

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

PBL/T-CELL AND B-CELL CROSSMATCHING METHOD OF SAMPLE 2A07+2A08/2016

DESPATCHED ON 07TH JUNE 2016

| 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 | Pre Comp T-cell | Pre Comp B-cell | Post Comp T-cell | | | | Post Comp B-cell |
| 9 | | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.1M | 5 | 95 | 1 | 5 | 30 | 30 | 30 | 30 | 30 | Fluoroquench | | |
| 11 | | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.1M | 1 | 1 | 1 | 5 | 30 | 30 | 45 | 45 | 60 | Fluoroquench | | |
| 12 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.01M | 1 | 1 | 1 | 5 | 30 | 30 | 30 | 30 | 60 | EB/AO | | |
| 14 | | Yes | Yes | Yes | Yes | Cedarlane | Yes | 10% | 0.1 | 0.9 | 1 | 5 | 10 | 10 | 45 | 45 | 75 | Fluoroquench | | |
| 15 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.02M | 1 | 2 | 1 | 5 | 30 | 30 | 60 | 60 | 60 | Fluoroquench | | |
| 20 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.01M | 1 | 1 | 1 | 5 | 30 | 30 | 30 | 30 | 60 | EB/AO | | |
| 23 | | Yes | Yes | Yes | Yes | Cedarlane | Yes | | | | | 5 | | | 40 | 40 | 40 | EB/AO | | |
| 24 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.005M | 4:36 | 1 | 1 | 5 | 30 | 30 | 45 | 45 | 90 | Fluoroquench | | |
| 25 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 1:10 | 9 | 81 | 1 | 5 | 30 | 30 | 45 | 45 | 60 | Fluoroquench | | |
| 28 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.01M | 1 | 1 | 1 | 5 | 30 | 30 | 30 | 30 | 45 | EB/AO | | |
| 34 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.01M | 1 | 1 | 1 | 5 | 30 | 30 | 30 | 30 | 90 | EB/AO | | |
| 35 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.1M | 1 | 1 | 1 | 5 | 10 | 10 | 45 | 60 | 90 | EB/AO | | |
| 38 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.01M | 1 | 1 | 1 | 5 | 60 | 60 | 60 | 60 | 60 | EB/AO | | |
| 39 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.01M | 0.1 | 1 | 1 | 5 | 10 | 10 | 45 | 45 | 90 | Fluoroquench | | |
| 41 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.1M | 1 | 1 | 1 | 5 | 30 | 30 | 45 | 45 | 45 | EB/AO | | |
| 42 | Yes | Yes | Yes | Yes | Yes | TSC Biosciences | Yes | 12mg/ml | 1 | 1 | 1 | 5 | 30 | 30 | 60 | 60 | 120 | Fluoroquench | | |
| 45 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.005M | 1 | 1 | 1 | 5 | 30 | 30 | 60 | 30 | 90 | EB/AO | | |
| 51 | | Yes | Yes | Yes | Yes | Cedarlane | Yes | | 1 | 1 | 1 | 5 | 0 | 0 | 60 | 60 | 60 | Fluoroquench | | |
| 54 | | Yes | Yes | Yes | Yes | Cedarlane | Yes | | 1 | 1 | 1 | 5 | 60 | 60 | 60 | 60 | 60 | PI/AO | | |
| 58 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | | 1 | 2 | 1 | 5 | 45 | 45 | 60 | 60 | 90 | Fluoroquench | | |
| 62 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | | | | | | | | | | | EB/AO | | |
| 101 | Yes | Yes | Yes | Yes | Yes | BAG | Yes | 0.05M | | | | | 45 | 45 | | | | Fluoroquench | | One hour incubation with cells, two hours with complement |
| 112 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 1M | 0.1 | 0.9 | 1 | 5 | 30 | | 60 | 120 | Stain Fix | | | |
| 114 | Yes | Yes | Yes | Yes | Yes | One Lambda | Yes | 0.05M | 0.1 | 1 | 1 | 5 | 30 | 30 | 30 | 60 | 60 | Fluoroquench | | |
