Legends to Supplemental Figures

Ramirez-Carrozzi et al.

Supplemental Figure 1. Efficiency of Brg1 and Brm Knockdown by Retroviral shRNA Transduction.

Brg1/Brm knockdown experiments were performed as described in the Experimental Procedures and in Ramirez-Carrozzi et al. (2006). The Brg1 (left panel) and Brm (right panel) Western blots in this figure show the knockdown efficiency at the time of LPS stimulation, which was generally 5 days after retroviral transduction. HMG1 antibodies were included in the Western blots as a loading control. The effects of Brg1/Brm knockdown on gene transcription are shown in Figure 1.

Supplemental Figure 2. Normalized Precursor Transcript Levels for CpG-Island and Non-CpG-Island Genes

The results of two independent experiments are shown (parts A and B) examining relative precursor transcript levels for several LPS-induced genes containing CpG-island (red) or non-CpG-island (black) promoters. Two housekeeping genes, *Gapd* and *Act*, were examined as controls. Real-time RT-PCR signals for the various genes were normalized using genomic DNA. RT-PCR primers (see Suppl. Table 1C) used to examine most genes spanned an exon-intron junction, thereby restricting amplification to precursor transcripts. However, the primers for five of the genes (four Class A genes, *Irf1*, *Junb*, *Zfp36*, *Cxcl1,* and one Class D gene, *Ifnb1*), amplified both precursor transcripts and mature mRNA because no introns exist in these genes for the selective amplification of precursor transcripts. The relative transcript levels for these genes therefore over-represent the precursor transcript levels. Primers for two other genes, *Egr1* and *Act*, also amplified both precursor transcripts and mRNA. Results are presented as Relative Transcripts, but corresponds to the amount of genomic DNA (in ng) required to yield the same signal observed by real-time RT-PCR when analyzing total RNA.

Precursor transcripts were analyzed in unstimulated bone marrow-derived macrophages (top panels) or macrophages stimulated with LPS for 30 min (second panels) or 120 min (third panels). The bottom panel shows the fold-induction at the 30 min and 120 min time-point for each gene relative to the unstimulated value, following normalization to the *Gapd* transcript levels.

Supplemental Figure 3. Changes in RNA Polymerase II Levels at CpG-Island and Non-CpG-Island Promoters Following LPS Stimulation.

ChIP experiments were performed with RNA polymerase II antibodies in bone marrow-derived macrophages left unstimulated (top panel) or stimulated for 30 min (second panel) or 120 min (third panel) with LPS. PCR amplification efficiencies for the various primer pairs were normalized using genomic DNA. In each panel, genes are ordered from lowest to highest RNA polymerase II levels. The bottom panel directly compares the changes in polymerase levels at each gene at each time point. In this panel, the genes are ordered as in the top panel. The results show that polymerase levels change only modestly at most CpG-island promoters following induction, with more dramatic changes at most non-CpG-island promoters and a few CpG-island promoters (e.g. *Cxcl2*). Results are presented as the average values with standard deviations from 3 independent immunoprecipitation experiments using two different chromatin preparations.

Supplemental Figure 4. Modest Changes in Histone H3 Levels Following LPS Stimulation

ChIP experiments were performed with histone H3 antibodies in bone marrow-derived macrophages left unstimulated (top panel) or stimulated for 30 min (second panel) or 120 min (third panel) with LPS. PCR amplification efficiencies for the various primer pairs were normalized using genomic DNA. In each panel, genes are ordered from highest to lowest histone H3 levels. The bottom panel directly compares the change in histone H3 levels at each gene at each time point. In this panel, the genes are ordered as in the top panel. The results

show that, although histone H3 levels decrease considerably at some CpG-island and non-CpG-island genes following induction, the H3 levels remain relatively unchanged at many other genes, with a consistent trend in both unstimulated and stimulated cells toward lower histone H3 levels at CpG-island promoters. Results are presented as the average values with standard deviations from 3 independent immunoprecipitation experiments using two different chromatin preparations.

Supplemental Figure 5. **Unusually High DNase I Hypersensitivity Scores at Class A Genes in Resting CD4+ T Cells**

(A) Maximum DNase I hypersensitivity scores for the human homologues of 64 of the 67 genes in our dataset are shown from an analysis of quiescent CD4+ T cells (Boyle et al., 2008). Hypersensitivity scores greater than 2.5 are colored red. Scores were obtained from the Duke DNase sig track at the UCSC Genome Browser. The CpG content is shown for the corresponding mouse genes, although the presence or absence of CpG islands was found to be strongly conserved between the mouse and human genomes (data not shown). Expression studies (Wang et al. 2008b) revealed that at least 9 of the Class A genes are induced following activation of human CD4+ T cells. However, this number almost certainly represents an underestimate of the number of Class A genes that are induced in these cells because the studies examined mRNA levels only at late time points, after many Class A genes are known to be downregulated.

(B) Maximum DNase I hypersensitivity scores in unstimulated CD4+ T cells from the Boyle et al. (2008) analysis are shown for 7 non-CpG-island genes that exhibited the strongest induction in CD4+ T cells in mRNA expression analyses (Wang et al. 2008a, 2008b). None of these genes exhibit high scores in unstimulated cells.

Wang, Z., Zang, C., Rosenfeld, J.A., Schones, D.E., Barski, A., Cuddapah S., Cui, K., Roh, T.Y., Peng, W., Zhang, M.Q., and Zhao, K. (2008a). Combinatorial patterns of histone acetylations and methylations in the human genome. Nat. Genet. *40*, 897-903.

