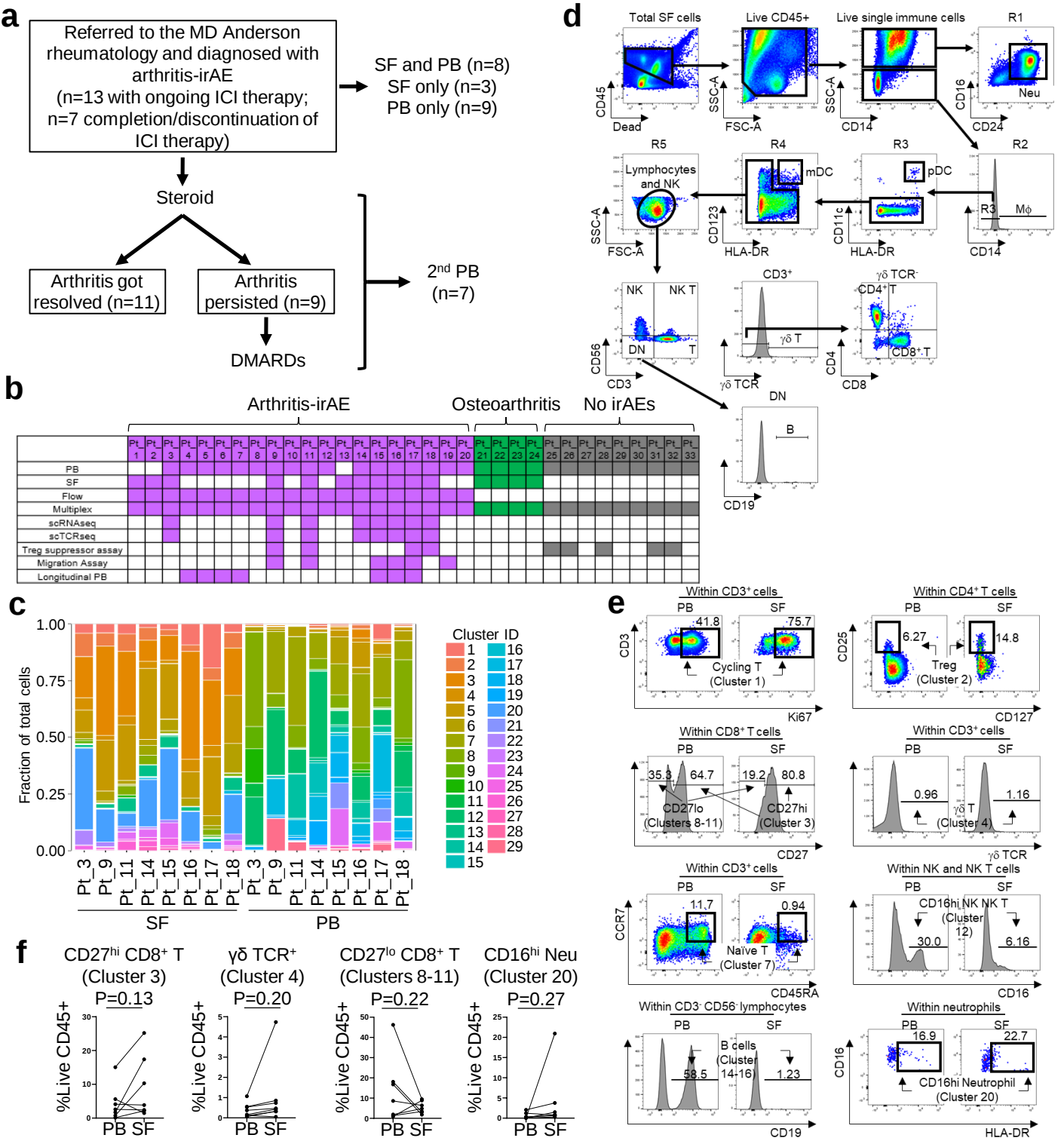


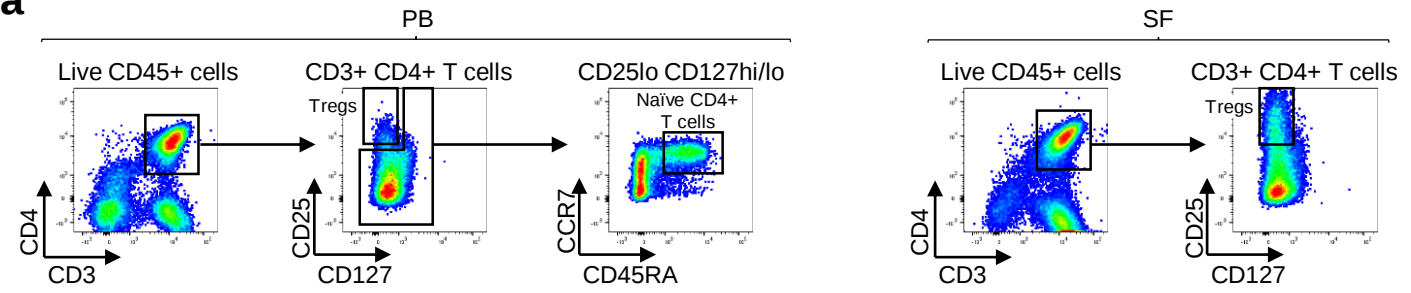
Supplementary Figure 1



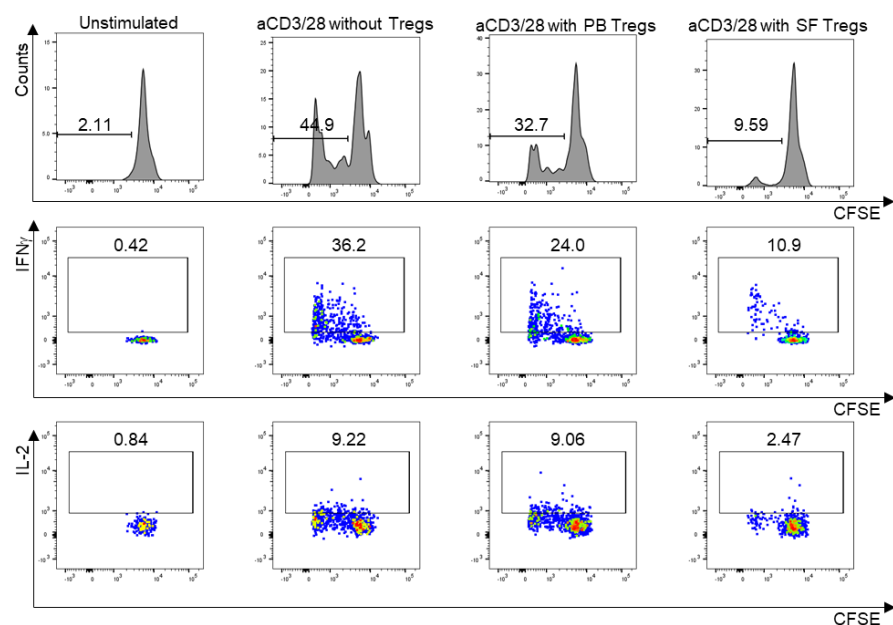
Supplementary Figure 1. Molecular and immunologic analysis of patients with arthritis as an immune-related adverse event (arthritis-irAE). (a-b) Collection of peripheral blood (PB) and/or synovial fluid (SF) samples. PB and/or SF samples were obtained from 20 patients with arthritis-irAE at the time of the active arthritis. We also collected the 2nd PB samples from 7 patients during arthritis-irAE treatment. The samples were analyzed with flow cytometry, single cell RNA sequencing (scRNAseq), single cell T cell receptor sequencing (scTCRseq), multiplex, in vitro regulatory T cell (Treg) suppressor assay, and/or migration assay. As a negative control, SF and PB samples were collected from 4 immune checkpoint inhibitor-naïve patients with osteoarthritis, and PB samples were collected from 9 patients who tolerated immune checkpoint inhibitors well for at least for 12 weeks. (c) Fraction of 29 cell clusters in Fig. 1b across individual patients. (d) Gating strategy for flow cytometry analysis. Live immune cells were detected by gating CD45 and live-dead. One of the most representative plots from an SF sample is shown. FSC-A, forward scatter area; SSC-A, side scatter area; Mφ, macrophages; neu, neutrophils; mDC, myeloid dendritic cells; pDC, plasmacytoid dendritic cells; NK, natural killer cells; DN, double negative, B, B cells; T, T cells. Corresponds to Figure 1. (e-f) Flow cytometry plots to identify cell populations matching with clusters in Fig. 1 (e) and quantitative analysis (f). n=7 (CD27hi CD8+ T; CD27lo CD8+ T; CD16hi Neu) or n=8 (γδ TCR+) PB and matching SF samples. Two-sided paired t-test. PB, peripheral blood; SF, synovial fluid. Source data are provided as a Source Data file.