| 116 | | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.05 | | 1.5 | 1 | 2.4 | 30 | 30 | 20 | 20 | 30 | EB/AO | | AMOS modification |
| 117 | Yes | Yes | Yes | Yes | Yes | Life Technology | Yes | | | 3, 2, 1 | 1 | 5 | | | 60 | 120 | 30 | Eosin, Fluoroquench | | DTT treatment of the sera is done in a separate tube 4µl DTT and 36µl sera. Cells extracted with Rosettesep |
| 119 | Yes | Yes | Yes | Yes | Yes | Biorad | Yes | | 1 | 1 | 1 | 5 | 30 | | 30 | 60 | 60 | Eosin | | |
| 120 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.005M | 1 | 1 | 1 | 6 | 30 | 30 | 60 | 60 | 60 | Fluoroquench | | Sera treatment with DTT prior to CDC incubation: 2µl DTT 0.1M + 38µl serum - incubation 30mn 37c |
| 128 | | Yes | Yes | Yes | Yes | One Lambda | Yes | | 1 | 1 | 1 | 5 | | | 45 | 90 | 90 | Eosin | | |
| 133 | Yes | Yes | Yes | Yes | Yes | Innotrain | Yes | 0.01M | 1 | 1 | 1 | 5 | 90 | 90 | 30 | 30 | 60 | Fluoroquench | | We resuspend the cell pellet with DTT in PBS (0.01M) and use 1 microliter of cell suspension per well |
| 136 | Yes | Yes | Yes | Yes | Yes | In-house | Yes | | 5 | 1 | 1 | 5 | | | 60 | 60 | 60 | EB/AO | | |
| 139 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.05M | 5 | 45 | 1 | 5 | 20 | | 30 | 60 | 60 | Fluoroquench | | Washes |
| 143 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 1:10 | 1 | 1 | 2 | 5 | 45 | | 30 | 60 | 60 | Eosin | | |
| 145 | Yes | Yes | Yes | Yes | Yes | Invitrogen | Yes | | | | | | | | 45 | 45 | 45 | EB/AO | | |
| 147 | Yes | Yes | Yes | Yes | Yes | In-house | Yes | 0.005M | 1 | 19 | | 5 | 30 | 30 | 30 | 60 | 60 | Fluoroquench | | |
| 149 | Yes | Yes | Yes | Yes | Yes | Commercial | Yes | | 0.5 | 0.5 | 1 | 5 | | | 60 | 60 | 60 | Eosin | | |
| 154 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | | | | | | | | 30 | 30 | 60 | Eosin | | |
| 157 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.005M | 1 | 1 | 1 | 5 | | | 60 | 60 | 120 | Fluoroquench | | |
| 159 | Yes | Yes | Yes | Yes | Yes | In-House | Yes | 0.05M | 10 | 90 | 1 | 5 | 45 | 45 | 30 | 30 | 60 | Fluoroquench | | AHG incubation during 3 minutes |
| 162 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.01M | 10 | 90 | 1 | 5 | 45 | 45 | 60 | 60 | 60 | Fluoroquench | | |
| 163 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.1M | 1 | 1 | 1 | 5 | 30 | 30 | 30 | 30 | 60 | Fluoroquench | | |
| 169 | Yes | Yes | Yes | Yes | Yes | One Lambda | Yes | 50mM | 1 | 1 | 1 | 5 | | | 35 | 45 | 75 | Fluoroquench | | |
| 181 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.005M | 1 | 1 | 1 | 5 | | | 30 | 30 | 45 | Fluoroquench | | |
| 186 | Yes | Yes | Yes | Yes | Yes | Invitrogen | Yes | 5mM | 1 | 19 | 1 | 6 | 30 | 30 | 30 | 60 | 60 | Fluoroquench | | |
| 189 | Yes | Yes | Yes | Yes | Yes | Innotrain | Yes | 0.0016gr/ml | 1 | 1 | 2 | 5 | | | 60 | 60 | 30 | EB/AO | | |
| 193 | Yes | Yes | Yes | Yes | Yes | Servbio | Yes | 0.1mg/l | 1 | 19 | 1 | 5 | 30 | 30 | 30 | 60 | 60 | Fluoroquench | | |
| 194 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.1M | 2 | 38 | | 5 | 30 | 30 | 30 | 30 | 60 | Fluoroquench | | |
| 195 | Yes | Yes | Yes | Yes | Yes | In-House | Yes | 5% | 1 | 1 | 1 | 6 | 15 | 15 | 30 | 30 | 45 | Fluoroquench | | |
