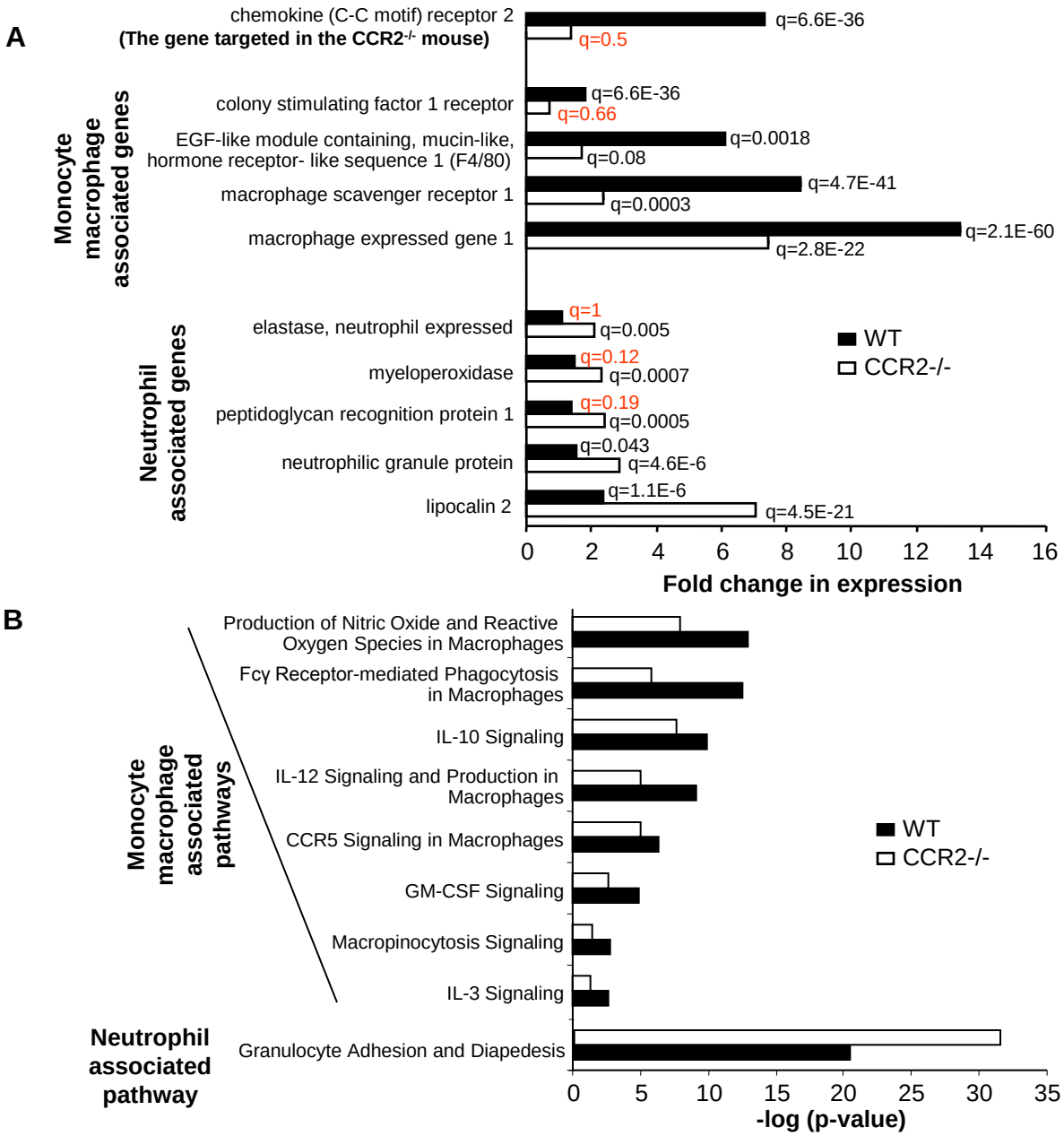
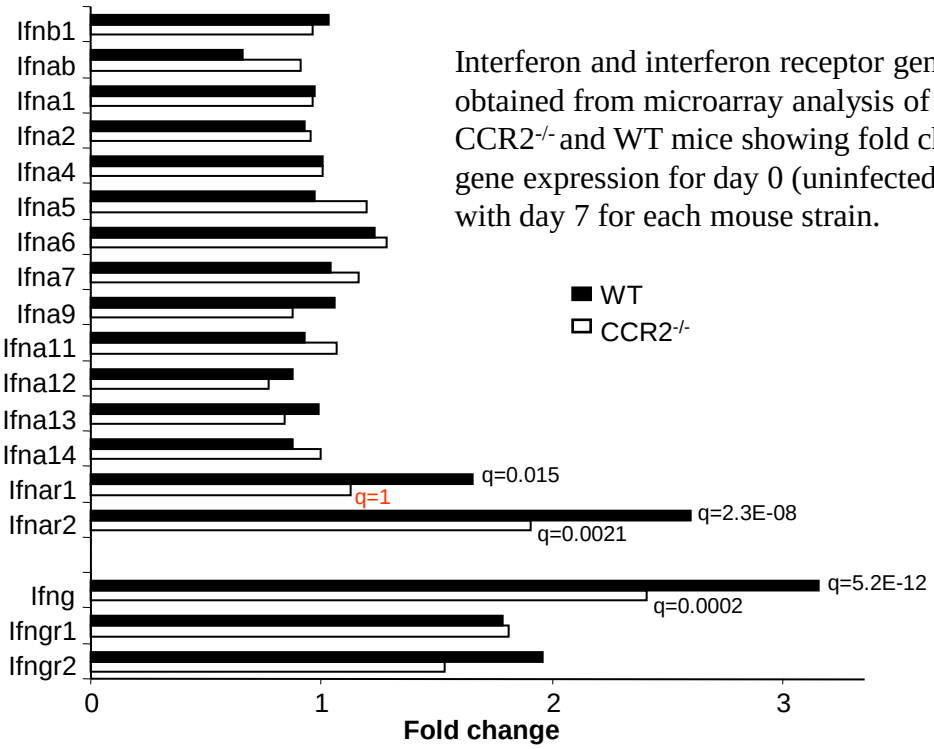
Supplemental material: Figures

Figure S1. Monocyte/macrophage and neutrophil associated genes and pathways in feet of CHIKV infected CCR2$^{-/-}$ and WT mice.