Supplementary Figure 2

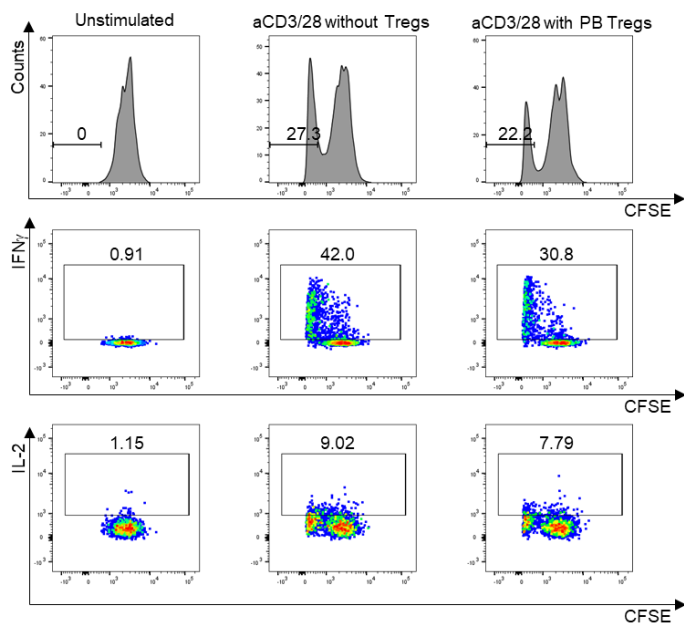
a



b

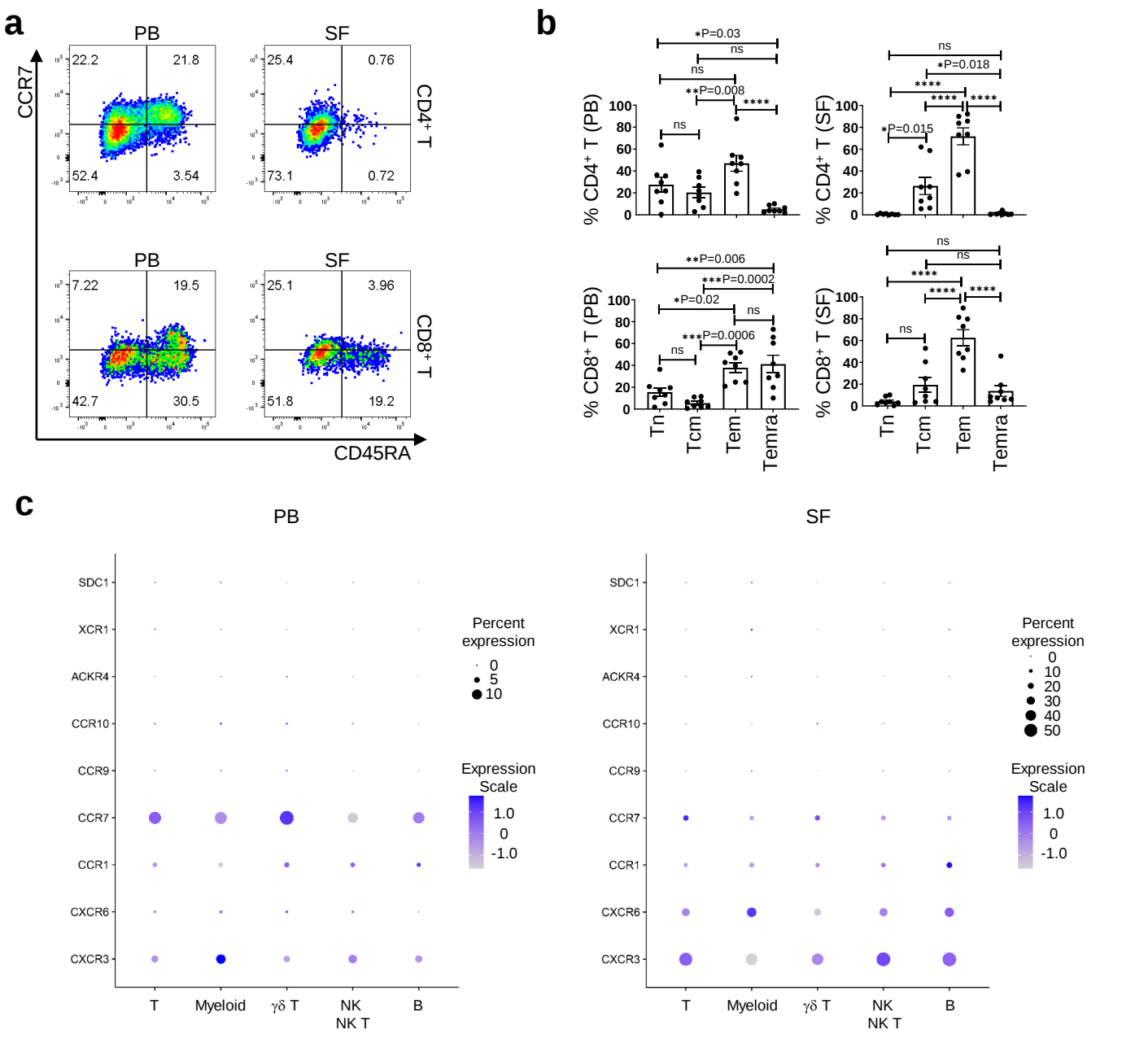


c



Supplementary Figure 2. Representative flow cytometry plots of in vitro regulatory T cell (Treg) suppressor assay. (a) Flow cytometry plots showing sorting strategies. PB, peripheral blood; SF, synovial fluid. Corresponds to Figure 3. (b) 10,000 CFSE-stained PB naïve CD4+ T cells were stimulated with anti-CD3/28 in the absence of autologous Tregs or in the presence of 20,000 autologous Tregs from PB or SF for 4 days. The percentage of CFSE-negative, interferon gamma (IFN γ)-producing, and interleukin-2 (IL-2)-producing populations within naïve CD4+ T cells is shown. (c) Parallel experiments were performed using PB samples from age-, sex-, and tumor-matched patients who did not develop immune-related adverse events at least 12 weeks after initiating immune checkpoint inhibitor therapy.

