

Expanded View Figures

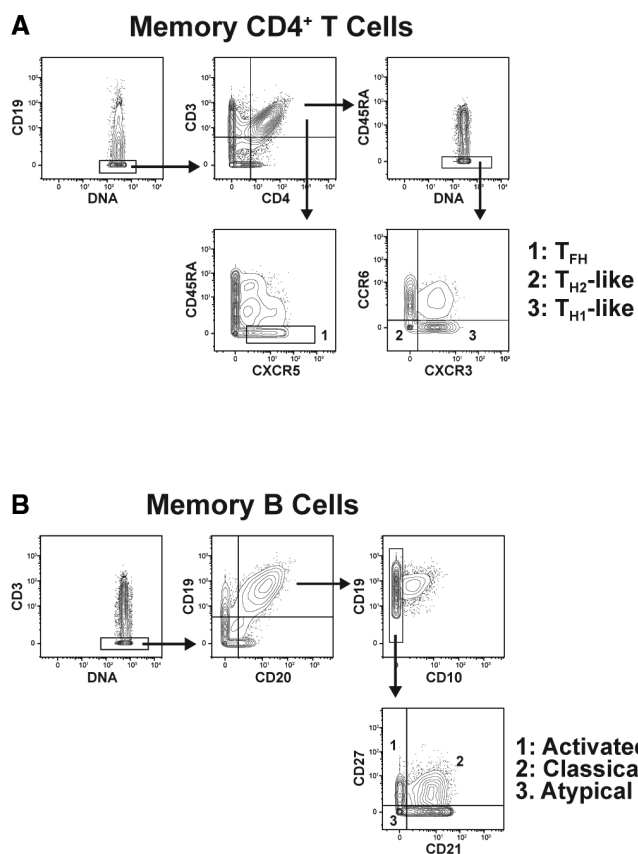


Figure EV1. Gating strategy to define major memory CD4⁺ T cell and MBC populations.

PBMCs from *P. falciparum* symptomatic ($n = 16$) and asymptomatic ($n = 24$) infected individuals as well as healthy immune controls ($n = 24$) were stained with a panel of metal-labeled antibodies and analyzed by CyTOF. Manual gating was used to select the following populations before FlowSOM clustering:

- A Individual T_{H1}-like memory CD4⁺ T cells (CD19⁻CD3⁺CD4⁺CD45RA⁻CCR6⁻CXCR3⁺), T_{H2}-like memory CD4⁺ T cells (CD19⁻CD3⁺CD4⁺CD45RA⁻CCR6⁻CXCR3⁻), circulating memory T_{FH} cells (CD19⁻CD3⁺CD4⁺CD45RA⁻CXCR5⁺) memory CD4⁺ T cells.
- B Classical (CD3⁻CD19⁺CD20⁺CD10⁻CD27⁺CD21⁺), atypical (CD3⁻CD19⁺CD20⁺CD10⁻CD27⁻CD21⁻), and activated MBCs (CD3⁻CD19⁺CD20⁺CD10⁻CD27⁺CD21⁻).

Figure EV2. Transcriptomic cohort characteristics.

P. falciparum symptomatic ($n = 30$, SM) and asymptomatic ($n = 40$, AM) infected individuals, as well as light-microscopy and PCR parasite-negative healthy immune controls ($n = 31$, HC) were recruited for the study (full bars). Subsets of 5–6 samples per group were selected for PBMC transcriptional profiling (striped bars).

- A–F Clinical parameters determined in the study included: age (A), gender (B), parasitemia (C), hemoglobin (g/dl blood) (D), hematocrit (E), and platelet count (F).
- G–M Antibody responses against the following antigens were evaluated in the study which included: *P. falciparum* parasite lysate (G), EBA-175 (H), EBA-140 (I), PfRh2 (J), PfRh4 (K), PfRh5 (L), and PfRipr (M).
- N–T Percentage of cell populations identified by CyTOF with statistically significant odds ratios are shown: CXCR3⁺ PD-1⁺ T_{H1} T_{FH} cells (N), IgM⁺ classical MBCs (O), IgM⁺ atypical MBCs (P), CD25^{low} CD27^{low} CD4⁺ T_{H2} memory cells (Q), IgD⁺IgM^{low} classical MBCs (R), Isotype switched T-bet⁺ atypical MBCs (S), and IgD⁺ IgM⁺ atypical MBCs (T).

