

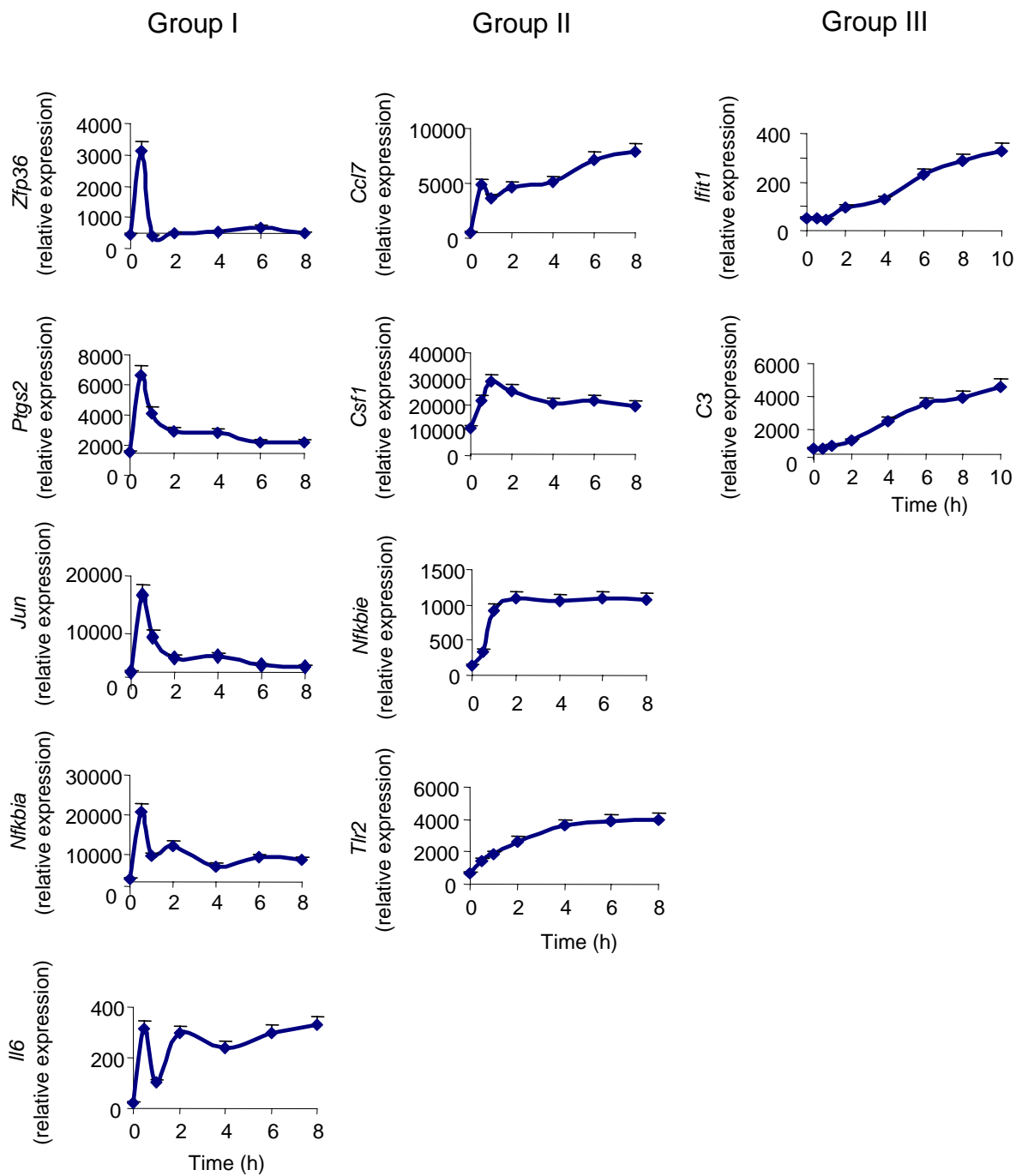
**Supplementary Figure 1.** Additional examples of genes within the three groups of TNF-activated genes. 3T3 fibroblasts were stimulated with TNF (10 ng/ml). At indicated times after addition of TNF, mRNA transcripts encoding genes in each group were measured by RT-qPCR and normalized to *Rpl32* as described in **Methods**. The results are shown as mean  $\pm$  s.d. of triplicate samples and representative of four independent experiments with similar results.

