Supplementary Table 1: Flow cytometry panel, initial (top) and modified (bottom). Mac = macrophage; Mono = monocyte; OB = osteoblast; RSM = Resident Synovial Macrophage; APC = Antigen Presenting Cell

Molecular Target	Fluorochrome	Clone	Vendor	Cell of Interest
CD11b	BV605	ICRF44	BioLegend	Myeloid
CD11c	BV785	3.9	BioLegend	Dendritic Cell
CD14	PE-Cy7	63D3	BioLegend	Mac/Mono
CD20	BV785	2H7	BioLegend	B cell
CD3	BV650	UCHT1	BioLegend	T Cell
CD45	AF700	2D1	BioLegend	Leukocytes
CD56	PE-Cy5	5.1H11	BioLegend	NK cell
CD68	PerCP-Cy5.5	Y1/82A	BioLegend	Macrophage
CX3CR1	BV421	2A9-1	BioLegend	RSM/Mac/Mono
HLA-DR	PE	Tu36	BioLegend	Activated APC
OPG/TNFRSF11B	AF488	155321	R&D Systems	RSM
RANKL/TNFSF11	AF350	685857	R&D Systems	OB/activated T cells
TREM2	APC	237920	R&D Systems	Macrophage
Viability	Live/Dead Aqua		Thermofisher	

Molecular Target	Fluorochrome	Clone	Vendor	Cell of Interest
CD11b	BV605	ICRF44	BioLegend	Myeloid
CD11c	BV785	3.9	BD Biosciences	Dendritic Cell
CD14	Spark Blue 550	63D3	BioLegend	Mac/Mono
CD20	BV785	2H7	BD Biosciences	B cell
CD3	BV510	SK7	BioLegend	T Cell
CD45	AF700	HI30	BioLegend	Leukocytes
CD56	PE-Cy5	5.1H11	BioLegend	NK cell
CD66b	PE	MIH24	BioLegend	Neutrophil
CD68	R718	Y1/82A	BD Biosciences	Macrophage
CX3CR1	BV711	2A9-1	BioLegend	RSM/Mac/Mono
F11R/JAM-1/JAM-A	BV421	M.Ab.F11	BD Biosciences	RSM
OPG/TNFRSF11B	AF488	155321	R&D Systems	RSM
TREM2	APC	237920	R&D Systems	Macrophage
ZO-1/TJP1	Coralite 594	polyclonal	ThermoFisher	RSM
Viability	Live/Dead Blue		ThermoFisher	

Supplemental Table 2: Patient Demography. Arthritis Type was determined by synovial fluid WBC count per mm^3, %PMNs, and/or culture results. * = 3 patients had Lyme Arthritis, 1 of which had both a *S. aureus* and positive IgG with confirmatory Western Blot.

Demographics	n=52	
Age	(years)	
	Mean (STD)	51.7 (21)
	Median	58
	Range	3-77
Gender	n, (%)	
	Male	35 (68)
	Female	17 (32)
Race and Ethnicity	n, (%); Hispanic n, (%)	
	White	45 (87); 1 (2)
	Black	3 (6); 0
	Asian	1 (2); 0
	Multiracial	1 (2); 0
	Unknown	1 (2); 0
Arthritis Type	n (%)	
	Normal/Non-I	9 (25)
	Inflammatory	8 (22)
	Septic*	18 (50)
	Hemorrhagic	1 (3)

