Manual gating agrees with unsupervised clustering of CyTOF data
Top: gating strategy used to define cell populations using canonical markers. Axes indicate CyTOF ArcSinh-scaled marker intensities.
Bottom: Pearson correlation between relative cell-type abundance determined by unsupervised clustering (x-axis) and by manual gating (y-axis). Each marker represents a single PBMC sample.
Manual gating and unsupervised clustering lead to similar cell-type abundances and activation

Top: Abundances of indicated manually gated populations, normalized for each animal to pre-challenge timepoints. x-axis shows day post infection (DPI), lines denote mean ± 1 standard error of the mean (SEM).

Bottom: Median CD38 expression on manually gated populations shown in (C), medians are normalized to each NHP’s average baseline expression. x-axis shows DPI, lines denote mean ± 1 SEM.
| Cocktail | Antigen | Clone | Vendor and Catalog | RRID | Label | Conc. (µg/mL) |
|----------|---------|-------|-------------------|------|-------|---------------|
| Surface | CD38 | AT-1 | Abcam ab131420 | AB_11158787 | 89 | 8.00 |
| Surface | CD45 | DO58-12B3 | BD 555666 | AB_394433 | 115 | 5.37 |
| Surface | CD233 | BRIC 6 | International Blood Group Reference Laboratory 9439 | 113 | 4.11 |
| Surface | CD61 | VI-PL2 | Biolegend 336402 | AB_1227584 | 140 | 5.37 |
| Surface | CD66 | YTH71.3 | Thermo MA5-17003 | AB_2538475 | 158 | 2.25 |
| Surface | CD20 | 2H7 | Biolegend 302302 | AB_314250 | 173 | 5.37 |
| Surface | IgM | G20-127 | BD 555780 | AB_396115 | 174 | 2.68 |
| Surface | HLA-DR | Immu-357 | Beckman Coulter | 176 | 0.34 |
| Surface | CD1c | AD5-8E7 | Millenyi | 162 | 1.34 |
| Surface | BDCA3 | 1A4 | BD 559780 | AB_397321 | 164 | 3.58 |
| Surface | CD123 | 7G3 | BD 54527 | AB_395455 | 148 | 1.34 |
| Surface | CCR7 | 150503 | BD 561271 | AB_10561679 | FITC | 8.94 |
| Surface | CD3 | SP34-2 | BD 551916 | AB_394293 | 157 | 2.68 |
| Surface | CD4* | OKT4 | Biolegend 317404 | AB_571961 | 156 | 1.25 |
| Surface | CD8 | RPA-T8 | Biolegend 301002 | AB_314120 | 155 | 1.41 |
| Surface | CD45RA | H1100 | Biolegend 304102 | AB_314406 | 166 | 2.68 |
| Surface | CD11b | ICRF44 | Biolegend 303132 | AB_314164 | 153 | 6.71 |
| Surface | CD11c | 3.9 | Biolegend 301602 | AB_314172 | 143 | 1.34 |
| Surface | CD14 | M5E2 | Biolegend 301810 | AB_314192 | 151 | 10.73 |
| Surface | CD16 | 3G8 | Biolegend 302033 | AB_2104002 | 159 | 2.68 |
| Surface | CD33 | AC104.3E3 | Millenyi | 142 | 4.29 |
| Surface | CD56 | NCAM16.2 | BD 559043 | AB_397180 | 175 | 1.79 |
| Surface | CD7 | M-T701 | BD 555359 | AB_395762 | 141 | 5.37 |
| Surface | CD161 | HP-3G10 | Biolegend 339902 | AB_1501090 | 168 | 3.08 |
| Surface | CD27 | LT27 | Biorad MCA755GA | AB_321554 | 209 | 3.00 |
| Intracellular | STAT1 pY701 | 4a | BD 612232 | AB_399555 | 147 | 3.83 |
| Intracellular | STAT3 pY705 | 4/P-STAT3 | BD 612357 | AB_399646 | 139 | 5.82 |
| Intracellular | STAT4 pY693 | 38/P-Stat4 | BD 612738 | AB_399957 | 170 | 5.66 |
| Intracellular | STAT5 pY694 | 47/Stat5 | BD 611965 | AB_399366 | 149 | 5.37 |
| Intracellular | STAT6 pY691 | 18/P-Stat6 | BD 611567 | AB_399013 | 165 | 5.38 |
| Intracellular | Ki67 | SolA15 | Thermo 17-5698-82 | AB_2688057 | 169 | 3.36 |
| Intracellular | Erk1/2 pT202/Y204 | D13.14.4E | CST 4370S | AB_2315112 | 152 | 16.44 |
| Intracellular | MAPKAPK2 pT334 | 27B7 | CST 3007S | AB_490936 | 144 | 2.29 |
| Intracellular | CREB pS133 | 87G3 | CST 9198S | AB_2561044 | 145 | 7.24 |
| Intracellular | IkBa amino-terminal | L38A5 | CST 4814S | AB_390781 | 163 | 3.79 |
| Intracellular | TBK1/NAK pS172 | D52C2 | CST 5483S | AB_10693472 | 161 | 10.78 |
| Intracellular | S6 pS235/236 | 2F9 | CST 4858S | 150 | 13.15 |
| Intracellular | Zap70/Syk pY319/Y322 | 17A/P-ZAP70 | BD 612575 | AB_399984 | 160 | 2.14 |
| Intracellular | 4E-BP1  pT37/46 | 236B4 | CST 2855S | AB_560835 | 172 | 3.13 |
| Intracellular | PLCy2 pY759 | K86-689.37 | BD | 146 | 2.18 |
| Intracellular | P38 pT180/Y182 | 36/p38 | BD 612289 | AB_399606 | 154 | 3.56 |
| Intracellular | FITC (for CCR7) | FIT-22 | Biolegend 408302 | AB_528901 | 171 | 5.33 |
| Intracellular | FoxP3 | PCH101 | Thermo 14-4776-82 | AB_467554 | 167 | 10.55 |

Antibody panel for cell staining prior to CyTOF. Related to Key Resources Table. See also (Bjornson-Hooper et al., 2019a, 2019b).