**Supplementary Figure 2.** Kinetic patterns of TNF-induced gene expression in human dermal primary fibroblasts. Cells were exposed to continual TNF (10 ng/ml) stimulation (TNF; solid diamond), or to 6 hours of TNF followed by medium alone (TNF removal; open square). At indicated times after initial addition of TNF, mRNA transcripts encoding indicated genes were measured by RT-qPCR and normalized to *Rpl32* as described in **Methods**. However, *Rpl32* in these cells was expressed in approximately 100-fold lower amounts than in 3T3 cells. Note that only one time point was taken after removal of TNF so the kinetics of the drop in mRNA concentration are not evident. The results are shown as mean  $\pm$  s.d. of triplicate samples from one representative experiment. Numbers in parentheses show the half life of the mRNA, determined by addition to ActD at 6h following TNF stimulation, which is similar to that determined at 0.5h of TNF stimulation for most genes (data not shown)

**Supplementary Figure 3.** Effect of TNF concentration on gene expression patterns. 3T3 fibroblasts were treated with 10 ng/ml (solid diamond) or 1 ng/ml (open square) TNF and mRNA transcripts encoding indicated genes were measured by qRT-PCR. The data are mean  $\pm$  s.d. of triplicate samples and representative of three independent experiments with similar results.

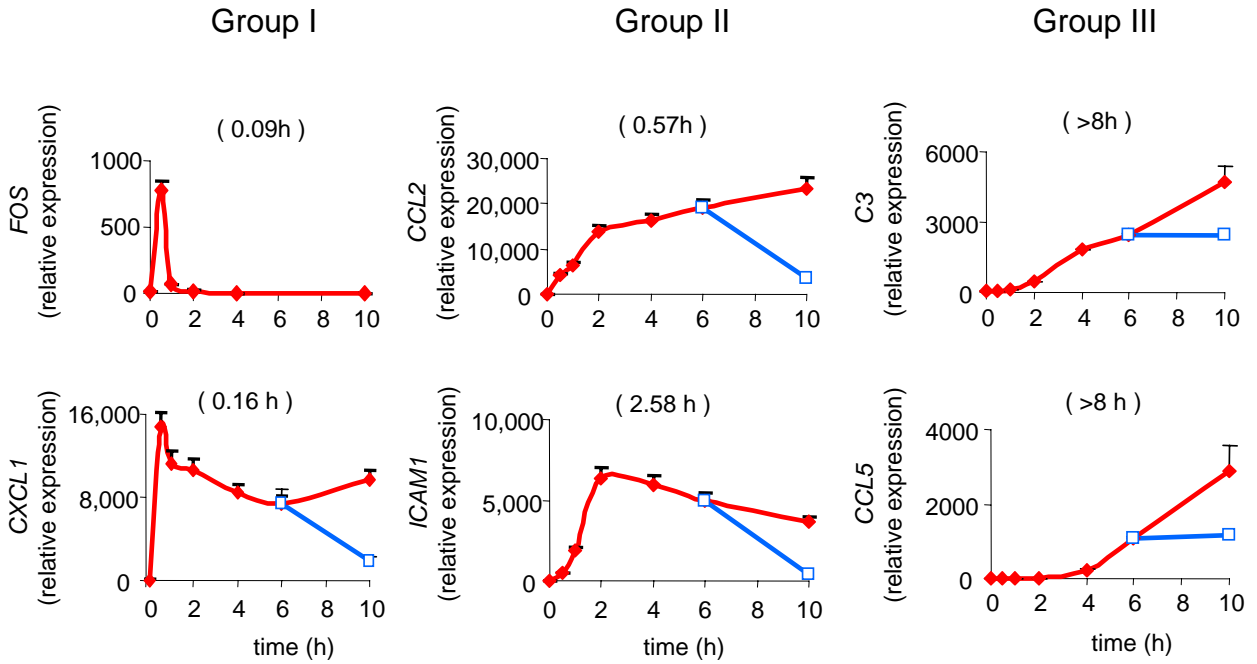
**Supplementary Figure 4.** Schematic of GFP transgene constructs with various promoters and 3'UTRs.

