

THE LANCET

Infectious Diseases

Supplementary appendix 2

This appendix formed part of the original submission and has been peer reviewed. We post it as supplied by the authors.

Supplement to: Agrati C, Cossarizza A, Valentina Mazzotta V, et al. Immunological signature in human cases of monkeypox infection in 2022 outbreak: an observational study. *Lancet Infect Dis* 2022; published online Nov 7. [https://doi.org/10.1016/S1473-3099\(22\)00662-4](https://doi.org/10.1016/S1473-3099(22)00662-4).

Appendix

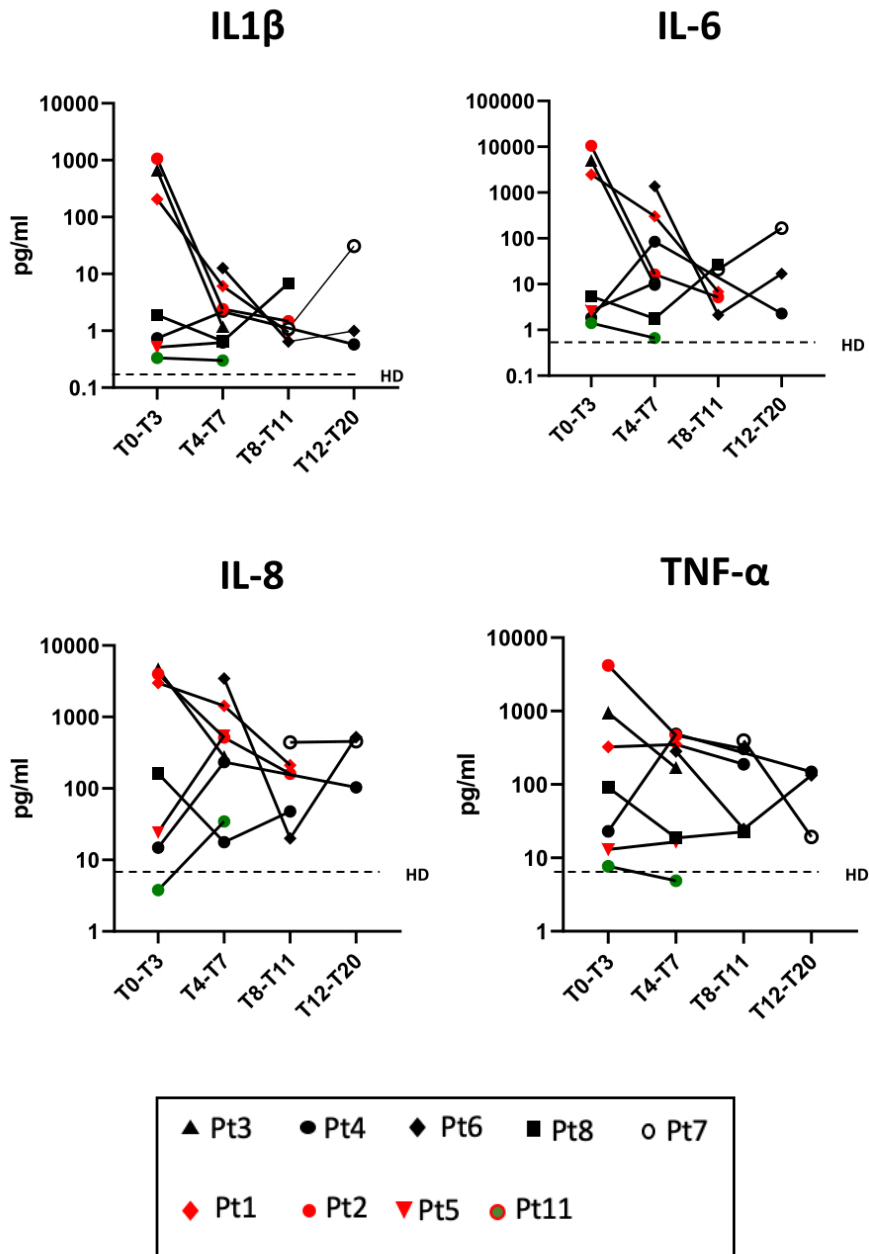
Immunological signature in human cases of monkeypox infection in 2022 outbreak

Agrati Chiara, Cossarizza Andrea, Mazzotta Valentina, Grassi Germana, Casetti Rita, De Biasi Sara, Pinnetti Carmela, Gili Simona, Mondì Annalisa, Cristofanelli Flavia, Lo Tartaro Domenico, Notari Stefania, Maffongelli Gaetano, Gagliardini Roberta, Gibellini Lara, Aguglia Camilla, Lanini Simone, D'Abramo Alessandra, Matusali Giulia, Fontana Carla, Nicastrì Emanuele, Maggi Fabrizio, Girardi Enrico, Vaia Francesco, Antinori Andrea.

