Panel: WBC subsets

Panel: T-cell activation



Supplementary Figure S1. Gating strategies for flow cytometry analysis of rhesus macaque peripheral mononuclear cells. First gated on CD45+ hematopoietic cells, then separate them into lymphocytes by FSC and SSC, and monocytes by being CD14+HLA-DR+. Then lymphocytes were separated into B cells, T cells and NK cells as being CD3-CD20+, CD3+CD20-, CD3-CD8+NKG2A+, respectively. T cells were further divided into CD4+CD8- and CD4-CD8+ subsets. CD123 and CD11c were used to divide CD3-CD20-CD14-HLA-DR+ subset into pDC and mDC. Through different antibody panels and 12-color flow cytometry, the activation, regulation and functional characteristics of T cells were further analyzed. First, CD4+ and CD8+ T cells were divided into three differentiation stages including naïve (CD95dimCD28+), central memory (CD95highCD28+), and effector memory (CD95+CD28-) cells. The expression levels of CD27, CXCR3, PD-1, HLA-DR, CXCR5, ICOS, CCR6, CD38, CD25 were used to illustrate the changes of T cell chemotaxis and immune activation after viral infection. T-bet, CCR7, Ki67, CD31 and CD45RA were used as markers for T cell differentiation and function regulation. The soluble

mediators, such as Granzyme B, IL-4, IL-2, Perforin, IL-13, TNF- α , IL-17A and IFN- γ , were used to describe antiviral function and immune response of T cells.



Supplementary Figure S2. Pulmonary infiltrates in rhesus macaques can be identified by mobile digital medical X-ray photography system.



Figure S3. Heat map of changes in the number or proportion of the main leukocyte subsets in peripheral blood from young or old Chinese rhesus macaques after SARS-Cov-2 infection. The rainbow-colored squares represent the average cell number or frequency in each group. The black asterisk indicates p < 0.05 compared to d0 by Fisher's LSD test post two-way ANOVA.

Table S1 Antibodies for flow cytometry

Panel: WBC Subsets

Antibody	Fluorochrome	Clone	Source
CD123	BV421	7G3	BD Pharmigen
CD20	BV510	2H7	BD Pharmigen
CD45	BV605	D058-1283	BD Pharmigen
CD56	BV650	NCAM-16.2	BD Pharmigen
CD14	BV786	M5E2	BD Pharmigen
CD11C	FITC	3.9	BD Pharmigen
NKG2A	PE	REA110	Miltenyi Biotec
CD16	PE-CF594	3G8	BD Pharmigen
CD4	PerCP-Cy5.5	L200	BD Pharmigen
HLA-DR	APC	G46-6	BD Pharmigen
CD8	APC-R700	RPA-T8	BD Pharmigen
CD3	APC-Cy7	SP34-2	BD Pharmigen

Panel: T-cell Activation

Antibody	Fluorochrome	Clone	Source
CD27	BV421	MT271	BD Pharmigen
CD28	BV510	CD28.2	BD Pharmigen
CXCR3	BV605	G025H7	Biolegend
PD-1	BV650	EH12.2H7	Biolegend
CD95	BV786	DX2	Biolegend
HLA-DR	FITC	G46-6	BD Pharmigen
CXCR5	PE	MU5UBEE	eBioscience
ICOS	PE-Dazzle594	C398.4A	Biolegend
CCR6	PerCP-Cy5.5	G034E3	Biolegend
CD38	APC	AT-1	NIH
CD8	APC-R700	RPA-T8	Biolegend
CD3	APC-Cy7	SP34-2	Biolegend
Panel: T-cell Regulation			
Antibody	Fluorochrome	Clone	Source

CD25	BV421	M-A251	BD Pharmigen
CD28	BV510	CD28.2	BD Pharmigen
T-bet	BV605	4B10	Biolegend
CCR7	BV650	3D12	Biolegend
CD95	BV786	DX2	Biolegend
p-STAT1(Tyr701))	Purified	5D86	CST
Ki67	PE	B56	BD Pharmigen
CD31	PE-Dazzle594	WM59	Biolegend
Foxp3	PE-Cy5	PCH101	eBioscience
CD45RA	APC	5H9	BD Pharmigen
CD8	APC-R700	RPA-T8	BD Pharmigen
CD3	APC-Cy7	SP34-2	BD Pharmigen
Mouse anti rabbit IgG	FITC	Polyclone	Boster

Panel: T-cell Function

Antibody	Fluorochrome	Clone	Source
Granzyme B	Pacific Blue	GB11	Biolegend
CD28	BV510	CD28.2	BD Pharmigen
IL-4	BV604	MP4-25D2	Biolegend
IL-2	BV650	MQ1-17H12	Biolegend
CD95	BV786	DX2	Biolegend
Perforin	FITC	Pf-344	Mabtech
IL-13	PE	JES10-5A2.2	Miltenyi Biotec
TNF-α	PE-Dazzle594	MAb11	Biolegend
IL-17A	PerCP-Cy5.5	BL168	Biolegend
IFN-γ	APC	4S.B3	Biolegend
CD8	APC-R700	RPA-T8	BD Pharmigen
CD3	APC-Cy7	SP34-2	BD Pharmigen