# Supplementary Figure 1



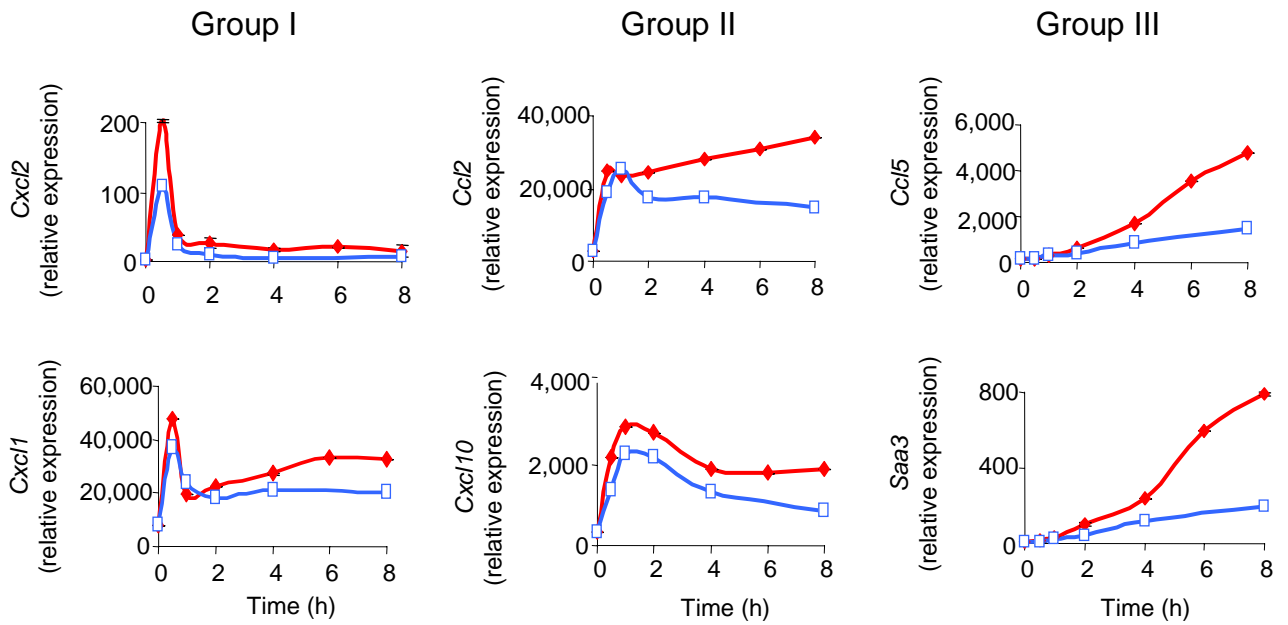
## Supplementary Figure 2.

◆ TNF  
□ TNF removal



# Supplementary Fig. 3

◆ TNF 10 ng/ml  
□ TNF 1 ng/ml





## Supplementary Table 1. List of the three groups of genes

Group I genes, (fold $\geq$ 2.0, p<0.01)

