Figure S1

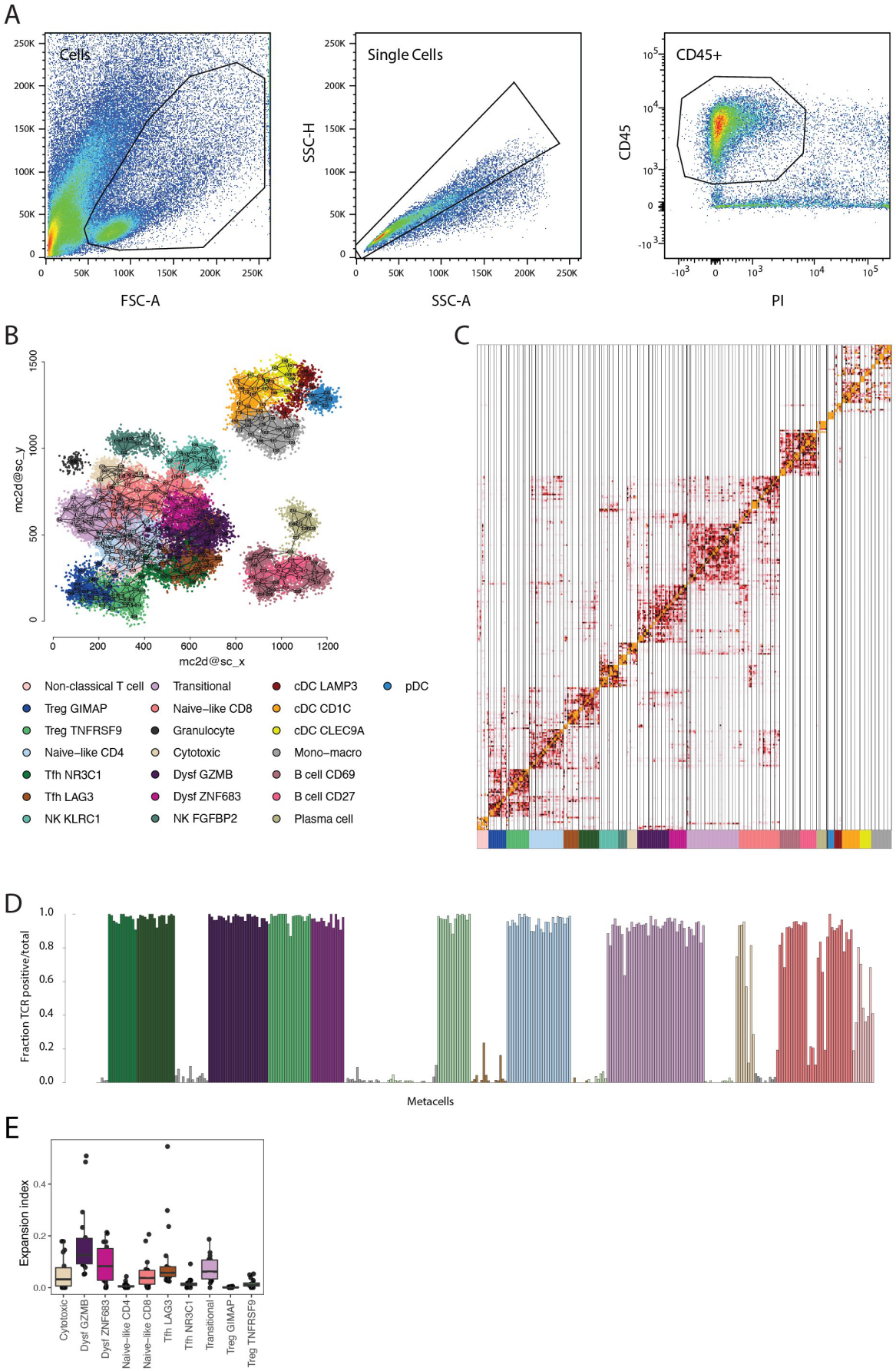
A
Cells
FSC-A
Single Cells
SSC-H
SSC-A
CD45+
CD45
PI
B
mc2d@sc_y
mc2d@sc_x
Non-classical T cell
Treg GIMAP
Treg TNFRSF9
Naive−like CD4
Tfh NR3C1
Tfh LAG3
NK KLRC1
Transitional
Naive−like CD8
Granulocyte
Cytotoxic
Dysf GZMB
Dysf ZNF683
NK FGFBP2
cDC LAMP3
cDC CD1C
cDC CLEC9A
Mono−macro
B cell CD69
B cell CD27
Plasma cell
pDC
C
D
Fraction TCR positive/total
Metacells
E
Expansion index
Cytotoxic
Dysf GZMB
Dysf ZNF683
Naive−like CD4
Naive−like CD8
Tfh LAG3
Tfh NR3C1
Transitional
Treg GIMAP
Treg TNFRSF9

Figure S1. Cell state annotations of CD45^{+} cells and clonality of T cells isolated from HNSCC biopsies

A) Gating strategy used for flow cytometry analysis and sorting of live CD45^{+} cells from pre- and on treatment biopsies of 18 HNSCC patients. B) 2D projection of all cells from all patients as in Figure 1B, metacell numbers and edges between metacells are depicted, colored by cell states. C) Matrix of metacell correlations, in which groups of related metacells cluster together. Cell state annotations are shown by the color code, corresponding to the code used in Figure 1B and S1B. D) Detection of TCR sequences per metacell, colored by cell state. Fraction of cells for which a TCR was detected is shown for each metacell, distinguishing metacells that consist of ab T cells from metacells that (partly) consist of non(-classical) T cells. E) Quantification of clonal expansion in each cell state across all patients using the STARTRAC expansion index[37]. Boxes show the median and 25th and 75th percentiles. Whiskers depict 1.5 × interquartile range (IQR), and data points represent individual patients.