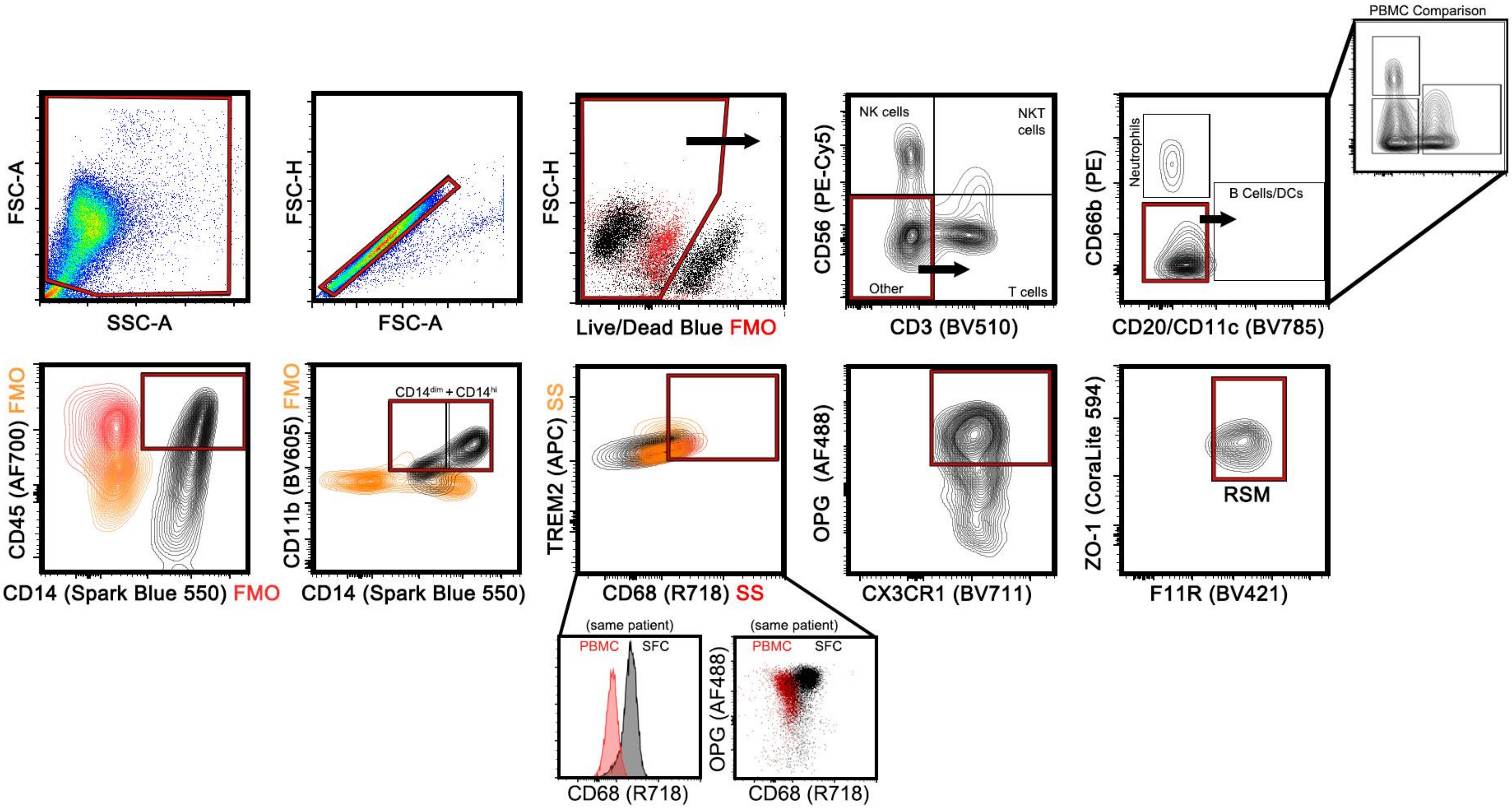
Supplementary Table 3: Patient Diagnosis. SF analysis was performed on patients when it was clinically indicated. SF diagnosis was based on WBC count (# nucleated cells) and % PMN based on neutrophil count. * = not performed. ** = Lyme Arthritis in addition to other bacterial arthritis.

Patient	Color	Clarity	Crystals	# Nucleated Cells	# Neutrophils	# Lymphocytes	Arthritis Type
2	Red	Turbid	CCP, MSU	29783	22635	101	Inflammatory
3	Yellow	Turbid	CPP	4716	2453	20	Inflammatory
5	Red	Turbid	none	822	674	107	Non-Inflammatory
6	Yellow	Turbid	none	29497	28022	0	Septic
7	Yellow	Hazy	none	714	29	64	Non-Inflammatory
9	Yellow	Hazy	none	63726	60539	1275	Septic
10	Yellow	Slightly Hazy	none	464	28	70	Non-Inflammatory
11	Red	Turbid	none	671	658	0	Non-Inflammatory
13	Orange	Hazy	none	831	632	58	Non-Inflammatory
14	None	Clear	none	29	0	7	Normal
15	Orange	Turbid	none	63184	60025	632	Inflammatory
16	Pink	Hazy	none	166627	128303	9998	Septic
18	Amber	Turbid	none	134280	119509	2686	Septic
19	Red	Turbid	none	11522	8987	1959	Inflammatory
21	N/A	N/A	none	N/A	N/A	N/A	Septic
22	Yellow	Turbid	CPP	34482	27586	2	Inflammatory
23	Yellow	Clear	none	348	28	52	Non-Inflammatory
24	Red	Turbid	none	1819	1564	14	Hemorrhagic
26	Yellow	Clear	none	26	5	12	Normal
27	Orange	Turbid	MSU	33805	26030	0	Inflammatory
28	Brown	Turbid	none	66945	*	*	Septic
30	Yellow	Slightly Hazy	none	26536	25209	531	Septic
31	Red	Hazy	none	48363	46428	484	Septic
32	Orange	Hazy	none	43355	42054	434	Septic
33	Red	Turbid	none	304	76	85	Septic
34	Red	Hazy	none	854	384	154	Non-Inflammatory
38	Red	Turbid	none	114000	1000320	2	Septic
39	Red	Turbid	MSU	76890	76121	0	Inflammatory
40	Pale Yellow	Turbid	CPP	67368	64673	0	Inflammatory
41	Pink	Turbid	None	79118	75162	0	Septic**
43	Orange	Turbid	None	104595	92044	2	Septic
46	Pink	Turbid	None	80562	74117	2417	Septic
47	Red	Turbid	None	9884	8303	692	Septic
48	Orange	Hazy	None	130	83	26	Normal
49	Yellow	Hazy	MSU	20566	18715	0	Inflammatory