Gene name	Alias, or sequence description	Accession #	relative basal level	TNF fold induction		
				0.5h	2h	12h
<b>Blood vessel regulators</b>						
<i>Ptgs2</i>	Cox-2	M94967	1.59	2.01	-1.46	-2.94
<i>Edn1</i>	endothelin 1	D43775	0.30	3.98	-1.19	1.02
<i>Ereg</i>	epiregulin	NM_007950	1.72	2.12	1.32	-1.08
<b>Cell death, survival</b>						
<i>Gadd45b</i>	growth arrest and DNA-damage-inducible 45 beta	AK010420	1.80	3.16	2.36	1.69
<i>Ier2</i>	immediate early response 2	NM_010499	1.20	2.35	-1.18	-1.82
<b>Chemokine or cytokine</b>						
<i>Cxcl1</i>	Gro1	NM_008176	0.70	28.08	11.98	6.20
<i>Cxcl2</i>	Scyb2	NM_009140	0.02	44.86	9.09	3.59
<i>Il6</i>	interleukin 6	NM_031168	0.31	14.20	8.40	6.30
<i>Lif</i>	leukemia inhibitory factor	AF065917	0.17	5.69	3.41	2.06
<b>mRNA stability</b>						
<i>Btg2</i>	B-cell translocation gene 2	NM_007570	0.24	3.07	1.22	-2.19
<i>Zfp36</i>	TTP	X14678	0.49	6.65	1.84	-1.04
<b>Signal transduction</b>						
<i>Tnfrsf25</i>	A20	NM_009397	1.41	9.71	5.99	2.51
<i>Ier3</i>	immediate early response 3	NM_133662.1	5.34	4.08	2.49	1.93
<i>Sqstm1</i>	sequestosome 1	NM_011018	0.36	2.04	1.48	-1.00
Transcription factors or modulators						
<i>Atf3</i>	activating transcription factor 3	NM_007498	0.38	2.04	1.62	1.95
<i>Axud1</i>	Axud1	BG070296	0.31	2.45	-1.66	-4.62
<i>c-Jun</i>	Jun oncogene	NM_010591	2.69	2.00	1.01	-1.10
<i>c-Fos</i>	FBJ osteosarcoma oncogene	AV026617	0.49	4.71	-1.92	-6.42
<i>Irf1</i>	interferon-related developmental regulator 1	BB531645	0.15	3.51	1.60	-1.14
<i>Nfkb1a</i>	I $\kappa$ Ba	NM_010907	1.42	10.03	7.47	3.63
<i>Nfkb2</i>	I $\kappa$ Bzeta	NM_030612	0.62	5.71	3.63	2.00
<i>Irf1</i>	interferon regulatory factor 1	NM_008390	0.77	7.69	5.41	2.86
<i>Junb</i>	Jun-B oncogene	NM_008416	1.44	2.14	1.12	-1.83
<i>Tieg</i>	TGFB inducible early growth response	NM_013692	1.91	2.26	1.18	-1.86
<b>Others</b>						
<i>Slc25a25</i>	solute carrier family 25	BC019978.1	0.28	2.35	1.15	-1.29

## Supplementary Table 1. Continued

### Group II genes

(only those fold>=3, p<0.01 are shown)

Gene Name	Sequence Description	Accession #	Relative basal level	TNF fold induction		
				0.5h	2h	12h
<b>Anti-viral and immune response</b>						
<i>Ifi47</i>	interferon gamma inducible protein, 47 kDa	NM_008330	0.02	3.50	37.17	20.91
<i>Tgtp</i>	T-cell specific GTPase	NM_011579	0.03	1.36	24.75	9.73
<b>Blood vessel regulators</b>						
<i>Gch</i>	GTP cyclohydrolase 1	NM_008102	0.16	1.24	6.11	5.67
<b>Cell adhesion and migration</b>						
<i>Icam1</i>	intercellular adhesion molecule 1	BC008626	0.03	3.09	30.01	22.70
<i>Vcam1</i>	vascular cell adhesion molecule 1	BB250384	1.29	2.34	5.30	4.36
<b>Cell death and survival</b>						
<i>Birc2</i>	baculoviral IAP repeat-containing 2	NM_007464	0.35	1.53	5.25	6.27
<i>Tnfrsf6</i>	Fas	NM_007987	0.50	1.11	3.54	2.66
<b>Chemokines or cytokines</b>						
<i>Ccl2</i>	MCP-1, Scya2	AF065933	0.74	9.10	17.44	19.62
<i>Ccl20</i>	MIP-3a, Scya20	AF099052	0.02	100	100	100
<i>Ccl7</i>	MCP-3, Scya7	AF128193	0.34	5.90	10.53	14.09
<i>Cx3cl1</i>	Scyd1, Fractalkine	AF010586	0.88	1.90	4.82	2.62
<i>Cxcl10</i>	IP-10, Scyb10	NM_021274	0.26	10.91	31.91	17.03
<i>Cxcl5</i>	ENA-78, Scyb5	NM_009141	2.87	2.93	3.36	2.72
<i>CSF1</i>	colony stimulating factor 1 (macrophage)	NM_007778	2.53	1.90	3.00	2.10
<i>Ch25h</i>	cholesterol 25-hydroxylase	NM_009890	0.09	7.87	15.15	13.50
<b>Signal transduction</b>						
<i>Ifngr2</i>	Interferon gamma receptor 2	BF537076	0.25	1.26	3.01	3.04
<i>p21</i>	cyclin-dependent kinase inhibitor 1A (P21)	NM_007669	0.21	2.46	4.26	1.19
<i>Ripk2</i>	RIP2,	NM_138952.1	0.44	1.62	8.22	2.41
<i>S1pr3</i>	sphingosine-1-phosphate receptor 3	AV238324	0.54	1.37	3.36	3.11
<i>Tlr2</i>	toll-like receptor 2	NM_011905	0.66	2.67	7.30	4.79
<i>Cebpd</i>	CEBP beta	BB831146	1.78	1.22	4.36	1.93
<i>Nfkbie</i>	IkBe	BB820441	0.15	3.03	28.35	20.00
<i>Relb</i>		NM_009046	0.09	2.78	9.40	8.05
<i>Tnfaip2</i>	B94	BB233088	0.72	1.53	3.83	1.95
<i>Npal1</i>	RIKEN cDNA 3830408G10 gene	AK014427	0.23	1.42	3.14	1.24
<i>Rnd1</i>		BE852181	0.99	2.67	3.16	3.25
<i>Tmem140</i>	transmembrane protein 140	BC020080.1	0.44	1.04	3.32	2.36
<i>Stx11</i>	RIKEN cDNA 5830405C08 gene	AK017897	0.52	1.82	4.71	2.23