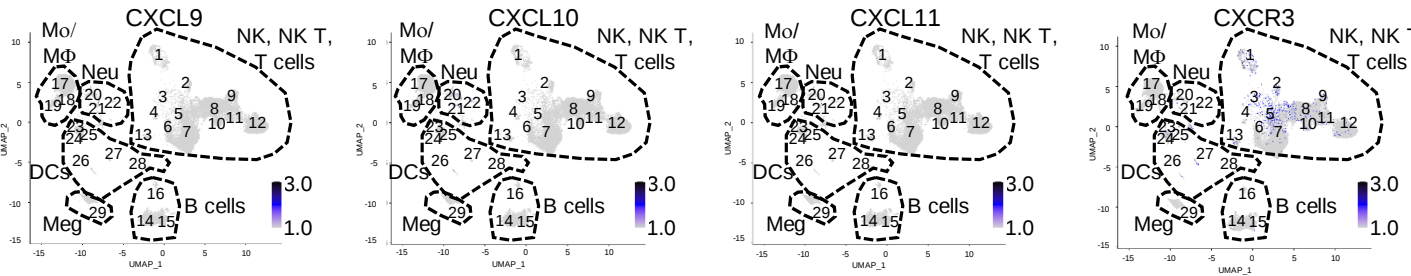
Supplementary Figure 3



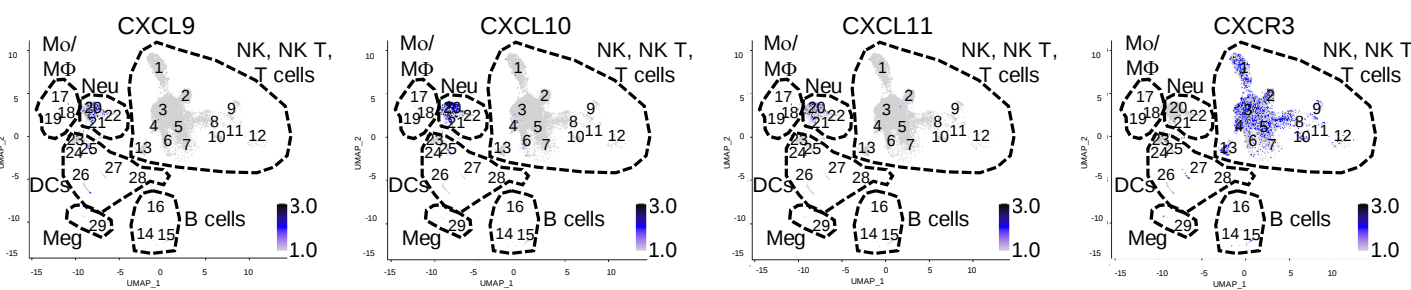
Supplementary Figure 4

a

PB

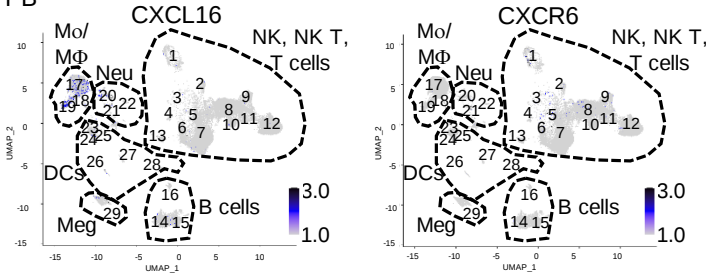


SF

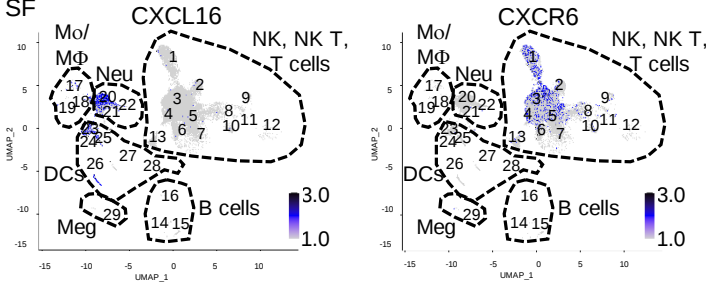


b

PB



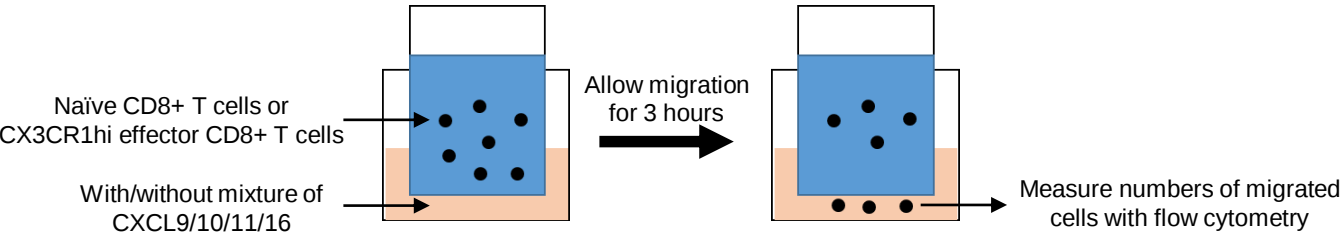
SF



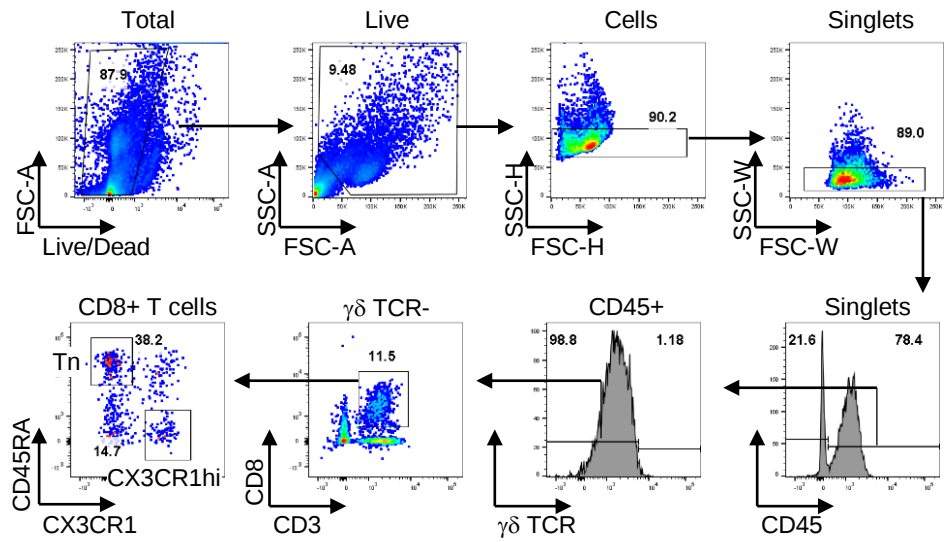
Supplementary Figure 4. Expression of CXCL9/10/11-CXCR3 and CXCL16-CXCR6 on live cells. (a-b) Feature plots showing expression of CXCL9/10/11-CXCR3 (a) and CXCL16-CXCR6 (b) on live cells. Cell clusters and their annotations are identical to those in Fig. 1. Created by authors using R 3.6.0. PB, peripheral blood; SF, synovial fluid.