Supplementary Figure 1

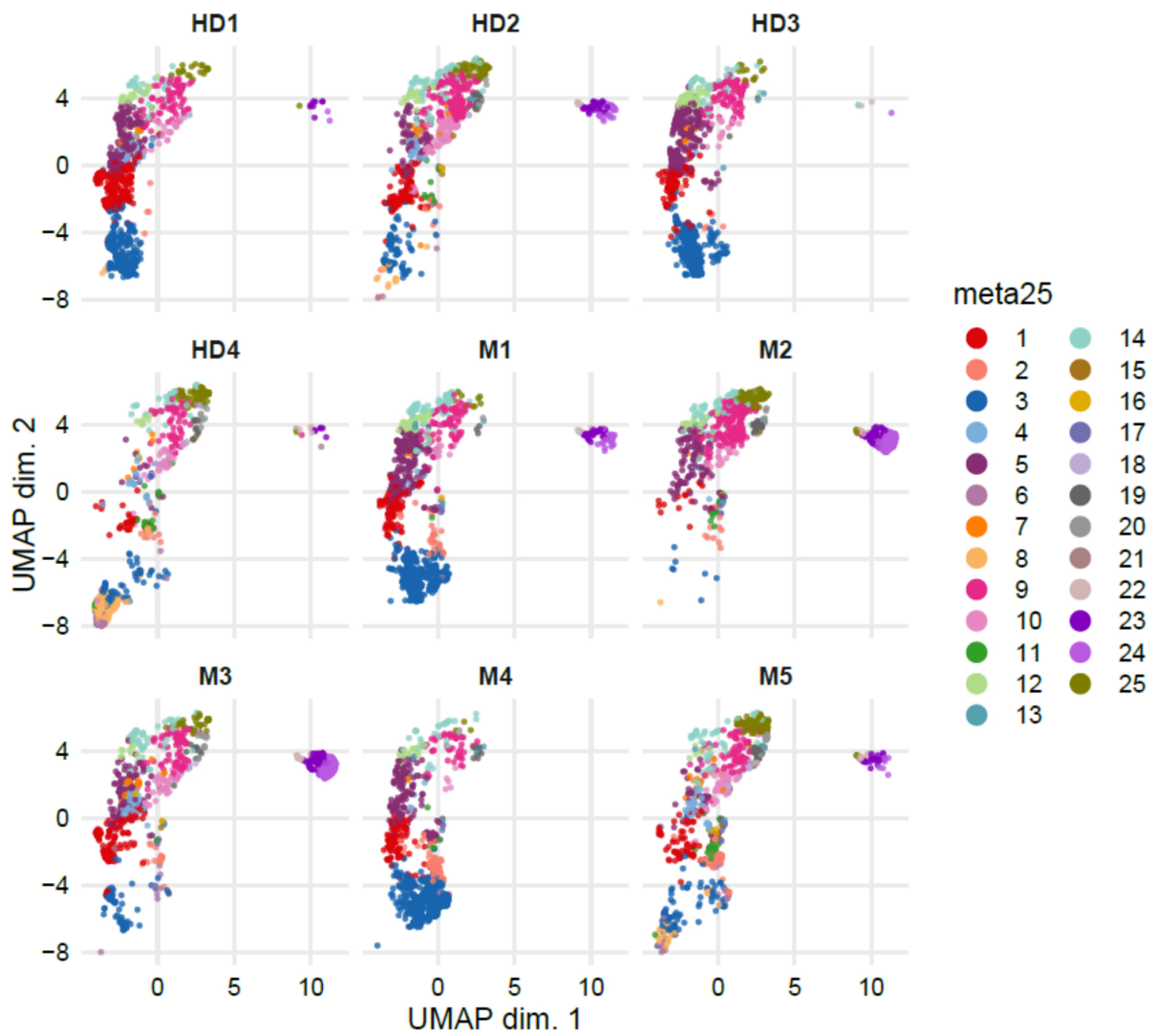
Kinetic of pro-inflammatory cytokines production during MPXV infection.