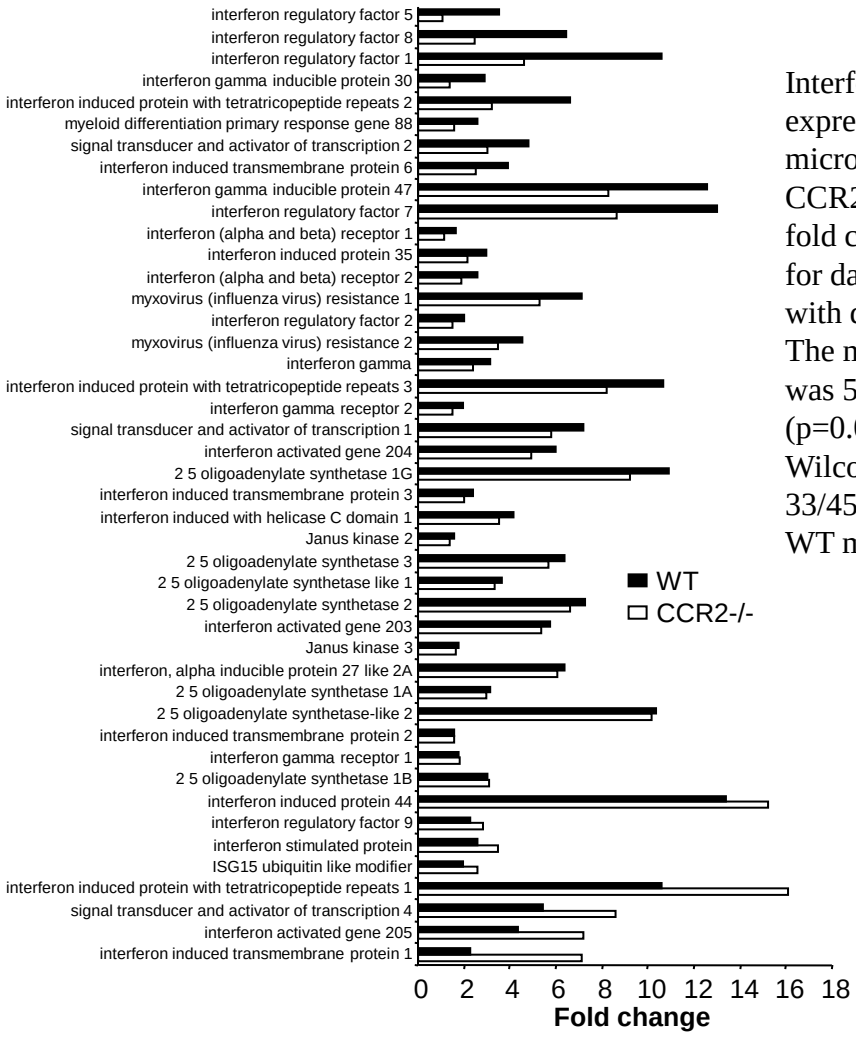


(**A**) Fold change in expression of up-regulated genes associated with monocyte/macrophage or neutrophils. Fold changes for DEGs from CCR2$^{-/-}$ d0 vs d7, and WT d0 vs d7 are shown with their q significance values (red values - not significant). The gene expression pattern is consistent with the histology and immunohistochemistry findings; neutrophil infiltrates predominate in arthritic feet of CCR2$^{-/-}$ mice and monocyte/macrophage infiltrates predominate in arthritic feet of WT mice.
(**B**) Ingenuity canonical pathway analysis of up-regulated DEGs from CCR2$^{-/-}$ d0 vs d7, and WT d0 vs d7. Monocyte/macrophage associated pathways and a neutrophil-associated pathway (Granulocyte Adhesion and Diapedesis) and are shown. (IPA has relatively few pathways dedicated to neutrophils). These results are again consistent with histology and immunohistochemistry findings.

Figure S2. Interferon-associated gene expression in feet of CHIKV infected CCR2$^{-/-}$ and WT mice