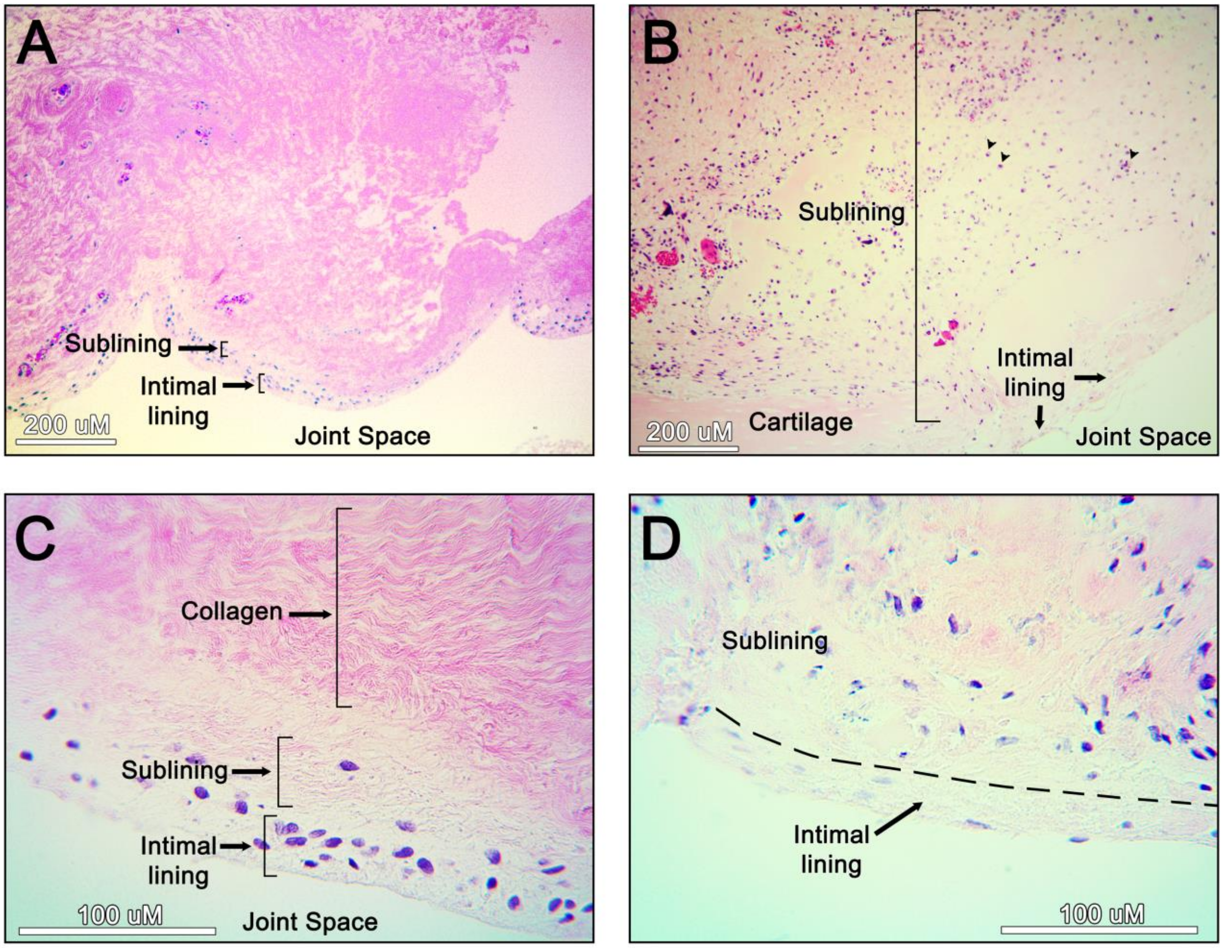
Supplementary Table 4: List of all significant conserved genes in RSM, Cytotoxic DC, and NK/NKT clusters.

Gene	Log2FC	Δ % Septic	Δ % Inflam	Cluster	Gene Function
PKIB	2.10657	0.71	0.726	DC	Also known as AKT, causes M2 formation
SKAP1	3.16933	0.89	0.954	NK/NKT	TCR adaptor protein
CD96	2.690931	0.77	0.874	NK/NKT	Inhibitory, marker of exhaustion
CD2	2.571958	0.78	0.817	NK/NKT	Regulates lytic activity
CD247	2.473159	0.79	0.869	NK/NKT	Subunit of T cell antigen receptor complex
ETS1	2.430819	0.85	0.88	NK/NKT	Required for NK differentiation and cytotoxic function
SLC38A1	2.222173	0.82	0.77	NK/NKT	Glutamine transporter
IDO1	3.679276	0.93	0.764	Cytotoxic DC	Immunosuppressive, expressed by DCs, macs, and epithelial cells
C1orf54	3.555745	0.94	0.794	Cytotoxic DC	cDC1 gene
CCSER1	3.110627	0.78	0.818	Cytotoxic DC	upregulated in anti-inflammatory conditions
CLEC9A	3.009585	1.00	0.888	Cytotoxic DC	DC receptor for necrotic cell death
CPNE3	2.908024	0.82	0.76	Cytotoxic DC	cDC1 gene
AUTS2	2.836438	0.78	0.74	Cytotoxic DC	associated with autism
CADM1	2.766723	0.99	0.936	Cytotoxic DC	expressed by pre-cDC1, cell adhesion
NEGR1	2.54191	0.97	0.798	Cytotoxic DC	immunoglobulin superfamily cell adhesion molecule subgroup IgLON, has been implicated in neuronal growth and connectivity
CLNK	2.441584	0.99	0.885	Cytotoxic DC	upregulated in response to IL-2 and IL-3, associated with SLP-76
DNASE1L3	2.326661	0.89	0.998	Cytotoxic DC	Cytokine secretion following inflammasome activation in response to DNA in circulating apoptotic bodies
ARL4C	2.315802	0.71	0.709	Cytotoxic DC	oncogene, tubulogenesis, wnt-beta catenin signaling
NAAA	2.166039	0.71	0.78	Cytotoxic DC	inflammatory, lysosomal
KIF16B	1.944567	0.79	0.778	Cytotoxic DC	microtubule formation associated with endosomes and cross presentation
DST	1.91132	0.77	0.764	Cytotoxic DC	junctional adhesion protein
PLPP1	1.872874	0.74	0.736	Cytotoxic DC	phospholipid lipase associated with mac inflammation, cDC1 marker
MCOLN2	1.841604	0.73	0.824	Cytotoxic DC	MHCII presentation, innate immune cell activation, enhances infectivity of certain viruses, interferon stimulating gene
CALCRL	1.840268	0.76	0.951	Cytotoxic DC	regulates DC function through NF-kB