The quantification of cytokines (IL1 β , IL-6, IL-8, TNF) in plasma samples was performed by automated multiplex immunoassays on EllaTM (San José, CA, USA). The hatched line identified the median level of healthy controls (n=10). The green dot identified paucisymptomatic patients, while the red dots identified HIV-positive patients.



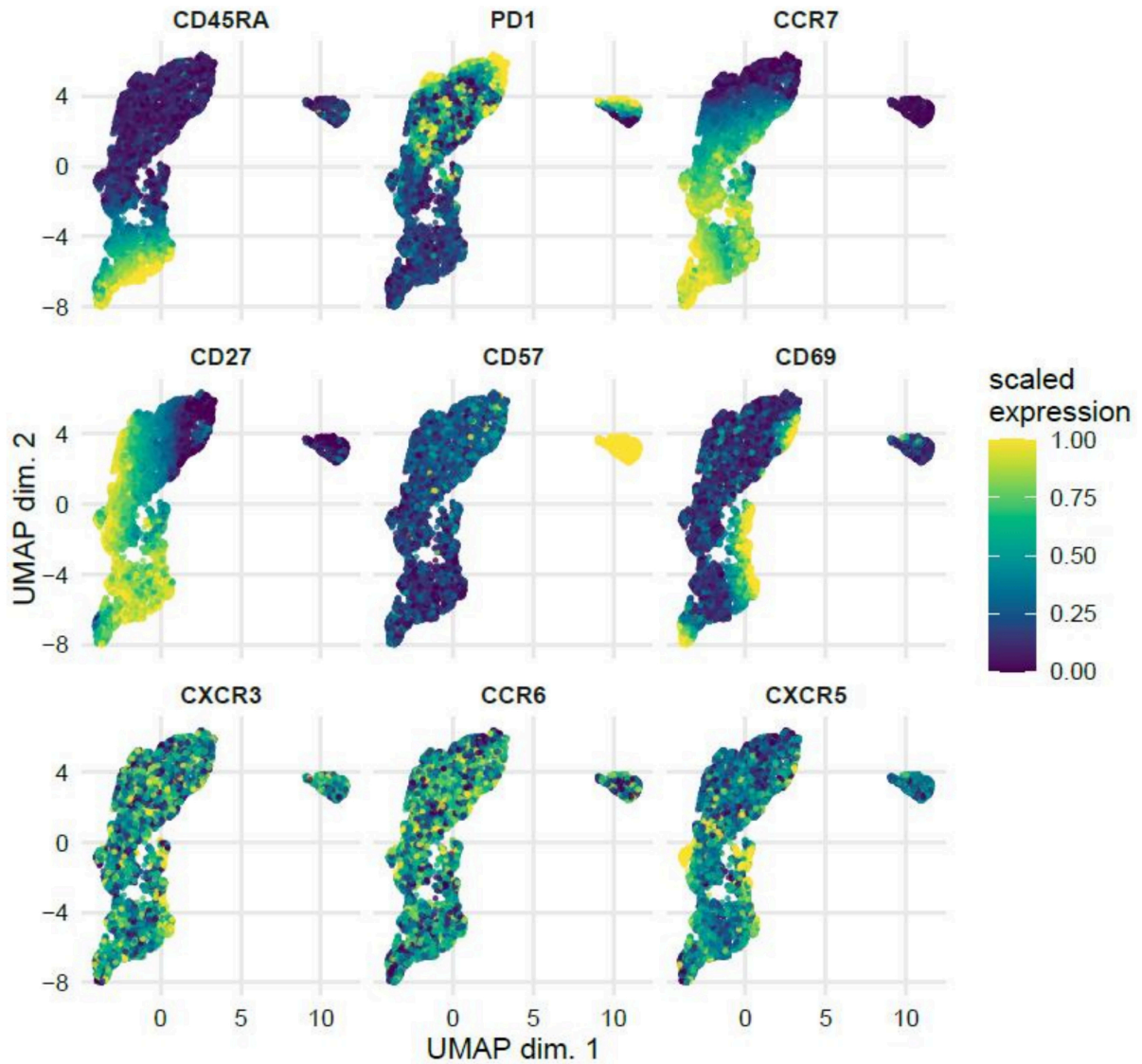
Supplementary Figures 2

Metaclusters among CD4+ T cells. Projection of UMAP graphs stratified by sample embedded with FlowSOM clusters.