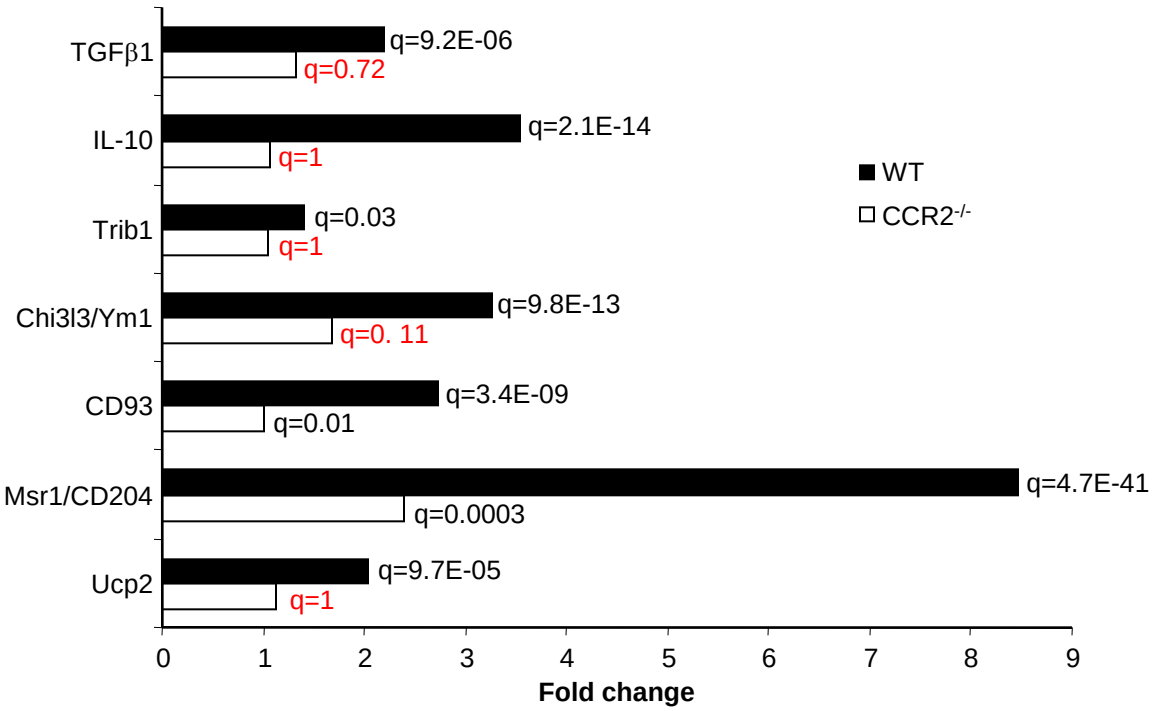


Interferon and interferon receptor gene expression obtained from microarray analysis of feet from CCR2$^{-/-}$ and WT mice showing fold change in gene expression for day 0 (uninfected) compared with day 7 for each mouse strain.



Interferon associated gene expression obtained from microarray analysis of feet from CCR2$^{-/-}$ and WT mice showing fold change in gene expression for day 0 (uninfected) compared with day 7 for each mouse strain. The mean fold change for WT was 5.15 and for CCR2$^{-/-}$ 4.63 ($p=0.012$, related samples Wilcoxan Signed Rank test), with 33/45 genes more up-regulated in WT mice.

Figure S3. Expression of genes associated with M2 and resolving macrophages in feet of CHIKV infected CCR2$^{-/-}$ and WT mice



Microarray expression data (with significance q values) for genes associated with M2 and resolving macrophages (for both strains of mice, day 7 versus day 0); TGFβ1, IL-10 (Ariel and Serhan. 2012. Front Immunol 3:4), Trib1 (Satoh et al., 2013. Nature. 495(7442):524-8), Ym1 (Stoermer et al., 2012. J Immunol 189;4047-59), CD93 (Beyer et al., 2012. PLoS One. 2012;7(9):e45466), Msr1 (London et al., 2013. J Immunol.. 190(7):3570-8), Usp2 (Park et al., 2011. Nature. 477;220-4). Ucp2 is up-regulated during efferocytosis (*ibid*) and was up-regulated in WT mice, but not in CCR2$^{-/-}$ mice, despite the abundance of apoptotic cells (Figure 3C), many of which were neutrophils (Figure 2M). (Red q values - not significant)

Supplemental material; Tables

Table S1. Primers used for qRT PCR

Gene	Sequences (5' to 3')	References
Arg-1	Forward: TGGACCTGGCCTTTGTTGA Reverse: GGTTGTCAGGGGAGTGTT	Wongtrakool et al., 2012. Pediatr Res 72: 147-53
CHIKV-E1	Forward: AGCTCCGCGTCCTTTACC Reverse: CAAATTGTCCTGGTCTTCCTG	Designed in house
CXCL1	Forward: TGTCAGTGCCTGCAGACCAT Reverse: CCTCGCGACCATTCTTGAGT	Feng et al., 2006. J Immunol 177: 7086-93
CXCL2	Forward: GTGAAC TGCGCTGTCAATGC Reverse: CGCCCTTGAGAGTGGCTATG	*Ibid*
G-CSF	Forward: CTCAACTTTCTGCCCAGAGG Reverse: AGCTGGCTTAGGCACTGTGT	Waight et al., 2011. PLoS One 6: e27690
IL-10	Forward: ACAGGAGAAGGGACGCCAT Reverse: GAAGCCCTACAGACGAGCTCA	Ellet et al., 2010. J Immunol 184: 5849-58
IL-1β	Forward: TCACAGCAGCACATCAACAAG Reverse: CCAGCAGGTTATCATCATCATCC	Bakker et al., 2008. Cardiovasc Res 78: 341-8
T-bet[a]	Forward: ACCAGAGCGGCAAGTGGG Reverse: TGGACATATAAGCGGTTCCCTG	Carlson et al., 2009. Blood 113: 1365-74
ROR-γT[b]	Forward: CCGCTGAGAGGGCTTCAC Reverse: TGCAGGAGTAGGCCACATTACA	*Ibid*
RPL13A	Forward: GAGGTCGGGTGGAAGTACCA Reverse: GCAT CTTGGCCTTTTCCTT	Mogal and Abdulkadir, 2006. Mol Cell Probes 20: 81-6

Sequences of primers used in the qRT-PCR experiments. [a]T-bet is a transcription factor expressed by Th1 cells. [b]ROR-γT is a transcription factor expressed by Th17 cell.

Table S2. Fold changes in chemokine and chemokine receptors in feet of CHIKV infected CCR2$^{-/-}$ and WT mice.