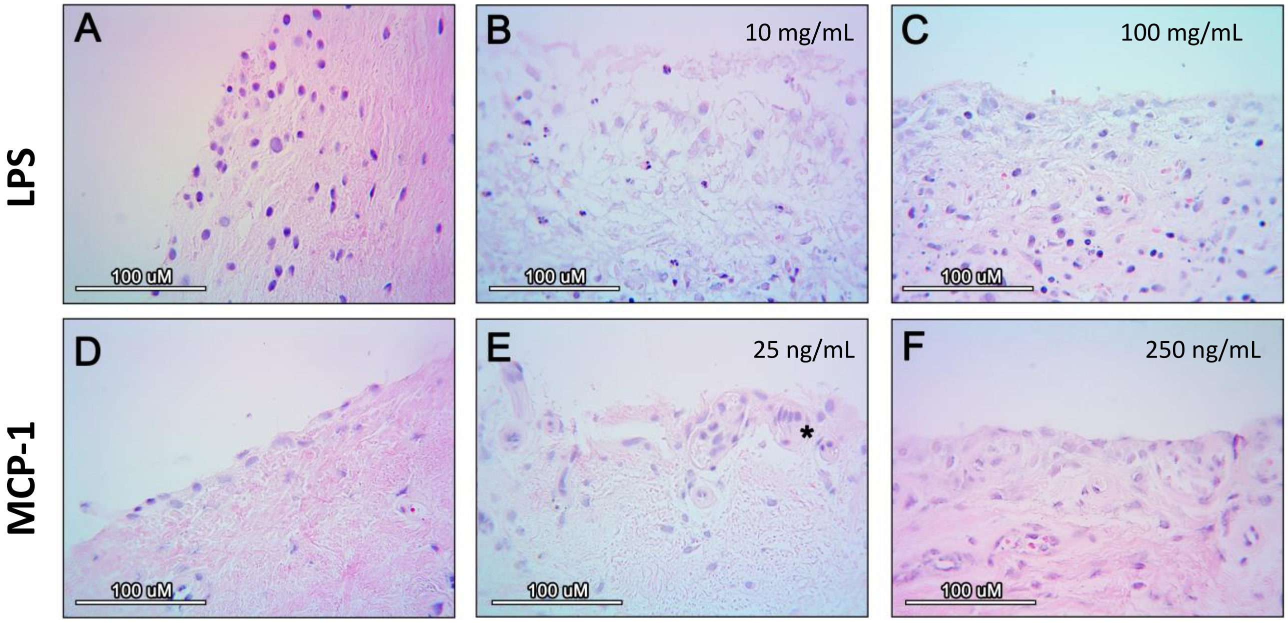
TACSTD2	1.765963	0.78	0.989	Cytotoxic DC	Marker for TGF-B1 dependent DCs
ZEB1	1.672937	0.74	0.941	Cytotoxic DC	cDC1 cells
C1orf21	1.647088	0.84	0.9	Cytotoxic DC	enriched in cytotoxic CD8 T cells
FLT3	1.645133	0.96	0.895	Cytotoxic DC	The growth factor Flt3 ligand (Flt3L) is central to dendritic cell (DC) homeostasis and development, controlling survival and expansion by binding to Flt3 receptor tyrosine kinase on the surface of DCs.
ENPP1	1.548458	1.00	0.777	Cytotoxic DC	suppresses innate immune response,
NUBPL	1.540362	0.83	0.911	Cytotoxic DC	mitochondrial gene, iron and sulfur
XCR1	1.53506	0.78	0.995	Cytotoxic DC	DC, induces CD8 responses
DAPP1	1.52997	0.72	0.717	Cytotoxic DC	immune related mucosal tissues, hyper-reactive airway disease, IDO1/CCR7 signaling, associated with DCs
LAMP3	3.181144	0.88	0.967	RSM	lysosome
CST7	2.710421	0.93	0.748	RSM	Cistatin F, lysosomal cathepsin inhibitor
DAPP1	2.225168	0.72	0.781	RSM	immune related mucosal tissues, hyper-reactive airway disease, IDO1/CCR7 signaling, associated with DCs
GPR157	2.163834	0.75	0.897	RSM	G protein coupled receptor 157
TBC1D4	2.161417	0.79	0.941	RSM	Rab GTPase Activating Protein
RFTN1	2.085313	0.77	0.736	RSM	glucose transport
CCR7	2.083676	0.92	0.837	RSM	M1 polarization
LAD1	1.96187	0.77	0.713	RSM	epithelialization
SLC41A2	1.415962	0.76	0.78	RSM	magnesium transporter
REPIN1	1.325367	0.76	0.72	RSM	increasing cell size and tumor metastasis
SPIB	1.005641	0.75	0.726	RSM	recruits TAMs via CCL4 signaling