Supplementary Figures 3

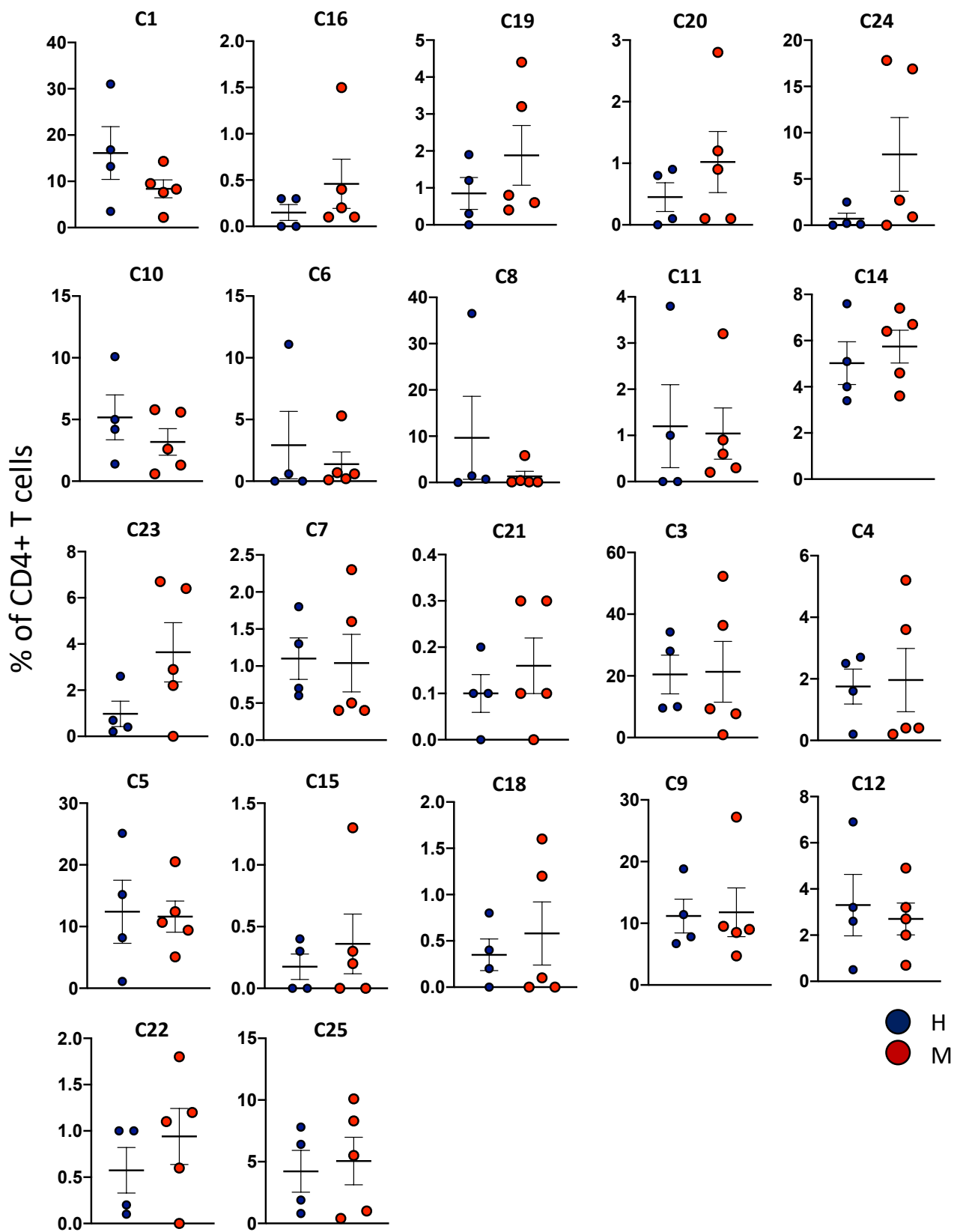
UMAP graphs coloured according to the expression of the 9 markers used for CD4+ T cell phenotyping.