Gene Symbol	CCR2-/- fold change	q value	WT fold change	q value	Ratio fold change CCR2$^{-/-}$ / WT
Cxcl2[a]	7.1	4.45E-21	1.4	0.306	5.205
Cxcl5[a]	7.8	2.25E-23	1.6	0.048	5.028
Cxcr2[b]	6.3	9.55E-19	1.3	0.336	4.691
Cxcl13	6.6	1.35E-19	2.3	2.86E-06	2.888
Ccrl2	3.7	1.76E-09	1.8	0.005	2.114
Ccl20	2.1	0.003	1.0	1.002	2.099
Ccl3/MIP-1α[a,b]	13.5	2.41E-37	7.5	1.18E-36	1.803
Cxcr6	6.5	1.88E-19	3.7	2.35E-15	1.778
Ccr3	2.0	0.008	1.2	0.870	1.717
Ccl19	3.7	1.76E-09	2.2	7.26E-06	1.670
Cxcl16	1.8	0.037	2.8	1.91E-09	0.660
Cxcr4	1.8	0.0661	2.8	2.54E-09	0.637
Ccl7[c]	8.3	9.48E-25	16.4	1.79E-70	0.507
Ccl12[c]	1.7	0.091	3.4	1.76E-13	0.503
Ccr5[c]	3.8	1.11E-09	18.9	1.33E-77	0.199
Ccr2	1.4	0.518	7.4	6.66E-36	0.191

Many chemokine and chemokine receptors were more up (red numbers) or more down (blue numbers) regulated in CCR2$^{-/-}$ mice compared with WT mice day 7 post infection; (only DEGS where differences between WT and/or CCR2$^{-/-}$ were >1.5 are shown). CCR2 (indicated in yellow) is the target of the knock out in CCR2$^{-/-}$ mice and is up-regulated in WT mice, whereas (as expected) expression is not significant (q=0.518) in CCR2$^{-/-}$ mice.

Chemokine and chemokine receptor expression levels reflect the increased neutrophil and reduced monocyte/macrophage infiltrates seen in arthritic feet of CCR2$^{-/-}$ mice compared with WT mice: [a] recruit neutrophils; [b]expressed by neutrophils (Soehnlein and Lindbom, 2010. Nat Rev Immunol. 10(6):427-39; Sadik et al., 2011. Trends Immunol 32(10);452-460); [c] highly expressed in inflammatory macrophages (http://www.immgen.org/databrowser/index.html).

Table S3. Details of the IPA pathways shown in Fig 6A (top graph).

Ingenuity Canonical Pathways	-log(p value)	Molecules
Granulocyte		
Granulocyte Adhesion and Diapedesis	7.52	CXCL3,IL36G,MMP3,CXCL13,CXCR2,PPBP, CCL3L1/CCL3L3,CCL20,IL1B,CXCL6,CCL19
Agranulocyte Adhesion and Diapedesis[a]	7.27	CXCL3,IL36G,MMP3,CXCL13,CXCR2,PPBP, CCL3L1/CCL3L3,CCL20,IL1B,CXCL6,CCL19
IL-17A in autoimmunity		
Role of IL-17A in Psoriasis	7.78	CXCL3,S100A9,CCL20,S100A8,CXCL6
Role of IL-17A in Arthritis	4.41	CXCL3,CCL20,PTGS2,NOS2,CXCL6
Rheumatoid arthritis		
Altered T Cell and B Cell Signaling in Rheumatoid Arthritis	2.55	IL36G,CXCL13,LTB,IL1B
Role of Osteoblasts, Osteoclasts and Chondrocytes in Rheumatoid Arthritis	1.73	IL36G,SFRP2,MMP3,IL1B,Bmpr1b
Role of Macrophages, Fibroblasts and Endothelial Cells in Rheumatoid Arthritis	1.71	IL36G,SFRP2,MMP3,LTB,IL1B,NOS2
Inflammation		
Acute Phase Response Signaling	3.77	IL36G,HP,C1S,RBP2,IL1B,SERPINA3,OSMR
Communication between Innate and Adaptive Immune Cells	3.41	IL36G,HLA-B,CCL3L1/CCL3L3,IL1B,CD8A
Atherosclerosis Signaling	2.83	CCR3,IL36G,MMP3,IL1B,S100A8
Dendritic Cell Maturation	2.13	STAT4,IL36G,HLA-B,LTB,IL1B
Aryl Hydrocarbon Receptor Signaling	1.76	GSTM3,ALDH1A2,NQO1,IL1B
LPS/IL-1 Mediated Inhibition of RXR Function	1.74	IL36G,GSTM3,ALDH1A2,IL1B,SMOX
Other		
LXR/RXR Activation	2.77	IL36G,IL1B,S100A8,PTGS2,NOS2
Retinoate Biosynthesis I	2.73	SDR16C5,ALDH1A2,RBP2
Cholecystokinin/Gastrin-mediated Signaling	2.23	IL36G,IL1B,EPHA4,PTGS2
Xenobiotic Metabolism Signaling	1.85	GSTM3,ALDH1A2,NQO1,IL1B,SMOX,NOS2

Details of the IPA pathways shown in Fig 6A for the 181 genes where "the fold change in $CCR2^{-/-}$ mice" divided by "the fold change in WT mice" was >1.5; i.e. pathways associated with genes more induced in $CCR2^{-/-}$ feet. [a]Same genes as in "Granulocyte Adhesion and Diapedesis". Only pathways with (i) a -log(p value) > 1.5 and (ii) 4 or more molecules are shown

Table S4. Details of the IPA pathways shown in Fig 6A (bottom graph)