Supplementary Figure 5

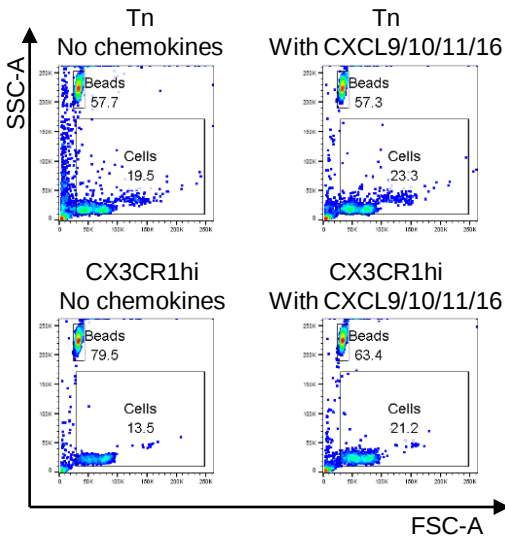
a



b

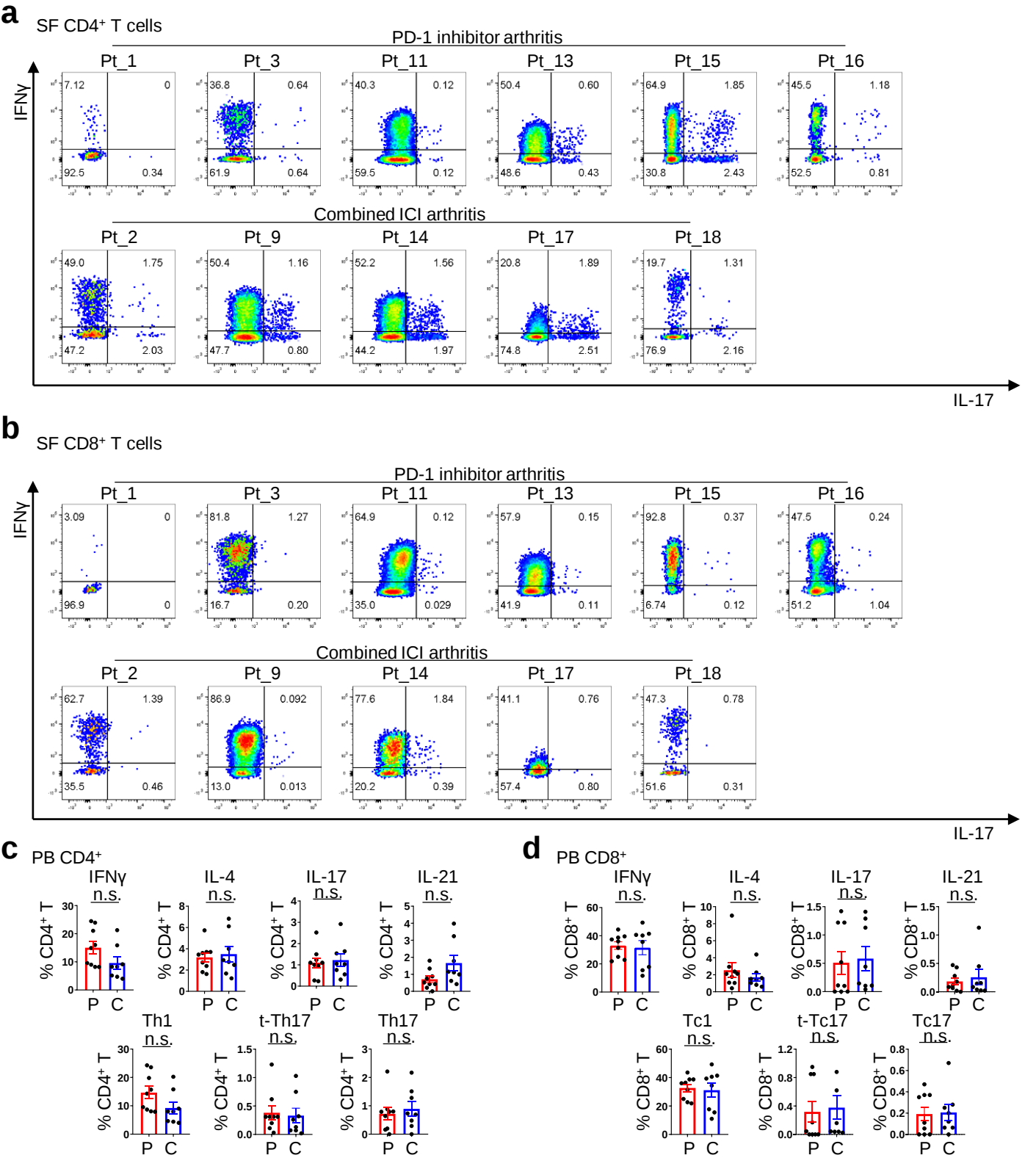


c



Supplementary Figure 5. CX3CR1hi effector CD8+ T cells migrated in response to CXCL9/10/11/16. (a) Schematics showing the experimental designs. (b) Flow cytometry plots showing sorting strategies. Corresponds to Figure 4. (c) After allowing migration of naïve or CX3CR1hi effector CD8+ T cells for three hours in the presence/absence of CXCL9/10/11/16, the cells in the lower chamber were collected. Representative flow cytometry plots taken at the time when 5,000 magnetic beads were collected. Tn, naïve CD8+ T cells; CX3CR1hi, CX3CR1hi effector CD8+ T cells.