Supplementary Figures 4

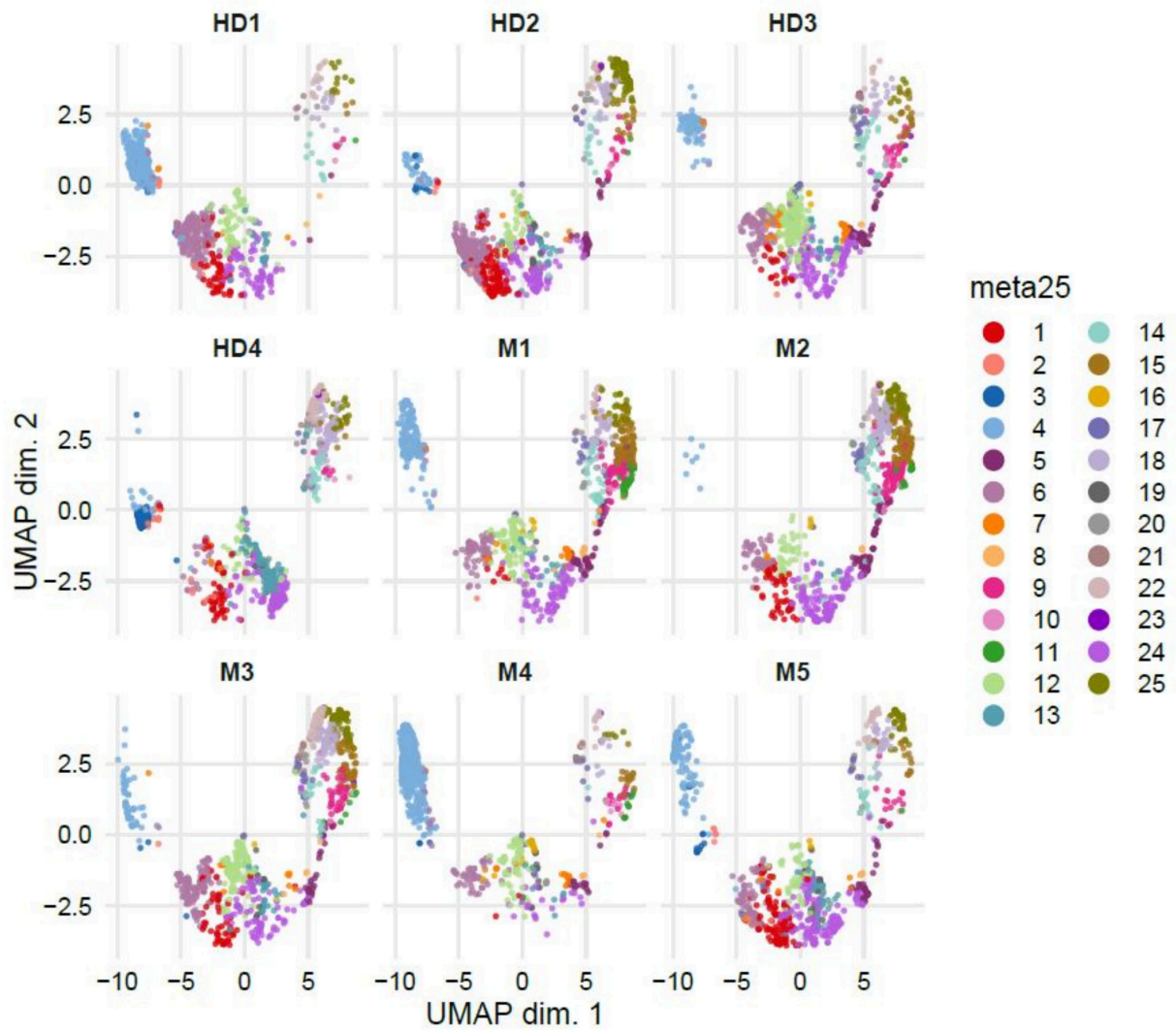
Proportion of CD4+ T-cells in different clusters.

Dotplots show the relative cell percentage of different CD4+ T-cell clusters between healthy donors (H; blue circles; n=4), and monkeypox patients (red circles; n=5). The central bar represents the mean \pm SEM. When significant, the statistical relevant adjusted p-values obtained by GLMM statistical test comparing H and M cluster percentages are reported in the figure.