Ingenuity Canonical Pathways	-log(p value)	Molecules
Monocyte/macrophage		
Dendritic Cell Maturation	12.4	PLCB2,LEP,NFKBIE,PIK3R5,HLA-DQA1,HLA-DRB1,HLA-DMB, CD83,HLADQB1,FCGR2B,TREM2,FCGR1A,PIK3CG,COL10A1,T NFRSF1B,FCGR3A,HLA-DMA,TYROBP,IL10,MYD88,HLA-B, IL15,CD40,PLCG2,PIK3R6,FCER1G,CD86,STAT2,PIK3CD, IRF8,TNF
Fcγ Receptor-mediated Phagocytosis in Macrophages and Monocytes	7.87	RAC2,PXN,TLN1,FYB,FCGR1A,INPP5D,PLD4,NCF1,PIK3CG,SY K,LYN,HCK,VAV1,FGR,FCGR3A,ACTA1,LCP2,PRKCB
Production of Nitric Oxide and Reactive Oxygen Species in Macrophages	7.74	PTPN6,PPP1R3C,NFKBIE,PIK3R5,PPP1R3A,APOC2,NCF4, SPI1,IRF1,NCF1,ALB,RHOG,PLCG2,PIK3CG,CYBA,NCF2, PIK3R6,CYBB,PIK3CD,SERPINA1,IRF8,TNFRSF1B,TNF,SIRPA, PRKCB
MSP-RON Signaling Pathway	6.24	CSF2RB,ITGB2,ITGAM,CCL12,PIK3CG,PIK3R6,PIK3R5,PIK3CD, CCR2,TNF,ACTA1
IL-12 Signaling and Production in Macrophages	4.36	IL10,MYD88,PIK3R5,APOC2,SPI1,IRF1,ALB,CD40,TGFB1, PIK3CG,PIK3R6,SERPINA1,PIK3CD,IRF8,TNF,PRKCB
Pattern recognition		
Role of Pattern Recognition Receptors in Recognition of Bacteria and Viruses	8.01	Tlr11,NLRP3,IL10,MYD88,PIK3R5,C1QC,C1QA,C1QB,IRF7,PLC G2,PIK3CG,SYK,TLR1,PIK3R6,PIK3CD,TNF,C3AR1,PRKCB
Autoimmunity		
Altered T Cell and B Cell Signaling in Rheumatoid Arthritis	7.96	HLA-DMA,Tlr11,CD79B,IL10,IL15,HLA-DQA1,HLA-DRB1,HLA-DMB,HLADQB1,CD79A,CD40,TGFB1,TLR1,FCER1G,CD86, Tlr13,TNF
Type I Diabetes Mellitus Signaling	7.88	HLA-DMA,SOCS3,CASP3,MYD88,NFKBIE,HLA-B,HLA-DQA1, APAF1,HLA-DRB1,HLA-DMB,HLA-DQB1,IRF1,PRF1,FCER1G, CD86,BID,TNFRSF1B,HLA-F,TNF
Autoimmune Thyroid Disease Signaling	6.88	HLA-DMA,PRF1,CD40,IL10,HLA-B,HLA-DQA1,FCER1G,HLA-DRB1,CD86,HLA-DMB,HLA-DQB1,HLA-F
Systemic Lupus Erythematosus Signaling	4.07	PTPN6,CD79B,IL10,HLA-B,PIK3R5,FCGR2B,FCGR1A, CD79A,INPP5D,CD40,PIK3CG,CD72,PLCG2,PIK3R6,FCER1G, LYN,CD86,PIK3CD,HLA-F,TNF,FCGR3A
B and T cells		
Role of NFAT in Regulation of the Immune Response	9.55	PLCB2,NFKBIE,PIK3R5,HLA-DQA1,HLA-DRB1,HLA-DMB, HLA-DQB1,FCGR2B,FCGR1A,CD79A,GNB4,PIK3CG, FCGR3A,HLA-DMA,CD79B,ITPR1,GNAI2,BTK,SYK,PLCG2, PIK3R6,FCER1G,LYN,CD86,MEF2C,PIK3CD,LCP2
T Helper Cell Differentiation	8.5	HLA-DMA,IL2RG,IL10,HLA-DQA1,HLA-DRB1,HLA-DMB,HLA-DQB1,CD40,TGFB1,FCER1G,IL10RB,IL10RA,CD86,IL2RA, TNFRSF1B,TNF
FcγRIIB Signaling in B Lymphocytes	8.36	BTK,CD79B,SYK,PIK3CG,PLCG2,LYN,PIK3R6,PIK3R5,PIK3CD, DOK1,FCGR2B,CD79A,INPP5D
iCOS-iCOSL Signaling in T Helper Cells	7.68	HLA-DMA,IL2RG,NFKBIE,CSK,HLA-DQA1,PIK3R5,HLA-DRB1,HLA-DMB,HLA-DQB1,ITPR1,INPP5D,CD40,PIK3CG, PIK3R6,FCER1G,VAV1,IL2RA,PIK3CD,LCP2
Antigen Presentation Pathway	7.3	PSMB9,NLRC5,HLA-DMA,HLA-B,HLA-DQA1,CIITA,HLA-DRB1,HLA-DMB,CD74,HLA-F,TAP1
B Cell Receptor Signaling	6.87	RAC2,PTPN6,CD79B,POU2F2,NFKBIE,CSK,PIK3R5,FCGR2B, CD79A,INPP5D,BTK,SYNJ1,DAPP1,SYK,PIK3CG,PLCG2,PIK3R6,LYN,VAV1,PIK3AP1,PIK3CD,PRKCB
PI3K Signaling in B Lymphocytes	6.45	PLCB2,CD79B,NFKBIE,ITPR1,FCGR2B,INPP5D,CD79A,BTK,DAPP1,CD40,CD180,SYK,PIK3CG,PLCG2,LYN,VAV1,PIK3AP1, PIK3CD,PRKCB
CD28 Signaling in T Helper Cells	6.41	HLA-DMA,PTPN6,NFKBIE,CSK,HLA-DQA1,PIK3R5,HLA-DRB1,HLA-DMB,HLA-DQB1,ITPR1,PIK3CG,SYK,PIK3R6, FCER1G,CD86,VAV1,PIK3CD,LCP2
Graft-versus-Host Disease Signaling	6.14	HLA-DMA,PRF1,HLA-B,HLA-DQA1,FCER1G,HLA-DRB1,CD86, HLA-DMB,HLA-DQB1,HLA-F,TNF
Tec Kinase Signaling	5.86	PIK3R5,ITGA5,BTK,GNAI2,GNB4,RHOG,PLCG2,PIK3CG,LYN, FCER1G,PIK3R6,HCK,VAV1,STAT2,PIK3CD,TNF,FGR,ACTA1, ITGA4,PRKCB
B Cell Development	5.85	HLA-DMA,CD79B,CD40,HLA-DQA1,HLA-DRB1,CD86, HLA-DMB, HLA-DQB1,CD79A
Allograft Rejection Signaling	5.72	HLA-DMA,PRF1,H2-T24,CD40,IL10,HLA-B,FCER1G,HLA-DQA1, CD86,HLA-DRB1,HLA-DMB,HLA-DQB1,HLA-F,TNF
IL-4 Signaling	5.49	HLA-DMA,IL2RG,PTPN6,HLA-DQA1,PIK3R5,HLA-DRB1, HLA-DMB,HLA-DQB1,INPP5D,SYNJ1,PIK3CG,PIK3R6,PIK3CD
CTLA4 Signaling in Cytotoxic T Lymphocytes	5.24	HLA-DMA,PTPN6,HLA-DQA1,PIK3R5,HLA-DRB1,HLA-DMB, HLA-DQB1,PIK3CG,SYK,PIK3R6,FCER1G,CD86,PIK3CD,LCP2