## Supplementary Table 1. Continued

Group III genes  
(only those fold  $\geq 3$ ,  $p < 0.01$  are shown)

Gene Name	Alias, or Sequence description	Accession #	relative basal level	TNF fold induction		
				0.5h	2h	12h
<b>Antigen presentation</b>						
<i>H2-D1</i>	histocompatibility 2, D region locus 1	M86502	1.29	1.20	1.75	4.02
<i>H2-K</i>	histocompatibility 2, K region	BC011306	0.37	1.02	2.56	29.68
<i>H2-L</i>	histocompatibility 2, L region	M34962	0.23	1.30	1.70	3.66
<i>Ii</i>	Ia-associated invariant chain	BC003476	0.16	1.05	2.49	7.31
<i>Psmb10</i>	proteasome subunit, beta type 10	NM_013640	0.81	1.13	2.52	5.02
<i>Tapbp</i>	TAP binding protein	AF043943	4.55	1.08	1.74	3.11
<b>Anti-viral and immune response</b>						
<i>C3</i>	complement component 3	K02782	0.07	2.65	6.14	55.51
<i>Gbp2</i>	guanylate nucleotide binding protein 2	NM_010260	0.06	1.26	7.69	36.09
<i>Ifit1</i>	interferon-induced protein with tetratricopeptide repeats 1	NM_008331	0.10	-1.00	2.40	5.84
<b>Cell adhesion and migration</b>						
<i>Casp4</i>	caspase 11	NM_007609	0.04	1.86	7.81	18.72
<i>Cdsn</i>	corneodesmosin	BM231053	0.61	1.08	1.65	5.89
<b>Cell death and survival</b>						
<i>Mt2</i>	metallothionein 2	AA796766	1.35	1.58	3.14	5.24
<i>Sod3</i>	superoxide dismutase 3, extracellular	NM_011435	0.27	1.13	1.67	3.04
<i>Serpina3g</i>	serine protease inhibitor 2-1,	BC002065	0.06	1.11	5.65	19.18
<i>Ubd</i>	ubiquitin D	NM_023137	0.09	-1.08	1.71	9.14
<b>Chemokines and cytokines</b>						
<i>Ccl5</i>	RANTES, Sca5	NM_013653	0.02	-1.74	33.6	100
<i>Ccl9</i>	small inducible cytokine A9	AF128196	0.24	1.41	2.47	4.11
<i>Lcn2</i>	lipocalin 2	X14607	0.16	-1.42	1.01	4.81
<b>Signaling transduction</b>						
<i>Il13ra1</i>	IL13 receptor a1	BI081033	0.19	1.02	2.17	3.44
<i>Il1rl1</i>	interleukin 1 receptor-like 1	NM_010743	0.88	1.25	2.35	6.13
<i>Naf1</i>	Nef-associated factor 1, Abin-1	AJ242777	1.00	1.01	2.41	4.30
<b>Tissue remodeling and wound healing</b>						
<i>Enpp2</i>	ectonucleotide pyrophosphatase/phosphodiesterase 2	BC003264	0.30	1.08	1.74	13.07
<i>Mmp13</i>	matrix metalloproteinase 13	NM_008607	0.15	1.53	4.86	16.48
<i>Mmp3</i>	matrix metalloproteinase 3	NM_010809	1.37	1.20	1.86	6.57
<i>Serpinf1</i>	, PEDF	NM_011340	2.18	1.09	1.26	4.08
<i>Ppicap</i>	peptidylprolyl isomerase C-associated protein	NM_011150	0.20	1.17	1.20	3.07
<b>Others (unknown function)</b>						
1110004C05Rik	RIKEN cDNA 1110004C05 gene	BC010291	3.38	1.24	1.97	3.77
<i>Abca1</i>	ATP-binding cassette, sub-family A1	BB144704	0.07	2.07	2.30	7.08
<i>Al561871</i>	expressed sequence Al561871	BI143915	0.17	-1.20	1.51	5.08
<i>Al882074</i>	expressed sequence Al882074	BB730912	0.09	1.29	3.32	5.91
<i>Arts1</i>	ESTs	AV287655	0.10	1.16	1.83	3.59
<i>Lhfp12</i>	lipoma HMGIC fusion partner-like 2	BG917242	0.02	1.82	6.77	23.29