| 197 | Yes | Yes | Yes | Yes | Yes | Invitrogen | Yes | 0.1M | 1, 2 | 1, 2 | 1 | 5 | 15 | 15 | 30 | 30 | 45 | Fluoroquench | | |
| 204 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | | 1/20 | 1 | 1 | 5 | 15 | 15 | 45 | 45 | 90 | Fluoroquench | | |
| 206 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.01M | 0.5 | 0.5, 1 | 0.5 | 3.3 | 30 | 30 | 30 | 45 | 45 | Fluoroquench | | |
| 209 | Yes | Yes | Yes | Yes | Yes | One Lambda, Ingen | Yes | 0.0015M | 1 | 2 | 1 | 5 | 10 | 10 | 30 | 30 | 60 | Fluoroquench | | CDC with AGH on T-lymphocytes during 2 minutes |
| 212 | Yes | Yes | Yes | Yes | Yes | Innotrain | Yes | 1/10 | 10 | 90 | 1 | 5 | | | 30 | 30 | 60 | PI/AO | | |
| 216 | Yes | Yes | Yes | Yes | Yes | Innotrain | Yes | | | | | | | | 45 | 45 | 60 | EB/AO | | |
| 227 | Yes | Yes | Yes | Yes | Yes | Immucor | Yes | 0.5M | 0.05 | 0.95 | 1 | 6 | | | 30 | 30 | 60 | Fluoroquench | | 15m incubation of serum with DTT at 37c before cell/serum deposite |
| 229 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 1/9 | 0.3 | 2.7 | 1 | 5 | | | 30 | 30 | 60 | Fluoroquench | | |
| 232 | Yes | Yes | Yes | Yes | Yes | Biotest | Yes | | | 0.5 | 0.5 | 2.5 | | | 45 | 60 | 60 | EB/AO | | |
| 235 | Yes | Yes | Yes | Yes | Yes | Biorad | Yes | 0.001M | 1 | 1 | 1 | 5 | 30 | | 30 | 60 | 60 | Eosin | | |
| 238 | Yes | Yes | Yes | Yes | Yes | Invitrogen | Yes | 1/20 | 2 | 38 | | 5 | 30 | 30 | 30 | 45 | 60 | Fluoroquench | | |
| 245 | Yes | Yes | Yes | Yes | Yes | BAG | Yes | 0.1M% | 5 | 95 | 1, 2 | 5 | 30 | 30 | 30 | 30 | 60 | EB/AO | | |
| 252 | Yes | Yes | Yes | Yes | Yes | Innotrain | Yes | | 0.1 | 1 | 1 | 5 | 30 | | 30 | 60 | 60 | Fluoroquench | | |
| 259 | Yes | Yes | Yes | Yes | Yes | Biorad | Yes | | | | | 5 | | | 30 | 30 | 60 | PI/AO | | |
| 260 | Yes | Yes | Yes | Yes | Yes | Innotrain | Yes | | 1 | 1 | 1 | 5 | 30 | 30 | 30 | 30 | 60 | Fluoroquench | | |
| 262 | Yes | Yes | Yes | Yes | Yes | Life Technologies | Yes | | 1/2 | 1 | 1 | 5 | | | 30 | 60 | 90 | Eosin | | |
| 269 | Yes | Yes | Yes | Yes | Yes | Gentrack | Yes | | 1/10 | 3 | 27 | 1 | 5 | | 30 | 30 | 60 | Fluoroquench | | XM LCT with AHG on T-cells |
| 284 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.1M/L | 1 | 19 | 1 | 5 | 35 | 35 | 40 | 40 | 60 | EB/AO | | T-cells tested with and without AHG |
| 292 | Yes | Yes | Yes | Yes | Yes | One Lambda | Yes | 0.015M | 1 | 1 | 1 | 3 | 15 | 15 | 30 | 30 | 60 | Fluoroquench | | |
| 293 | Yes | Yes | Yes | Yes | Yes | Invitrogen | Yes | | 50 | 450 | 2400 | 5 | 60 | 60 | 60 | 60 | 120 | Eosin | | |
| 294 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.1Mol | 1 | 19 | 1 | 3 | 15 | 15 | 30 | 30 | 30 | Fluoroquench | | |
| 297 | Yes | Yes | Yes | Yes | Yes | ID-Bio | Yes | 0.01M | 0.05 | 1 | 1 | 6 | 30 | 30 | 30 | 30 | 60 | EB/AO | | |
| 311 | Yes | Yes | Yes | Yes | Yes | Servbio | Yes | 0.1M | 5 | 1 | 1 | 5 | 30 | 30 | 30 | 30 | 60 | Fluoroquench | | |
| 315 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.1M | 4 | 76 | 1 | 5 | 15 | 15 | 30 | 30 | 50 | Fluoroquench | | |
| 321 | Yes | Yes | Yes | Yes | Yes | Cedarlane | Yes | 0.1M | 2 | 38 | | 7 | | | 30 | 30 | 60 | Fluoroquench | | |

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