Cytotoxic T Lymphocyte-mediated Apoptosis of Target Cells	5.17	HLA-DMA,PRF1,H2-T24,CASP3,HLA-B,FCER1G,APAF1, HLA-DQA1,BID,HLA-DRB1,HLA-DMB,HLA-DQB1,HLA-F
PKCθ Signaling in T Lymphocytes	5.13	HLA-DMA,RAC2,NFKBIE,HLA-DQA1,PIK3R5,HLA-DRB1,HLA-DMB,HLA-DQB1,PLCG2,PIK3CG,PIK3R6,FCER1G,CD86, VAV1,PIK3CD,LCP2
Communication between Innate and Adaptive Immune Cells	4.61	Tlr11,CD40,IL10,TLR1,IL15,HLA-B,FCER1G,CD86,Tlr13, HLA-DRB1,CD83,HLA-F,TNF
NK cells		
Natural Killer Cell Signaling	9.02	RAC2,PTPN6,Klra4 (includes others),LAIR1,TYROBP,PIK3R5, SH3BP2,INPP5D,SYNJ1,SYK,KLRC4-KLRK1/KLRK1,PLCG2, PIK3CG,PIK3R6,FCER1G,VAV1,PIK3CD,SH2D1B,LCP2,FCGR3A,PRKCB
Crosstalk between Dendritic Cells and Natural Killer Cells	8.4	IL2RG,TYROBP,CD69,IL15,HLA-B,HLA-DRB1,TLN1,CD83, TREM2,CSF2RB,PRF1,CD40,KLRC4-KLRK1/KLRK1, CD86,TNFRSF1B,HLA-F,TNF,ACTA1
Muscle		
Actin Cytoskeleton Signaling	4.79	RAC2,PXN,MYLPF,CSK,ACTN2,PIK3R5,ITGA5,MYH7,TLN1, IQGAP2,MYH2,PIK3CG,PIK3R6,MYH3,VAV1,PIK3CD,NCKAP1L, TMSB10/TMSB4X,PIP4K2A,ACTA1,MYH1,ITGA4
Calcium Signaling	4.74	RAP2B,TNNT1,CHRNA1,ATP2B1,TNNC2,TNNC1,TNNT2,TRDN, CHRNB1,Tpm1,MYH7,ITPR1,MYH2,MYH3,RYR1,MEF2C,TNNI1, SLC8A1,ACTA1,MYH1
Paxillin Signaling	4.76	ITGB2,PXN,ITGAM,PIK3CG,ACTN2,CSK,PIK3R6,PIK3R5,ITGA5, PIK3CD,TLN1,ITGA7,ACTA1,ITGA4
Inflammation		
TREM1 Signaling	9.57	Tlr11,TYROBP,IL10,MYD88,LAT2,ITGA5,CD83,FCGR2B,CD40, CCL12,CCL7,PLCG2,TLR1,CD86,Tlr13,TNF
Leukocyte Extravasation Signaling	8.81	RAC2,MMP14,PIK3R5,MMP25,PIK3CG,CYBA,CYBB,ACTA1, ITGA4,PXN,CXCR4,ACTN2,ARHGAP4,ITGA5,NCF4,SELPLG, GNAI2,BTK,ITGB2,NCF1,WIPF1,ITGAM,PLCG2,NCF2,PIK3R6, VAV1,PIK3CD,PRKCB
Atherosclerosis Signaling	6.36	ALOX12B,MSR1,CXCR4,APOC2,PLA2G7,F3,TNFRSF14, SELPLG,ITGB2,ALB,CD40,CCL12,TGFB1,COL10A1,SERPINA1, CCR2,TNF,ITGA4
Fc Epsilon RI Signaling	5.38	RAC2,PIK3R5,INPP5D,BTK,SYNJ1,PLCG2,PIK3CG,SYK,LYN, FCER1G,PIK3R6,VAV1,PIK3CD,TNF,LCP2,PRKCB
Integrin Signaling	4.96	RAP2B,RAC2,PXN,ARHGAP26,ACTN2,PIK3R5,ITGA5,TLN1, PARVB,ITGB2,WIPF1,RHOG,ITGAM,PIK3CG,PLCG2,PIK3R6, ZYX,PIK3CD,ITGA7,ACTA1,ITGA4
Role of JAK1 and JAK3 in γc Cytokine Signaling	4.13	SOCS3,IL2RG,FES,PIK3CG,SYK,IL15,PIK3R6,PIK3R5,IL2RA, PIK3CD
NF-κB Activation by Viruses	4.09	ITGB2,CCR5,PIK3CG,NFKBIE,PIK3R6,PIK3R5,ITGA5,PIK3CD, TNFRSF14,ITGA4,PRKCB
Other		
Virus Entry via Endocytic Pathways	4.4	RAC2,HLA-B,PIK3R5,ITGA5,ITGB2,FLNC,PLCG2,PIK3CG, PIK3R6,PIK3CD,ACTA1,PRKCB,ITGA4
Induction of Apoptosis by HIV1	4.32	CASP3,CXCR4,NFKBIE,APAF1,BID,BAX,TNFRSF1B,NAIP,TNF, BAK1
LXR/RXR Activation	4.26	TTR,MSR1,VTN,AHSG,NR1H3,APOC2,ABCG1,ALB,CCL12,CCL7,SERPINA1,PLTP,GC,TNFRSF1B,TNF
Role of Tissue Factor in Cancer	4.21	F10,ARRB2,CASP3,PIK3CG,LYN,HCK,PIK3R6,PIK3R5,PLAUR, FGB,PIK3CD,RPS6KA2,F3,FGR
ILK Signaling	4.2	PXN,FBLIM1,CASP3,ACTN2,PIK3R5,MYH7,PARVB,ITGB2, RHOG,MYH2,FLNC,PIK3CG,PIK3R6,MYH3,PIK3CD,TMSB10/TMSB4X,TNF,ACTA1,MYH1
Phospholipase C Signaling	4.1	PLCB2,CD79B,MYLPF,ITGA5,ITPR1,FCGR2B,CD79A,BTK, PLD4,TGM2,GNB4,RHOG,SYK,PLCG2,FCER1G,LYN,MEF2C,ARHGEF2,ADCY7,LCP2,ITGA4,PRKCB