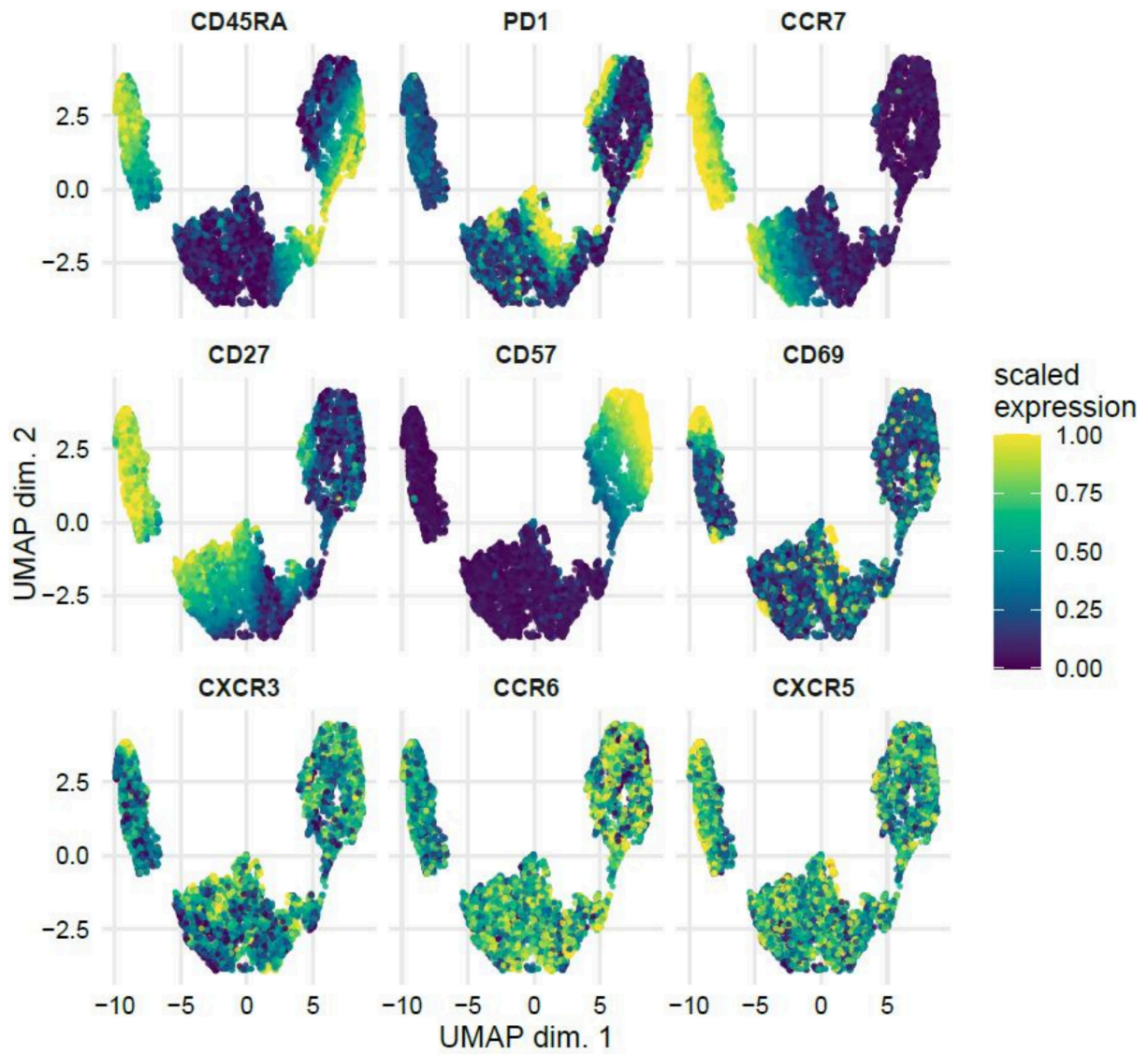
Supplementary Figures 5

Metaclusters among CD8+ T cells. Projection of UMAP graphs stratified by sample embedded with FlowSOM clusters.