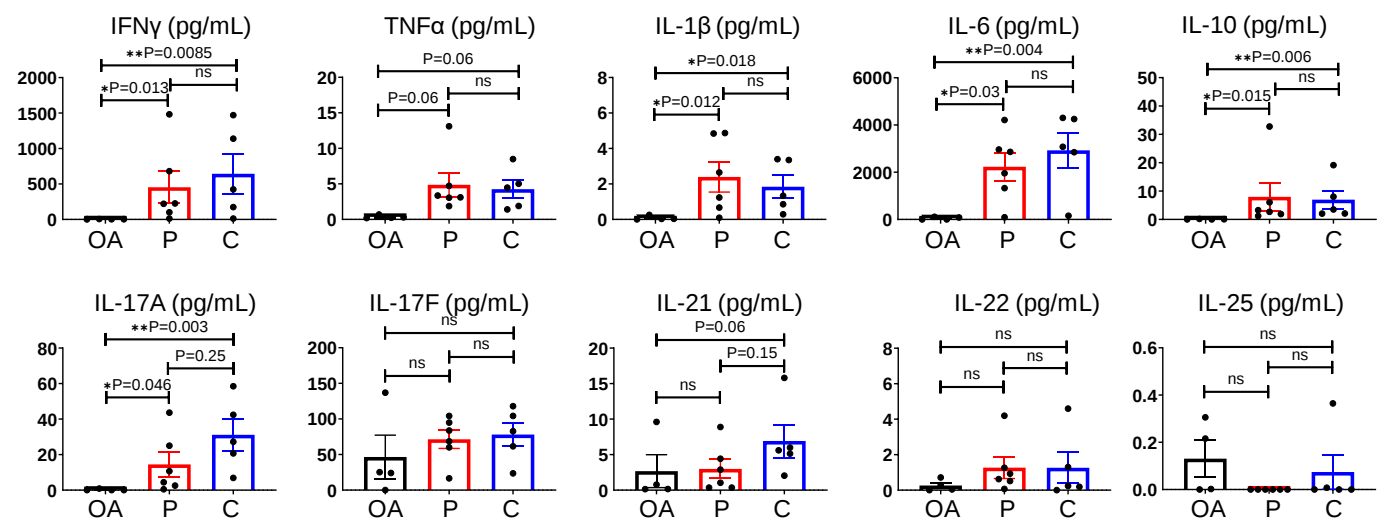
Supplementary Figure 6



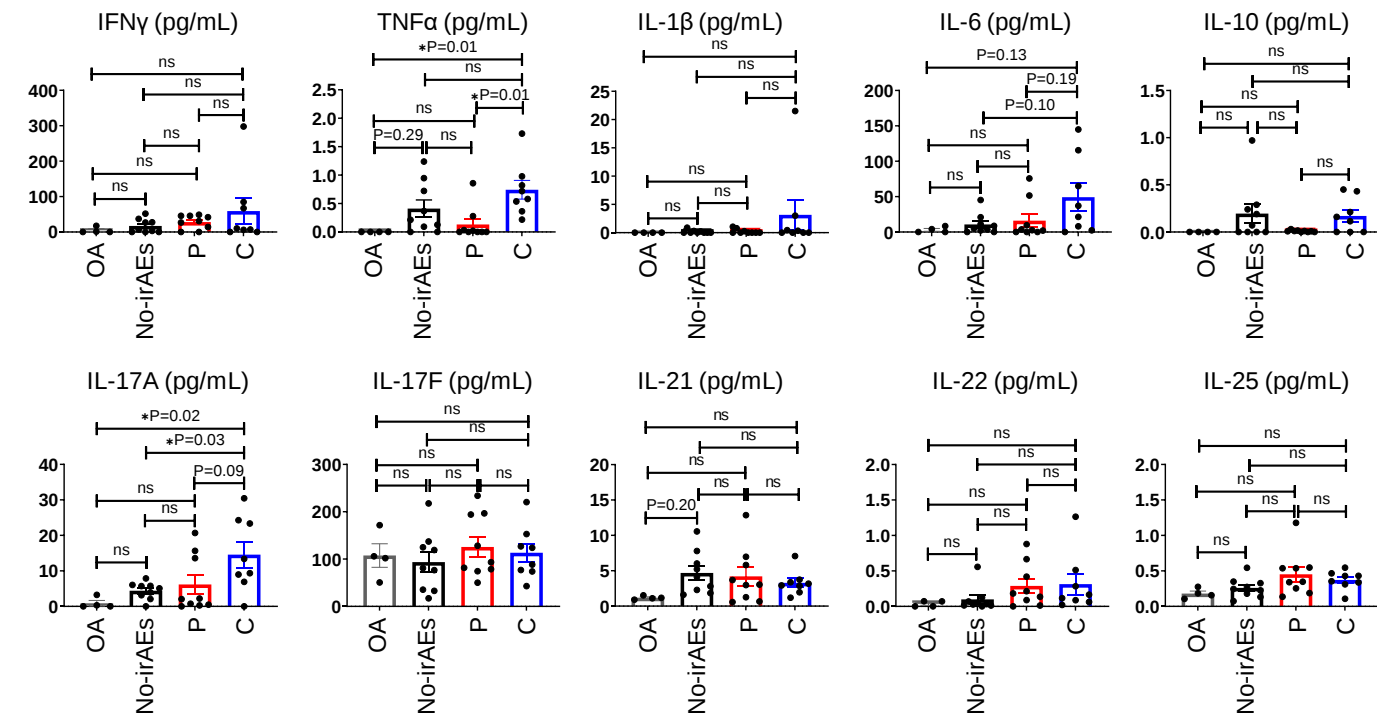
Supplementary Figure 6. Analysis of T cells producing key effector cytokines. (a-b) Intracellular staining of CD4⁺ T cells (a) and CD8⁺ T cells (b) in synovial fluid (SF) of individual patients who developed arthritis after PD-1 inhibitor monotherapy (PD-1 inhibitor arthritis) or combined immune checkpoint inhibitor (ICI) therapy (combined ICI arthritis). IFN γ , interferon gamma; IL, interleukin. (c-d) Proportion of cytokine-producing CD4⁺ T cells (c) and CD8⁺ T cells (d) in peripheral blood (PB; upper panels) and proportions of Th1/Tc1, transient (t-)Th17/t-Tc17, and Th17/Tc17 cells (lower panels). Two-sided unpaired t-test. Bars indicate the mean and SEM. P, PD-1 inhibitor arthritis; C, combined ICI arthritis. n=9 PB samples from the PD-1 inhibitor arthritis group and 8 PB samples from the combined ICI arthritis group. Source data are provided as a Source Data file.