Details of the IPA pathways shown in Fig 6A for the 920 gene where "the fold change in WT mice" divided by "the fold change in $CCR2^{-/-}$ mice" was >1.5; i.e. pathways associated with the genes less induced in $CCR2^{-/-}$ feet. Only pathways with a -log(p value) > 4 are shown.

Table S5. Ingenuity upstream regulator analysis of the 181 gene more induced in CCR2$^{-/-}$ mice

Upstream Regulator	Activa-tion z-score	p-value of overlap	Target molecules in dataset
STAT3	1.774	4.29E-14	ADM,ALAS2,ANGPTL4,CCL20,CHI3L1,CXCL13,CXCR2,HLA-B,HP,Ifi204 (includes others),IFIT1,IFIT1B,IFITM1,IL1B,MMP3,NOS2, PTGS2,SERPINA3,SERPINB1,SERPINB3,SERPINB4
MKL2	-2.646	3.16E-13	CXCL6,ELANE,HLA-B,IFITM1,LCN2,LTF,Ngp,PGLYRP1,PPBP, S100A8,S100A9,SERPINB10
MKL1	-2.646	3.52E-12	CXCL6,ELANE,HLA-B,IFITM1,LCN2,LTF,Ngp,PGLYRP1, PPBP,S100A8,S100A9,SERPINB10
RELB	0.097	1.01E-10	CCL19,CCL20,CCR3,CXCL13,IL1B,LTB,NOS2,PTGS2,STAT4
NFKBIA	2.068	2.78E-10	CCL20,CCL3L1/CCL3L3,CCR3,CXCL3,CXCL6,CYP7B1,HLA-B,Ifi204 (includes others),IFIT1B,IL1B,LCN2,MMP3,NOS2,PTGS2, PTX3,S100A8,S100A9,TFRC
STAT1	1.526	6.64E-10	ALAS2,CCL19,CCRL2,Ifi204 (includes others),IFIT1,IFIT1B,IFITM1,IL1B, NOS2,PTGS2,Retnla,SERPINA3,SERPINB3,SERPINB4
RELA	2.748	3.70E-09	CCL19,CCL20,CHI3L1,CXCL3,CXCL6,CYP17A1,EHF,HLA-B,IFIT1B, IL1B,LTB,NOS2,PTGS2,PTX3,TREM1
CEBPE	2.199	3.01E-08	ELANE,LCN2,LTF,Ngp,PTGS2,Retnla,RETNLB
NFKB1	2.062	1.29E-07	CCL19,CCL20,CCL3L1/CCL3L3,CXCL3,EHF,IFIT1B,IL1B,LTB,NOS2, PTGS2,PTX3
HMGB1	2.543	1.31E-07	CCL20,CXCL3,IFIT1,IL1B,MMP3,NOS2,PTGS2

COLOUR CODING

Muscle and vasculature associated upstream regulators
(Note these have negative activation z scores suggesting down regulation)

NF-κB associated upstream regulators (often associated with inflammation)

Interferon associated upstream regulators

Transcription factor associated with granulocyte differentiation
(Lekstrom-Himes, 2001. Stem Cells. 19(2):125-33)

Inflammatory mediator associated with RA
(Chen et al. 2013. Rheumatology (Oxford). 2013 Apr 12. [Epub ahead of print])

Ingenuity upstream regulator analysis of the 181 genes where "the fold change in CCR2$^{-/-}$ mice" divided by "the fold change in WT mice" was >1.5; i.e. upstream regulators more active in CCR2$^{-/-}$ feet. Activation z scores and significance (p values) are shown for upstream regulators where p<1.4E-7 (Direct only). Upstream regulators are colour coded according to the legend (COLOUR CODING). Note chemokine ligands and receptors are well represented in "Target molecules in dataset".