LOC677168	cDNA, RIKEN clone:2900034J12,	AK019325	0.25	-1.06	2.32	5.91
AW049765		BC026642.1	0.10	-1.41	2.57	21.95
<i>Prnd</i>	prion protein dublet	AF165166	0.02	1.18	3.38	28.41
<i>U90926</i>	cDNA sequence U90926	NM_020562	0.38	-1.16	1.03	3.72

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## Supplementary Table 2. Stability of mRNA transcripts measured using two different approaches

mRNA stability (half life, h, mean  $\pm$  s.d., n=3 )

Genes		6h TNF pretreatment	
		TNF $\alpha$ removal	ActD treatment
Group II	<i>Ccl20</i>	1.30 $\pm$ 0.12	3.33 $\pm$ 1.02
	<i>Ccl2</i>	1.38 $\pm$ 0.09	0.82 $\pm$ 0.01
	<i>Ccl7</i>	1.56 $\pm$ 0.21	1.32 $\pm$ 0.06
	<i>Tlr2</i>	1.93 $\pm$ 0.25	1.27 $\pm$ 0.32
	<i>Nfkbie</i>	1.69 $\pm$ 0.07	1.90 $\pm$ 0.13
	<i>Cxcl10</i>	1.87 $\pm$ 0.05	1.92 $\pm$ 0.05
	<i>Icam1</i>	1.66 $\pm$ 0.20	2.30 $\pm$ 0.56
	<i>CSF1</i>	5.33 $\pm$ 2.51	2.60 $\pm$ 0.43
	<i>Mmp10</i>	10.5 $\pm$ 18.0	stable
Group III	<i>Ifit1</i>	stable	1.97 $\pm$ 1.22
	<i>Stat1</i>	9.76 $\pm$ 18.5	11.7 $\pm$ 12.7
	<i>C3</i>	stable	stable
	<i>Ccl5</i>	stable	stable
	<i>Mmp13</i>	stable	stable
	<i>Mmp3</i>	stable	stable
	<i>Saa3</i>	stable	stable