Supplementary Figure 7

a
Synovial fluid supernatant



b
Serum



Supplementary Figure 7. Quantification of inflammatory cytokines in SF supernatant (a) and serum (b). One-way analysis of variance. *P<0.05, **P<0.01. Bars indicate the mean and SEM. No-irAEs; patients who had no irAEs for at least 12 weeks after initiating of immune checkpoint inhibitor (ICI) therapy; OA, osteoarthritis; P, PD-1 inhibitor arthritis; C, Combined ICI arthritis. IFN γ , interferon gamma; IL, interleukin. Supplementary Figure 7a: n=4 (OA), 6 (PD-1 inhibitor arthritis), and 5 (Combined ICI arthritis); Supplementary Figure 7b: n=4 (OA), 9 (no-irAE), 9 (PD-1 inhibitor arthritis), and 8 (combined ICI arthritis). Source data are provided as a Source Data file.

Supplementary Table 1. Information of peripheral blood and synovial fluid samples^a.

Characteristic	No. (%)		
	Arthritis-irAE, n=20	Osteoarthritis, n=4	No irAEs, n=9
PB	9 (45)	0 (0)	9 (100)
SF	3 (15)	4 (100)	0 (0)
Both SF and PB	8 (40)	0 (0)	0 (0)
Median time from first ICI infusion to sample donation (range)	45.8 weeks (5.5-380.5)	N/A	N/A
Median time from development of arthritis to sample donation (range)	8.5 weeks (0.5-38.5)	N/A	N/A
Intra-articular steroids <12 weeks prior to sample donation	0 (0)	0 (0)	N/A
Systemic steroids <4 weeks prior to sample donation	8 (40)	0 (0)	0 (0)
Systemic steroids at the time of sample donation	2 (10)	0 (0)	0 (0)
Characterization of SF			
Median white blood cell count (range)	4,800 cells/ μ L (774-46,360)	643 cells/ μ L (22-643)	N/A
Median percentage of neutrophils (range)	75.0% (0-89)	3% (2-14)	N/A
Median percentage of lymphocytes (range)	13.5% (4-82)	54% (32-82)	N/A
Median percentage of histiocytes (range)	10.5% (4-26)	43% (4-63)	N/A
Crystal positive	0 (0)	0 (0)	N/A
Cultures positive	0 (0)	0 (0)	N/A

^aAbbreviations: PB, peripheral blood; SF, synovial fluid; irAE, immune-related adverse event; ICI, immune checkpoint inhibitor; N/A, not applicable.

Supplementary Table 2. Demographic and clinical characteristics of patients with PD-1 inhibitor arthritis and patients with combined ICI arthritis^a.

Characteristic	No. (%)	
	PD-1 inhibitor arthritis, n=11	Combined ICI arthritis, n=9
Median age (range)	57 years (34-73)	56 years (42-77)
Male	7 (63)	7 (78)
Median BMI (range)	28.2 (23.5-37.0)	28.8 (21.7-37.9)
Tumor type		
Melanoma	4 (36)	7 (77)
RCC	4 (36)	0 (11)
NSCLC	3 (27)	1 (11)
Neuroendocrine tumor	0 (0)	1 (11)
Pattern of arthritis		
Rheumatoid arthritis-like	3 (27)	3 (33)
Reactive arthritis-like	8 (73)	6 (67)
CTCAE grade II-IV irAEs prior to the arthritis		
Colitis	3 (27)	4 (44)
Myocarditis	1 (9)	0 (0)
Type I diabetes mellitus	1 (9)	0 (0)
Hypothyroidism	0 (0)	1 (11)
Neuritis	1 (9)	0 (0)
Median time from first ICI infusion to arthritis symptoms (range)	28.5 weeks (6-70)	87 weeks (0.5-365)
Stopped ICI therapy before arthritis development	4 (36)	3 (33)
Colitis-irAE	1 (9)	0 (0)
Myocarditis-irAE	1 (9)	0 (0)
Neuritis-irAE	1 (9)	0 (0)
Tumor progression	1 (9)	1 (11)
Completion of ICI therapy	0 (0)	2 (22)
Arthritis-irAE CTCAE grade		
II	10 (80)	6 (67)
III	1 (7)	3 (33)
Median CDAI (range)	22.0 (17-25)	20 (7-60)
Median ESR ^b (range)	44 mm/hour (8-95)	58 mm/hour (21-108)
Median CRP ^c (range)	44.7 mg/L (6.8-249)	137 mg/L (6.5-300)
ANA-positive	3 (27)	0 (0)
RF-positive	0 (0)	2 (22)
Anti-CCP antibody-positive	0 (0)	0 (0)

^aAbbreviations: ICI, immune checkpoint inhibitor; BMI, body mass index; RCC, renal cell carcinoma; NSCLC, non-small cell lung carcinoma; CTCAE, common terminology criteria for adverse events; irAE, immune-related adverse event; CDAI, clinical disease activity index; ESR, erythrocyte sedimentation rate; CRP, C-reactive protein; ANA, anti-nuclear antibody; RF, rheumatoid factor; CCP, cyclic citrullinated peptide.

^bNormal values of ESR range from 0 to 20.

^cNormal values of CRP range from 0 to 10.