yes	Yes	Yes	Yes	Yes	Innotrain	Yes	0.1M	1	1	1	5	30	30	30	30	60	60	Fluoroquench			
260	Yes	Yes	Yes	Yes	Yes	Innotrain	Yes	1/2	1	1	1	5	30	30	30	30	60	60	Fluoroquench			
262	Yes	Yes	Yes	Yes	Yes	Life Technologies	Yes	1/10	3	27	1	5	30	30	30	30	60	90	Eosin			
268	Yes	Yes	Yes	Yes	Yes	Gentrack	Yes	0.1M/L	1	19	1	5	35	35	40	40	60	60	Fluoroquench			
284	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.015M	1	1	1	3	15	15	30	30	60	60	EB/AO			
292	Yes	Yes	Yes	Yes	Yes	One Lambda	Yes	0.05M	50	450	1	6	60	60	60	60	120	120	Fluoroquench			
293	Yes	Yes	Yes	Yes	Yes	Invitrogen	Yes	0.1Mol	1	19	1	3	15	15	30	30	30	30	Eosin			
294	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.001Mol/ml	1	1	1	6	30	30	30	30	60	60	Fluoroquench			
297	Yes	Yes	Yes	Yes	Yes	Servbio	Yes	0.1M	1	3	57	1	5	30	30	30	30	60	60	Fluoroquench		
311	Yes	Yes	Yes	Yes	Yes	Servbio	Yes	1M	4	76	1	5	15	15	30	30	50	50	Fluoroquench			
315	Yes	Yes	Yes	Yes	Yes	Servbio	Yes	0.1M	2	38	1	7	30	30	30	30	60	60	Fluoroquench			
321	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.005M	10	90	1	5	30	30	30	30	60	60	Fluoroquench			
351	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.005M	10	90	1	5	30	30	30	30	60	60	Fluoroquench			
411	Yes	Yes	Yes	Yes	Yes	Cedarlane	Yes	0.005M	10	90	1	5	30	30	30	30	60	60	Fluoroquench			

Denotes 2017 methodology/No methodology received

EB - ethidium bromide  
AO - acridine orange  
PI - propidium iodide  
Hb - haemoglobin  
CFDA - carboxy-fluorescein diacetate