### Supplementary Table 3. Sequences of qPCR primers

name	alias	species	primer name	sequence
<i>Tnfrsf3</i>	A20	mouse	A20_(1)_F	GAACAGCGATCAGGCCAGG
			A20_(1)_R	GGACAGTTGGGTGTCTCACATT
<i>actb</i>	$\beta$ -actin	mouse	actin_F	AGGTGTGCACCTTTTATTGGTCTCAA
			actin_R	TGTATGAAGGTTTGGTCTCCCT
<i>C3</i>	Complement 3	mouse	C3_(2)_F	CTGAGAAGCGTCTCCATCAAG
			C3_(2)_R	ACGGGCAGTAGGTTGTTGTC
<i>Ccl2</i>	MCP-1	mouse	Ccl2_F	CTGAAGACCTTAGGGCAGAT
			Ccl2_R	AAGGAATGGGTCCAGACATAC
<i>Ccl20</i>	MIP-3 $\alpha$	mouse	CCL20_F	GCCTCTCGTACATACAGACGC
			CCL20_R	CCAGTTCTGCTTTGGATCAGC
<i>Ccl5</i>	Rantes	mouse	Ccl5_(1)_F	GCTGCTTTGCCTACCTCTCC
			Ccl5_(1)_F	TCGAGTGACAAACACGACTGC
<i>Ccl7</i>	MCP-3	mouse	CCL7_(1)_F	GCTGCTTTCAGCATCCAAGTG
			CCL7_(1)_R	CCAGGGACACCGACTACTG
<i>Ptgs2</i>	COX-2	mouse	COX-2_F	TGAGCAACTATTCCAAACCAGC
			COX-2_R	GCACGTAGTCTTCGATCACTATC
<i>CSF1</i>	M-CSF	mouse	CSF1_(2)_F	AGTCAACAGAGCAACCAAACC
			CSF1_(2)_R	CAAAAATCCCTCACTAGCCTCTC
<i>Cxcl1</i>	KC	mouse	Cxcl1_(1)_F	CTGGGATTCACCTCAAGAACATC
			Cxcl1_(1)_R	CAGGGTCAAGGCAAGCCTC
<i>Cxcl10</i>	IP-10	mouse	Cxcl10_F	AGGACGGTCCGCTGCAA
			Cxcl10_R	CATTCTCACTGGCCCGTCAT
<i>Cxcl2</i>	MIP-2	mouse	Cxcl2_(1)_F	CCCTCAACGGAAGAACCAAAG
			Cxcl2_(1)_R	TTCCCGGGTGCTGTTTGT
<i>Edn1</i>	endothelin-1	mouse	ET-1_(1)_F	GCACCGGAGCTGAGAATGG
			ET-1_(1)_R	GTGGCAGAAGTAGACACACTC
<i>Fos</i>	c-Fos	mouse	Fos_(2)_F	CCCATCCTTACGGACTCCC
			Fos_(2)_R	GAGATAGCTGCTCTACTTTGCC
<i>Icam1</i>	ICAM1	mouse	ICAM1_F	TGTCAGCCACTGCTTGTTGTA
			ICAM1_R	CAGGATCTGGTCCGCTAGCT
<i>Ifit1</i>	Ifit1	mouse	Ifit1_(1)_F	CTGAGATGTCACTTCACATGGAA
			Ifit1_(1)_R	GTGCATCCCCAATGGGTTCT
<i>Nfkbia</i>	I $\kappa$ B $\alpha$	mouse	I $\kappa$ B $\alpha$ _F	CTGCAGGCCACCAACTACAA
			I $\kappa$ B $\alpha$ _R	CAGCACCCAAAGTACCAAGT
<i>Nfkbie</i>	I $\kappa$ B $\epsilon$	mouse	I $\kappa$ B $\epsilon$ _(1)_F	TGGACCTCCAAGTGAAGAAGT
			I $\kappa$ B $\epsilon$ _(1)_R	TTCTCTGCAATGTGGCAATG
<i>Il1b</i>	interleukin-1 $\beta$	mouse	IL-1b_(1)_F	GCAACTGTTCTGAACTCAACT
			IL-1b_(1)_R	ATCTTTTGGGGTCCGTCAACT
<i>IL6</i>	interleukin 6	mouse	IL6_F	TAGTCCTTCCCTACCCCAATTTCC
			IL6_R	TTGGTCCTTAGCCACTCCTTC
<i>Irf1</i>	Irf1	mouse	IRF1_(2)_F	AGGCCGATACAAAGCAGGAGA
			IRF1_(2)_R	GCTGCCCTTGTTCTACTCTG
<i>Jun</i>	c-Jun	mouse	Jun_(2)_F	ACTCGGACCTTCTCACGTC
			Jun_(2)_R	CGGTGTAGTGGTGATGTGCC
<i>Rpl32</i>	L32	mouse	L32_F	AAGCGAAACTGGCGGAAAC
			L32_R	TAACCGATGTTGGGCATCAG
<i>Mmp10</i>	MMP-10	mouse	MMP-10_(2)_F	AACACGGAGACTTTTACCCTTTT
			MMP-10_(2)_R	GGTGCAAGTGTCCATTTCTCAT
<i>Mmp13</i>	MMP-13	mouse	MMP-13_(2)_F	ACCTCCACAGTTGACAGGCT
			MMP-13_(2)_R	AGGCACTCCACATCTTGTTTT