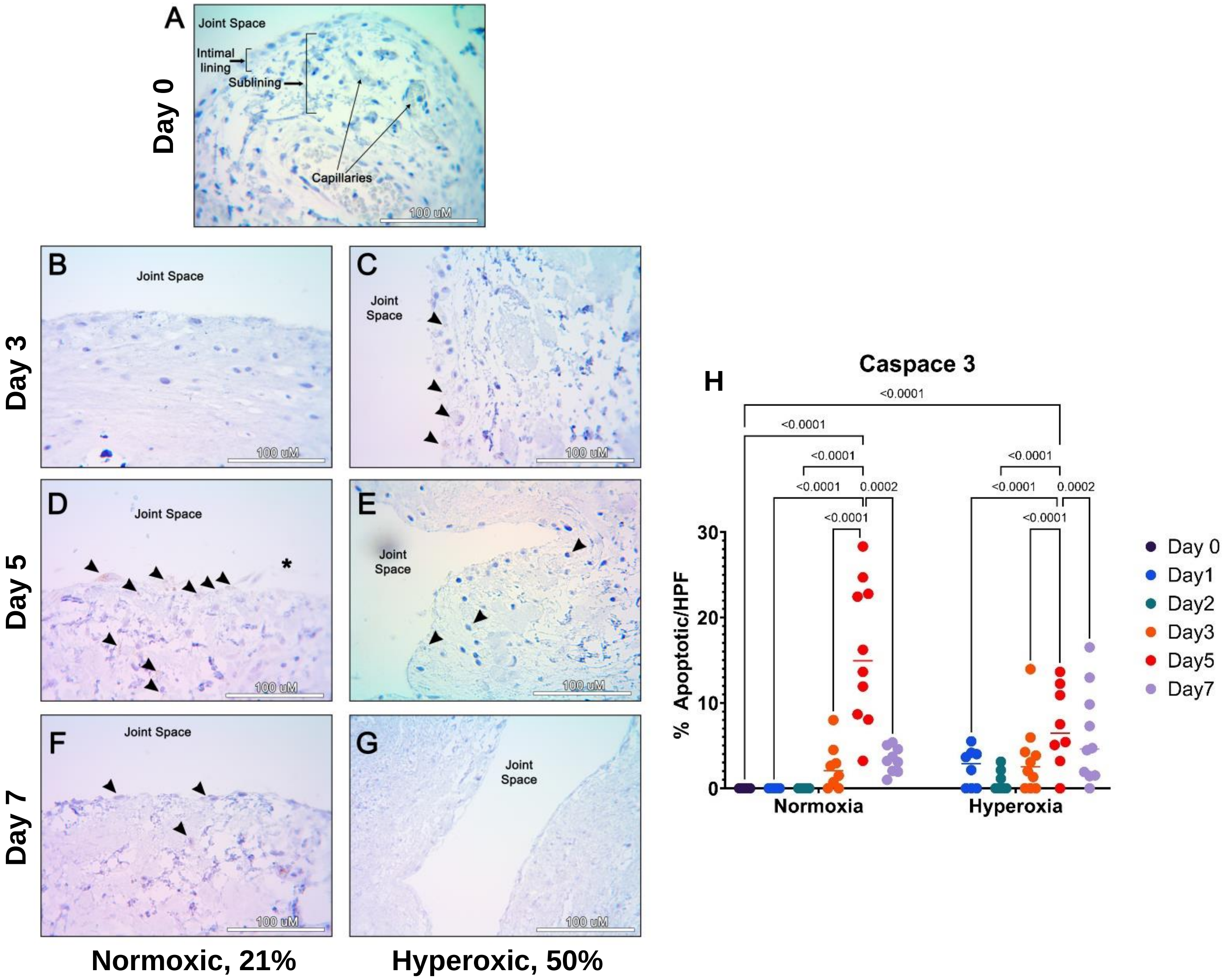
Supplemental Figure 1: Final gating strategy to identify TREM2$^+$CX3CR1$^+$OPG$^+$F11R$^+$ZO-1$^+$ macrophages consistent with RSM. FMO and single stain (SS) shown where used to define gates.