Data information: Boxes represent the 25th to 75th percentiles, whiskers show the range (minimum to maximum), and lines represent the median. Significance was determined by the Mann–Whitney test, * $p < 0.05$, **** $p < 0.01$, ***** $p < 0.005$, ***** $p < 0.001$.

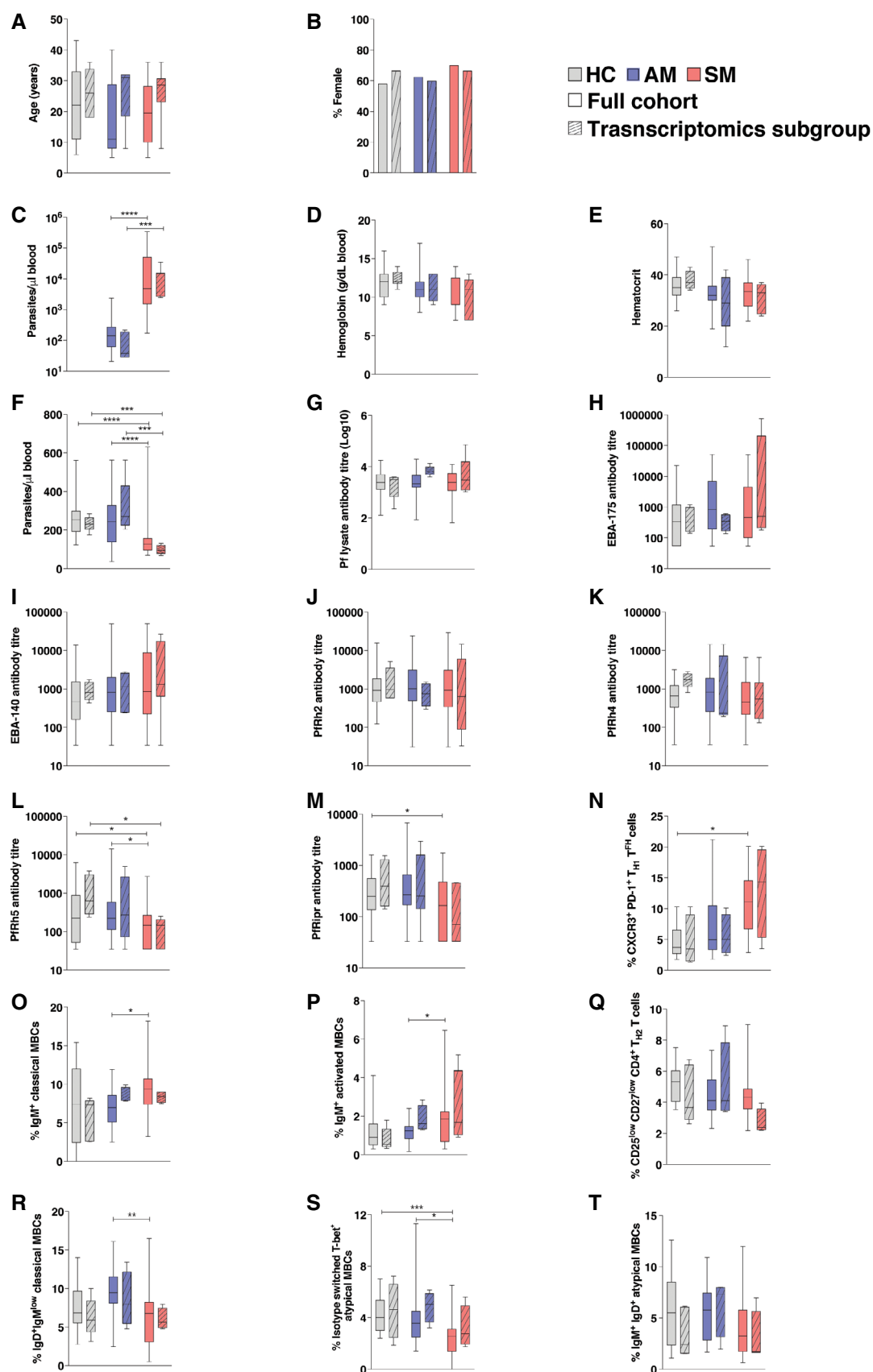


Figure EV2.