### Supplementary Table 3. Continued

<b>name</b>	<b>alias</b>	<b>species</b>	<b>primer name</b>	<b>sequence</b>
<i>Mmp3</i>	MMP-3	mouse	MMP-3_(2)_F MMP-3_(2)_R	TGTCCCGTTTCCATCTCTCTC TGGTGATGTCTCAGGTTCCAG
<i>Saa3</i>	Saa3	mouse	Saa3_(1)_F Saa3_(1)_R	TGCCATCATTCTTTGCATCTTGA CCGTGAACTTCTGAACAGCCT
<i>Serpina3g</i>	Spi2A	mouse	Spi2A_(1)_F Spi2A_(1)_R	CTTCCAACGGCTGGAATCTA ACTGTCCAATCAGGCATAGCG
<i>TLR2</i>	TLR2	mouse	TLR2_(3)_F TLR2_(3)_R	CCAGACACTGGGGTAACATC CGGATCGACTTTAGACTTTGGG
<i>Tnf</i>	TNF $\alpha$	mouse	TNF_F TNF_R	CTACTCCCAGGTTCTCTTCAA GCAGAGAGGAGGTTGACTTTT
<i>Vcam1</i>	Vcam1	mouse	Vcam1_(1)_F Vcam1_(1)_R	AGTTGGGGATTTCGGTTGTCT CCCCTCATTCCTTACCACCC
<i>Zfp36</i>	TTP	mouse	Zfp36_(1)_F Zfp36_(1)_R	TCTCTGCCATCTACGAGAGCC CCAGTCAGGCGAGAGGTGA
<i>Zfp36</i>	TTP	mouse	Zfp36_(2)_F Zfp36_(2)_R	TCGAAGAGACCCTAACCCAGGC GCGTAGTCATCAGGATCGGA
<i>C3</i>	C3	human	hC3_(1)_F hC3_(1)_R	CCTGCTACTAACCACCTCC AACAGTGACTGGAACATCCCC
<i>CCL2</i>	MCP-1	human	hCCL2_(1)_F hCCL2_(1)_R	CAGCCAGATGCAATCAATGCC TGGAATCCTGAACCCACTTCT
<i>CCL5</i>	Rantes	human	hCCL5_F hCCL5_R	CCCAGCAGTCGTCTTTGTCA TCCCGAACCCATTTCTTCTCT
<i>CXCL1</i>	CXCL1	human	hCXCL1_F hCXCL1_R	CTGCGCTGCCAGTGCTTGCA TGTGGCTATGACTTCGGTTTG
<i>CXCL10</i>	IP-10	human	hCXCL10_(1)_F hCXCL10_(1)_R	GTGGCATTCAAGGAGTACCTC GCCTTCGATTCTGGATTCAGACA
<i>FOS</i>	c-FOS	human	hFOS_(3)_F hFOS_(3)_R	GGGCAAGGTGGAACAGTTATC CCGCTTGAGTGTATCAGTCA
<i>GAPDH</i>	GAPDH	human	hGAPDH_F hGAPDH_R	AACAGCGACACCCACTCCTC CATACCAGGAAATGAGCTTGACAA
<i>ICAM1</i>	ICAM1	human	hiCAM1_(1)_F hiCAM1_(1)_R	TCTGTGTCCCCCTCAAAAGTC GGGGTCTCTATGCCCAACAA
<i>NFKBIA</i>	I $\kappa$ B $\alpha$	human	hIkBa_(1)_F hIkBa_(1)_R	CTCCGAGACTTTTCGAGGAAATAC GCCATTGTAGTTGGTAGCCTTCA
<i>IL6</i>	Interleukin6	human	hIL-6_(1)_F hIL-6_(1)_R	AAATTCGGTACATCCTCGACGG GGAAGGTTTCAGGTTGTTTTCTGC
<i>Rpl32</i>	Rpl32	human	hL32_F hL32_R	AGCTCCCAAAAATAGACGCAC TTCATAGCAGTAGGCACAAAGG
<i>GFP</i>	GFP		GFP_(BMC)_F GFP_(BMC)_R	GGAGCGCACGATCTTCTTCA AGGGTGTCCGCTCGAA
<i>GFP</i>	GFP		GFP_(6)_F GFP_(6)_R	ACTTCAAGATCCGCCACAACA TGGGTGCTCAGGTAGTGGTTG