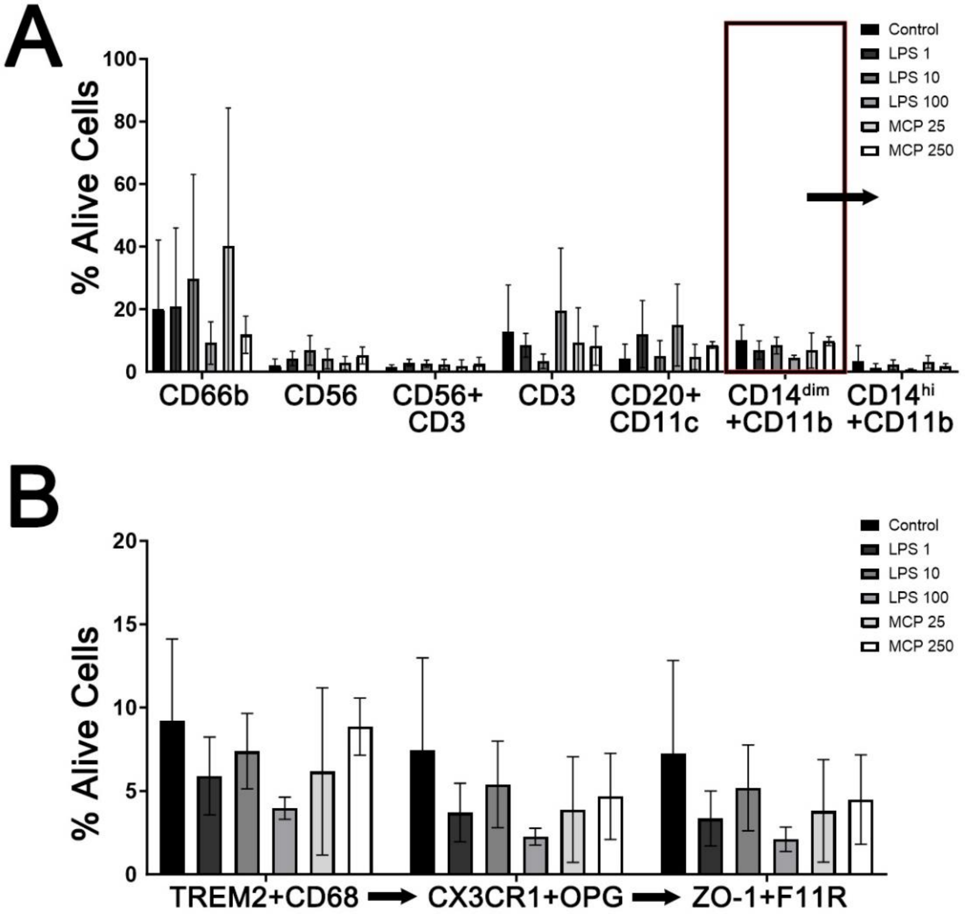
Supplemental Figure 2: *In vivo* negative (A, C, same patient, 10X and 40X respectively) and positive (B, D, second patient, 10X and 40X respectively) controls. Negative control patient displays fibrosis, but positive control (chronic bacterial infection) patient has evident dysfunction of synovium with cartilage formation, evident migration of inflammatory cells into the sublining, and thinning/loss of cellularity of the initial lining (B, D).



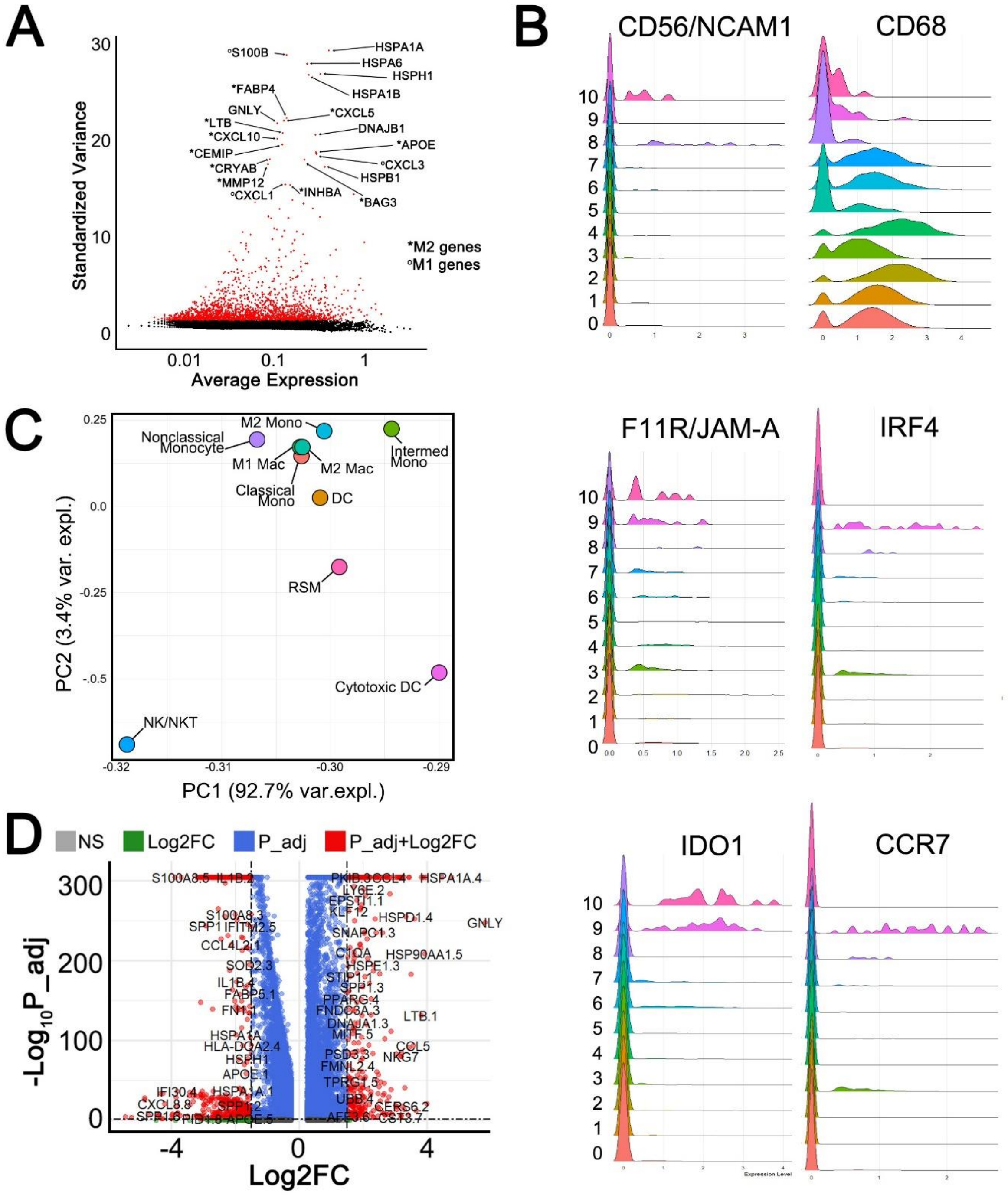
Supplementary Figure 3: disruption of the synovial intimal lining and migration of inflammatory cells at 24 hours is dependent upon stimulus dose. A-C are from same patient, D-F are from a second patient, all at 24 hours.