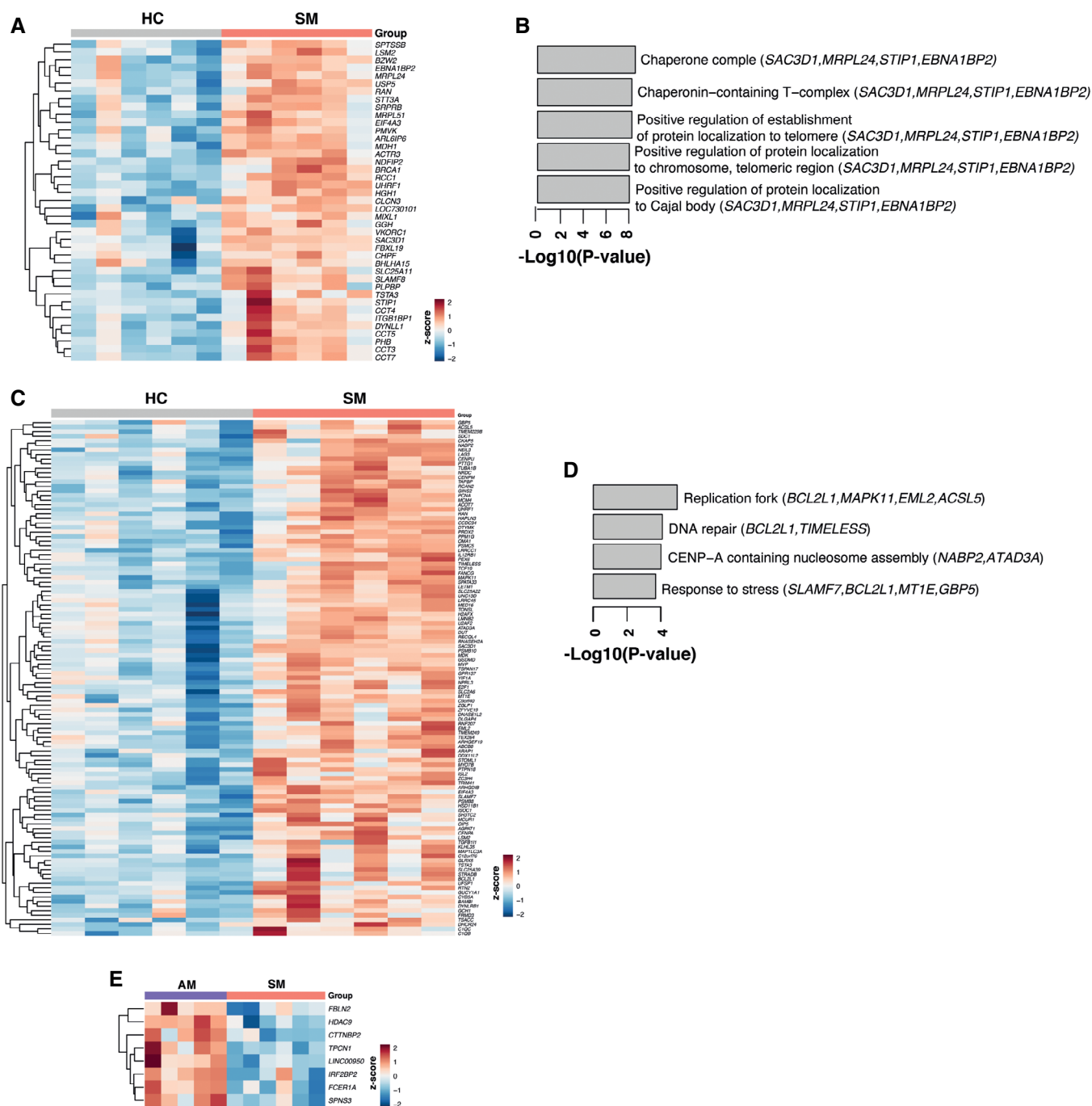


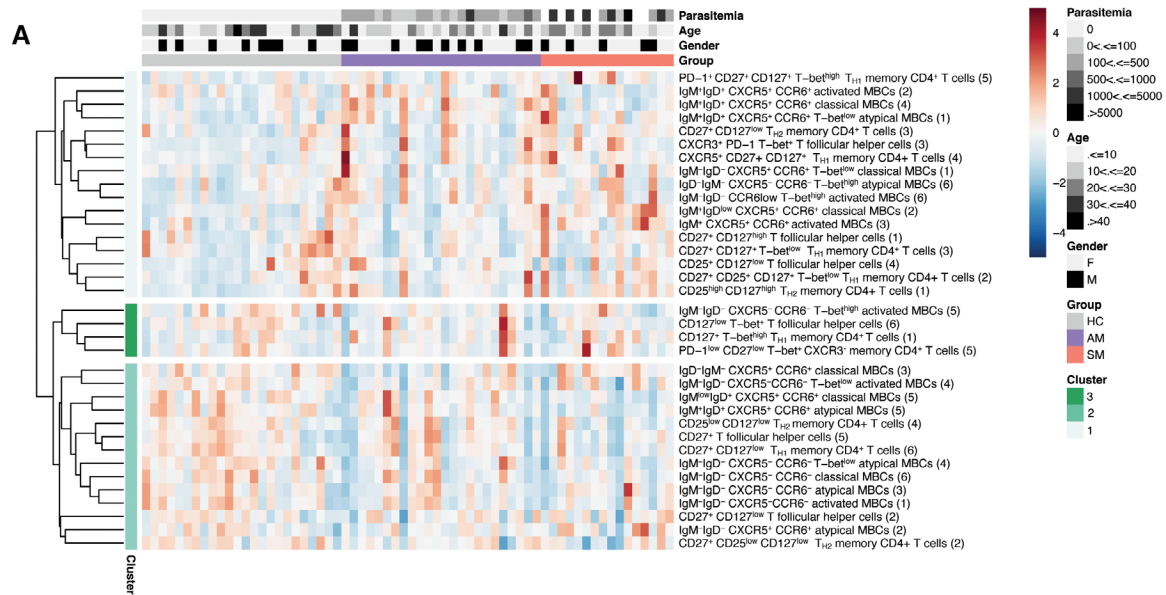
Figure EV3. Correlations between cell populations identified by CyTOF and *P. falciparum* malaria blood transcriptional signatures.

- A, B Hierarchical clustering heatmap (A) and bar plots showing enriched GO pathways (B) using genes upregulated in symptomatic individuals that were significantly correlated (Benjamini–Hochberg adjusted $P < 0.05$) with IgM⁺ activated MBCs.
- C, D Hierarchical clustering heatmap (C) and bar plots showing enriched GO pathways (D) using genes upregulated in symptomatic individuals that were significantly correlated with T-bet⁺ CD4⁺ T cells.
- E Hierarchical clustering heat map of genes upregulated in asymptomatic *P. falciparum*-infected individuals that were significantly correlated (Benjamini–Hochberg adjusted $P < 0.05$) with CD27⁺ T_{H2} CD4⁺ cells.

Figure EV4. Correlations between cell populations identified by CyTOF in response to *P. falciparum* infection.