Table S6. Ingenuity upstream regulator analysis of the 920 genes less induced in CCR2$^{-/-}$ mice

Upstream Regulator	Activation z-score	p-value of overlap	Target molecules in dataset
STAT1	5.849	1.40E-19	APOBEC3B,APOC2,BATF2,BAX,BCL2L11,CASP3,CCL12,CD274,CD40,CD86,CH25H, Chi3l3/Chi3l4,CIITA,CYP2E1,FAM26F,FCER1G,FCGR1A,FGL2,GBP1,GBP5,Gbp6 (includes others),HLA-DRB1,Ifi204 (includes others),Ifi47,IFIT2,IL10,IL15, IRF1,IRF5,IRF7,IRF8,IRGM,KLRC4-KLRK1/KLRK1,Neurl3,PRF1,PSMB10, PSMB9,Rnf213,Slfn1,SLFN13,Slfn2,SOCS3,SP110,STAT2,TAP1
SPI1	3.547	1.45E-19	BAX,C1QC,CD180,CD68,CD72,CD79A,CD79B,CIITA,CSF1R,CSF2RB,CTSS,CYBB,E MR1,FCER1G,FCGR2B,FES,HK3,IL10,IL2RA,ITGA4,ITGA5,ITGAM,ITGB2,Klra4 (includes others),Lyz1/Lyz2,MEF2C,NCF1,NCF2,NCF4,P2RY10,PIK3CG, PTPN6,SPI1,TFEC,TNF,TNFRSF14,VAV1
MYOD1	3.654	2.42E-13	ACTA1,ADCY7,ASS1,BIN1,CKM,CRYAB,DES,ENO3,GMFG,Ifi204 (includes others),ITGA7,MEF2C,MUSK,MYF6,MYH3,MYH7,MYLPF,MYOD1,PFKM,PLEKHO1,R PS6KA2,RYR1,SCN4A,SGCA,SLC2A4,TGFB1,TNNC1,TNNC2,TNNI1,TNNT1,TNNT2
IRF8[a]	2.854	1.25E-12	ASAH1,BAX,CASP3,CCR2,CD40,CD83,CD86,CIITA,CSF1R,CTSS,CXCL16,CYBB, EMR1,GBP1,IFIT2,IL15,IRF8,ITGAM,MSR1,PRDM1,TNF,TYROBP
IRF3	5.077	1.63E-12	APOBEC3B,BC147527,CCL12,CD69,CD86,DHX58,FAM26F,FCGR1A,GBP1,GBP5,HL A-F,Ifi204 (includes others),Ifi47,IFIT2,IL15,IRF5,IRF7,IRGM,Irgm2, NRAP,PARP14,PLAC8,PLAG1,PLCG2,Slfn1,STAT2,TAP1,TNF
IRF7	5.210	7.19E-12	BC147527,CD40,CD69,DHX58,FAM26F,FCGR1A,GBP1,GBP4,GBP5,Ifi204 (includes others),Ifi47,IFIT2,IL15,IRF1,IRF7,IRF8,IRGM,Irgm2,ITGAM,NRAP,PARP14,PLAC8,PL SCR1,PSMB10,PSMB9,Slfn1,STAT2,TAP1
STAT3	4.296	8.59E-12	ADIPOQ,AHSG,BAK1,BCL2L11,CCL12,CCR5,CD40,CD74,CIITA,FCER1G, FCGR1A,FGB,HLA-B,HLA-DMA,HLA-DQA1,HLA-DRB1,Ifi204 (includes others),IFI30,IL10,IL2RA,IRF1,IRF7,ITGAM,ITGB2,Lyz1/Lyz2,MYD88,MYH7,PLAUR, PLSCR1,PRDM1,PSMB9,PTPN6,Slfn1,SLFN13,Slfn2,SOCS3,SP110,TAP1,TGFB1, TNF,TNFRSF1B,Tnfsf9,TRIM14,ZFP36
MEF2C	2.722	1.32E-11	ABRA,ACTA1,CKB,CKM,CPT1B,DES,MEF2C,MYH1,MYH7,MYOD1,MYOT,MYOZ1, SLC2A4,SLC8A1,SMYD1,TNNC1,TNNI1,TNNT2
IRF1	3.641	3.56E-11	ADAM8,BAK1,BAX,CASP3,CCL12,CD40,CIITA,CTSS,CXCL16,CYBB,FGL2,IFIT2,IL10 IL15,IL18BP,IRF1,IRF5,IRF7,PSMB10,PSMB9,SPI1,STAT2,TAP1,TGFB1,TNF

COLOUR CODING

.	**Interferon associated upstream regulators**
.	**Myeloid cell development**
	Muscle and vasculature associated upstream regulators

Ingenuity upstream regulator analysis of the 920 genes where "the fold change in WT mice" divided by "the fold change in CCR2$^{-/-}$ mice" was >1.5; i.e. upstream regulators less active in CCR2$^{-/-}$ feet. Activation z scores and significance (p values) are shown for upstream regulators where p<1E-10 (Direct only). Upstream regulators are colour coded according to the legend (COLOUR CODING). [a]IRF8 is involved in myeloid differentiation into monocytes and is less strongly expressed in neutrophils, with IRF8 deficiency shown to cause major neutrophil infiltration (Rocca et al., 2013. Plos One 8(5): e62751).