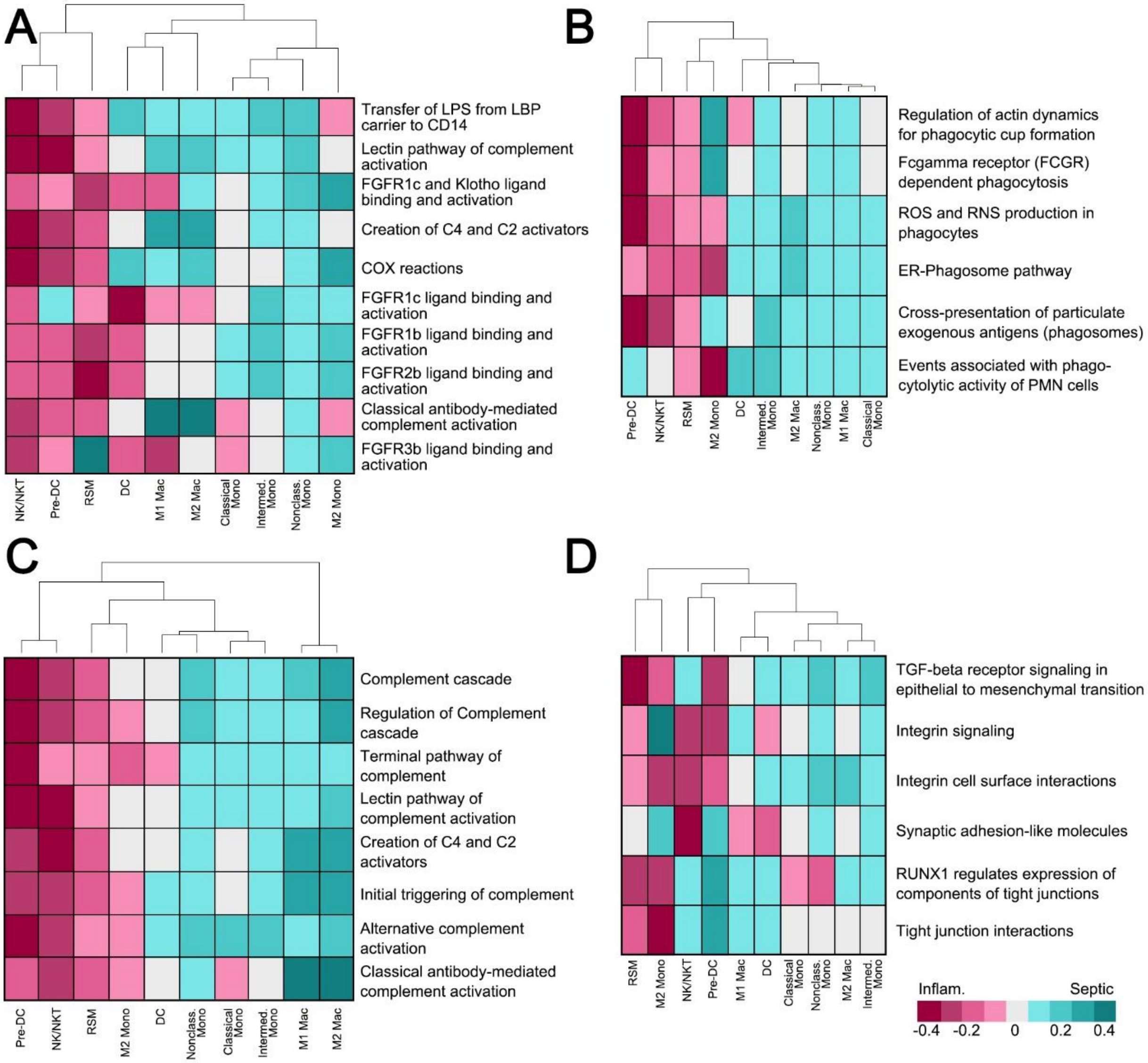
Supplemental Figure 4: Caspace-3 IHC and quantification for explant culture out to 7 days in normoxic (B, D, F) and hyperoxic (C, E, G) conditions. Arrowheads point out positive IHC for Caspace-3. Apoptosis became evident in normoxic conditions on day 3 of culture, compared to day 1 for hyperoxic conditions. Two-way ANOVA with Tukey's *post-hoc* correction (H).