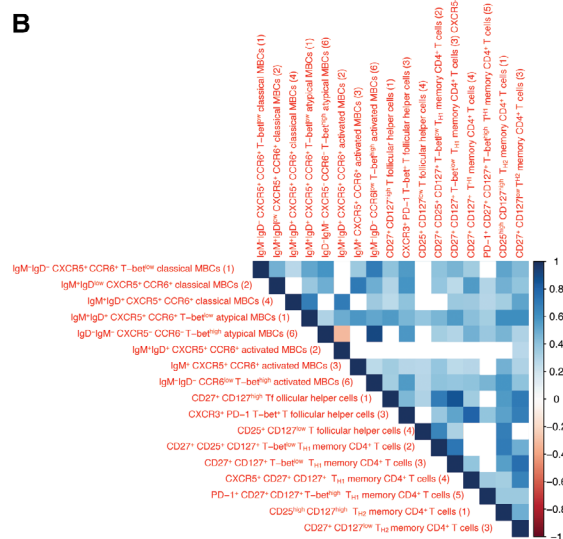
PBMCs from *P. falciparum* symptomatic ($n = 16$, SM) and asymptomatic ($n = 24$, AM) infected individuals as well as healthy immune controls ($n = 24$, HC) were stained with a panel of metal-labeled antibodies and analyzed by CyTOF.

- A Hierarchical clustering subdivides population identified by CyTOF into three main clusters, with cluster 1 featuring populations abundant in symptomatic patients and cluster 3 cells abundant in healthy immune controls.
- B–D Spearman correlation matrices were used to determine the relationship between cell populations within each separate cluster (Benjamini–Hochberg adjusted $P > 0.05$). The numbers between parentheses next to each cell population indicate the cluster number within each subset defined by FlowSOM clustering in Fig 2.



Cluster 1 – populations enhanced in SM, BH adjusted $p < 0.05$

Cluster 2 – populations enhanced in HC, BH adjusted $p < 0.05$



Cluster 3 – T-bet^{low} populations, BH adjusted $p < 0.05$

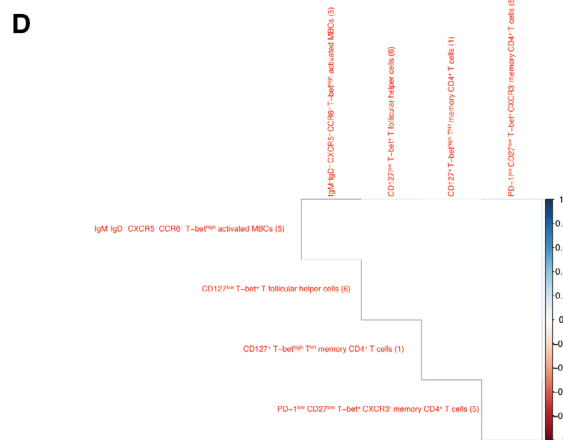
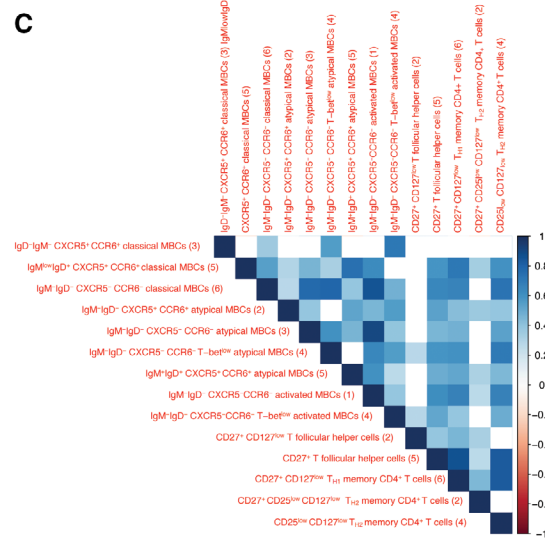


Figure EV4.