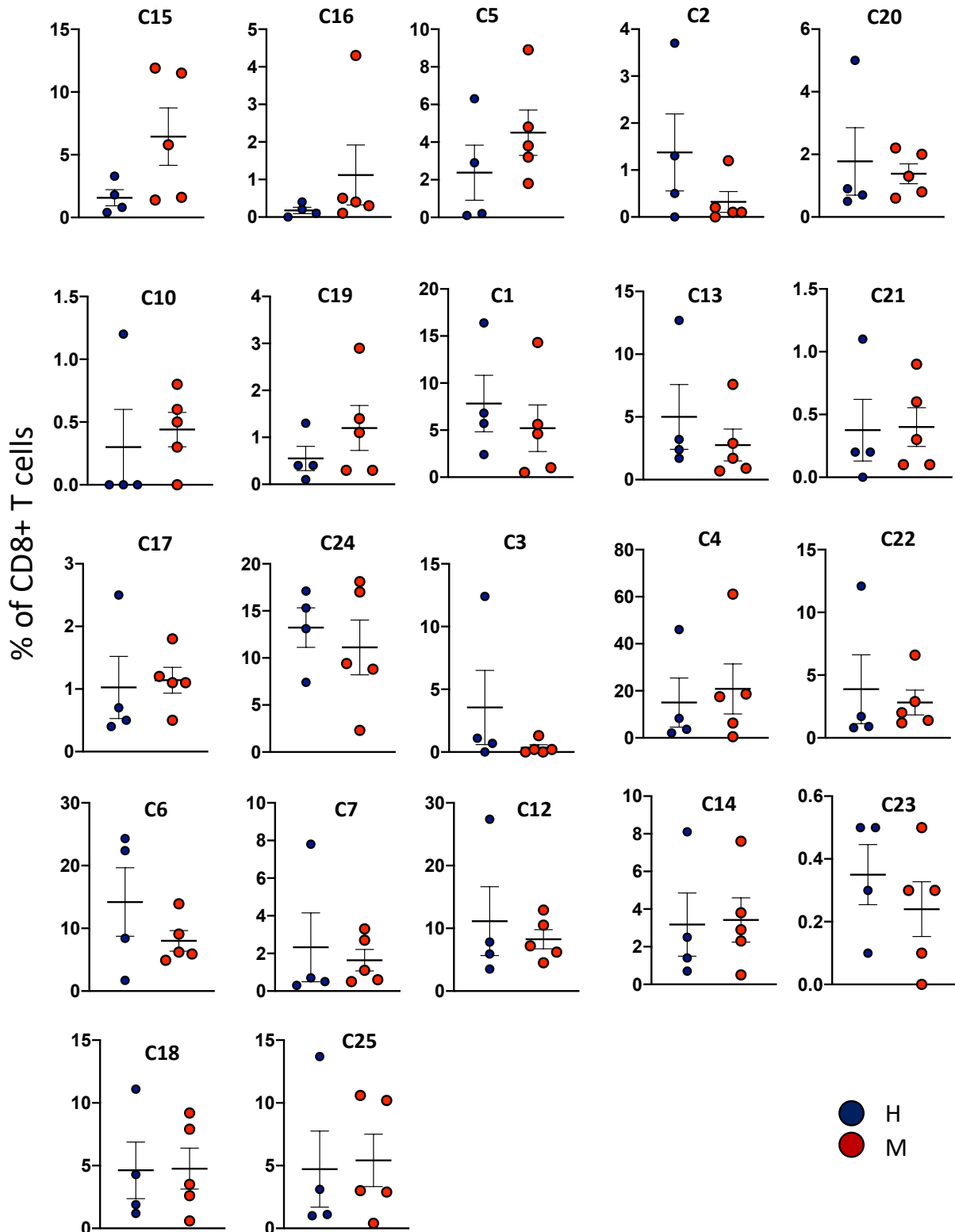
Supplementary Figures 6

UMAP graphs coloured according to the expression of 9 markers used for CD8+ T cell phenotyping.



Supplementary Figures 7

The proportion of CD8+ T-cells in different clusters. Dotplots show the relative cell percentage of different CD8+ T-cell clusters between healthy donors (H; blue circles; n=4), and monkeypox patients (red circles; n=5). The central bar represents the mean \pm SEM. When significant, the statistical relevant adjusted p-values obtained by GLMM statistical test comparing H and M cluster percentages are reported in the figure.



INMI Monkeypox Study Group

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