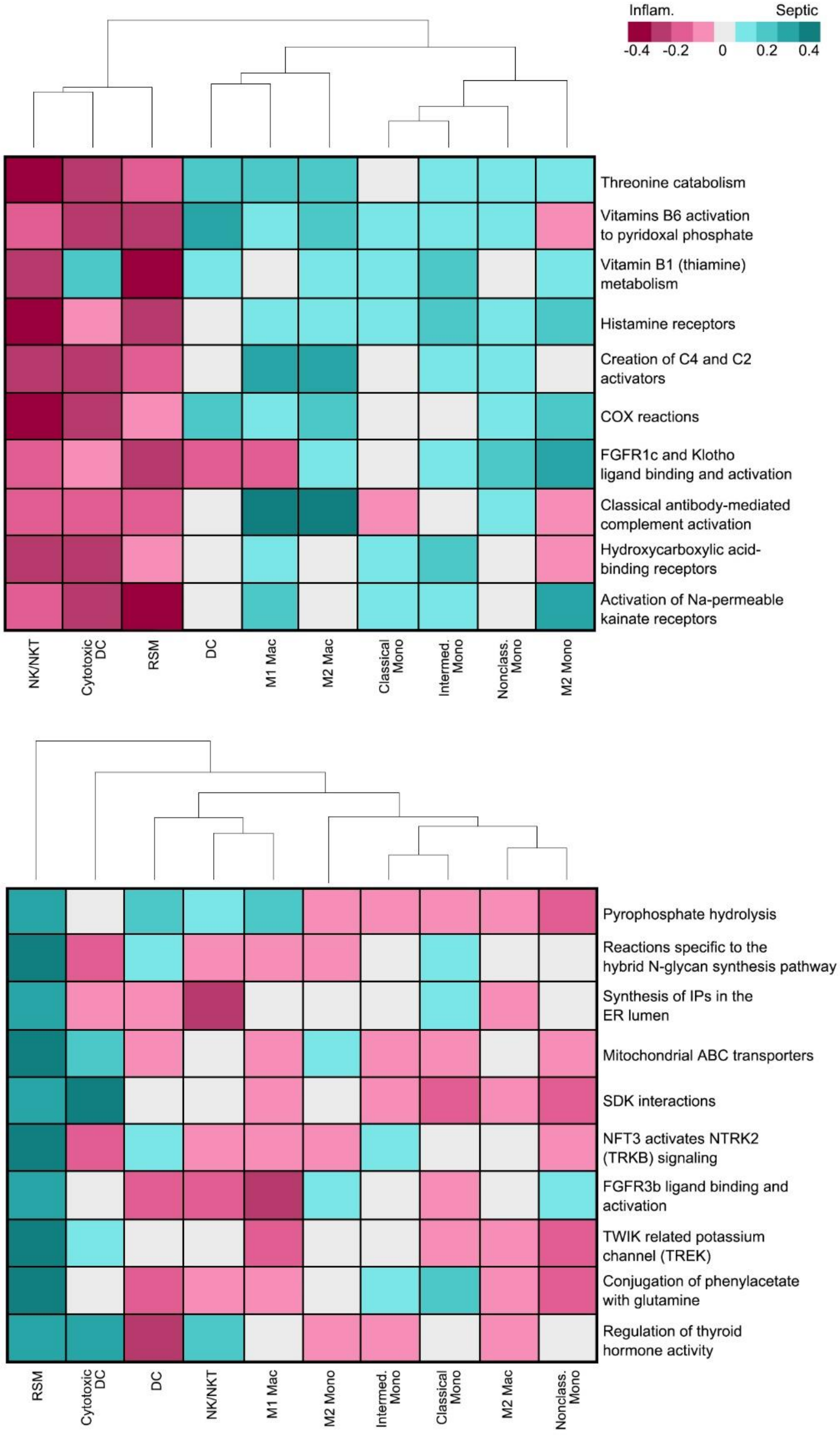
Supplementary Figure 5: Evaluation of migratory cell populations in the bottom transwell. Neutrophils (CD66b), NK (CD56), NKT (CD3 and CD56), T cells (CD3), B cells or Dendritic Cells (CD20 and CD11c, same fluorophore), and CD14 high or dim myeloid (CD11b) cells (**A**). Sequential analysis of the CD14^{dim} myeloid population demonstrating nearly all CD14^{dim} macrophages are positive for OPG, CX3CR1, ZO-1, and F11R (**B**). LPS in µg/mL, MCP-1 in ng/mL.



Supplemental Figure 6: Evaluation of scRNA-seq data. Variance analysis with top 20 genes labeled (A). Ridge plots of NK, M2, and RSM associated genes (B). Principle component analysis of all 10 clusters not including dead cells (C). Volcano plot of most up- and down-regulated genes, where minimum Log2FC was 1.5 for significance given homogeneity of sample (D).



Supplemental Figure 7: GSA for top 10 immune-relevant pathways (A), phagocytosis (B), complement (C), and adhesion-related (D).



Supplemental Figure 8: GSA for RSMs: top ten genes upregulated in inflammatory arthritis (top) and septic arthritis (bottom).