Wang, M., Windgassen, D., and Papoutsakis, E.T. (2008b). Comparative analysis of transcriptional profiling of CD3+, CD4+, and CD8+ T cells identifies novel immune response players in T-cell activation. BMC Genomics *9*, 225.

Supplemental Figure 6. Consensus IRF3 Binding Sites are Over-Represented in Class D Promoters

(A) DNA motifs that perfectly match the IRF3 consensus sequence A/TAANNGAAA were identified between -1 and -300 relative to the transcription start site of each of the 67 genes. IRF3 consensus sequences were found in this interval in 6 of the 10 Class D genes, but in only 6 of the remaining 57 genes, two of which exhibited IRF3-dependence (*Cxcl11* and *Gbp2*). Consensus Sp1 and NF-κB sites are also shown. Although perfect matches to the Sp1 and NF-κB consensus sequences were identified in only a small number of genes, the factors are thought to regulate many more genes by binding to DNA motifs that diverge from the strict consensus.

(B) The fraction of promoters in each class with consensus IRF3 binding sites between -1 and -300 relative to the main transcription start site (found in the Dbtss database) is shown, along with the consensus sequences used to survey the promoters.

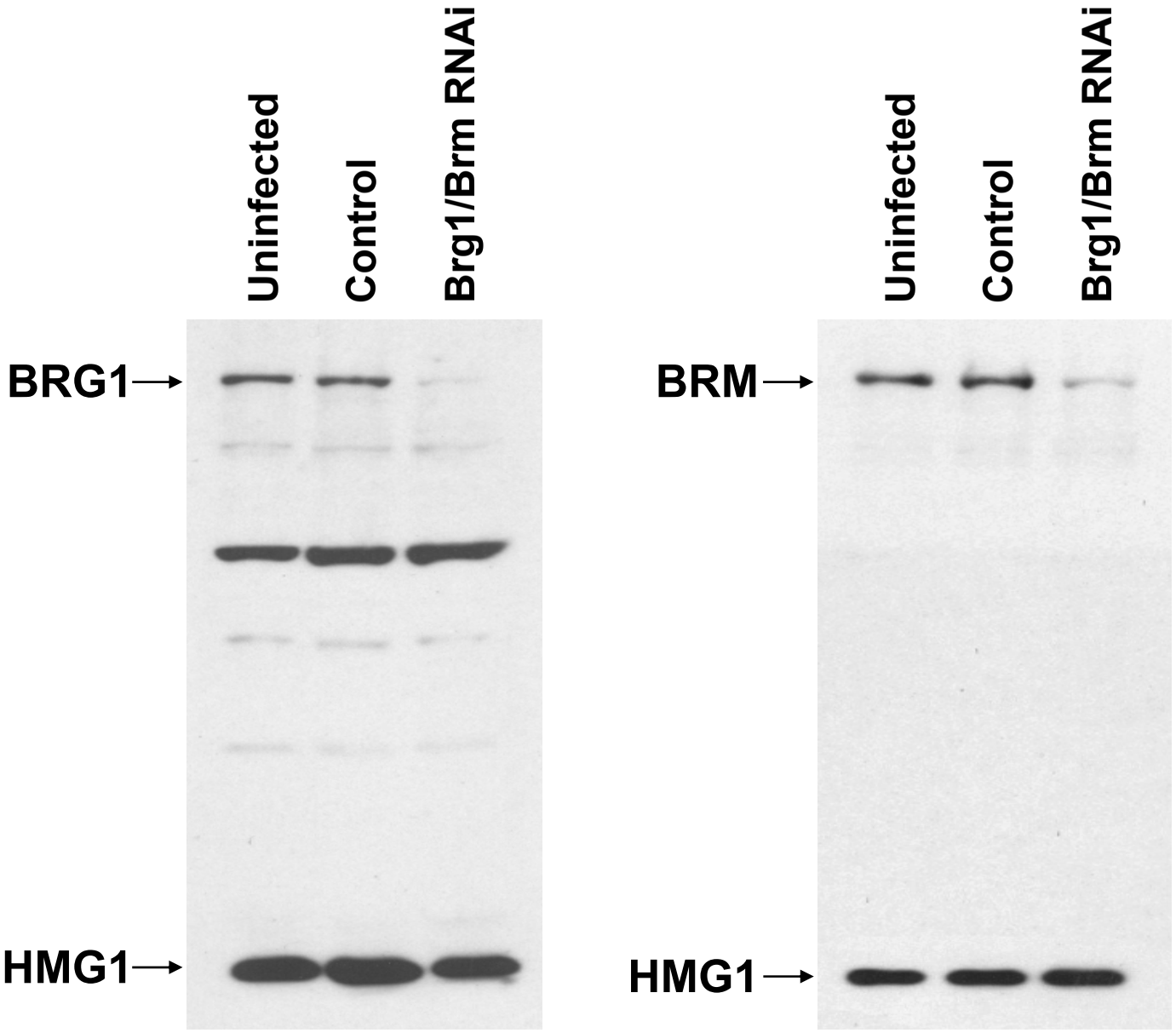
(C) ChIP experiments were performed with antibodies directed against IRF3, as well as GST as a negative control and C/EBPβ as a positive control, using chromatin from unstimulated and LPS-stimulated J774 macrophages. Results are presented as a percentage of input values.

Supplemental Figure 7. Fold-Induction Values for Genes Induced by Various Stimuli

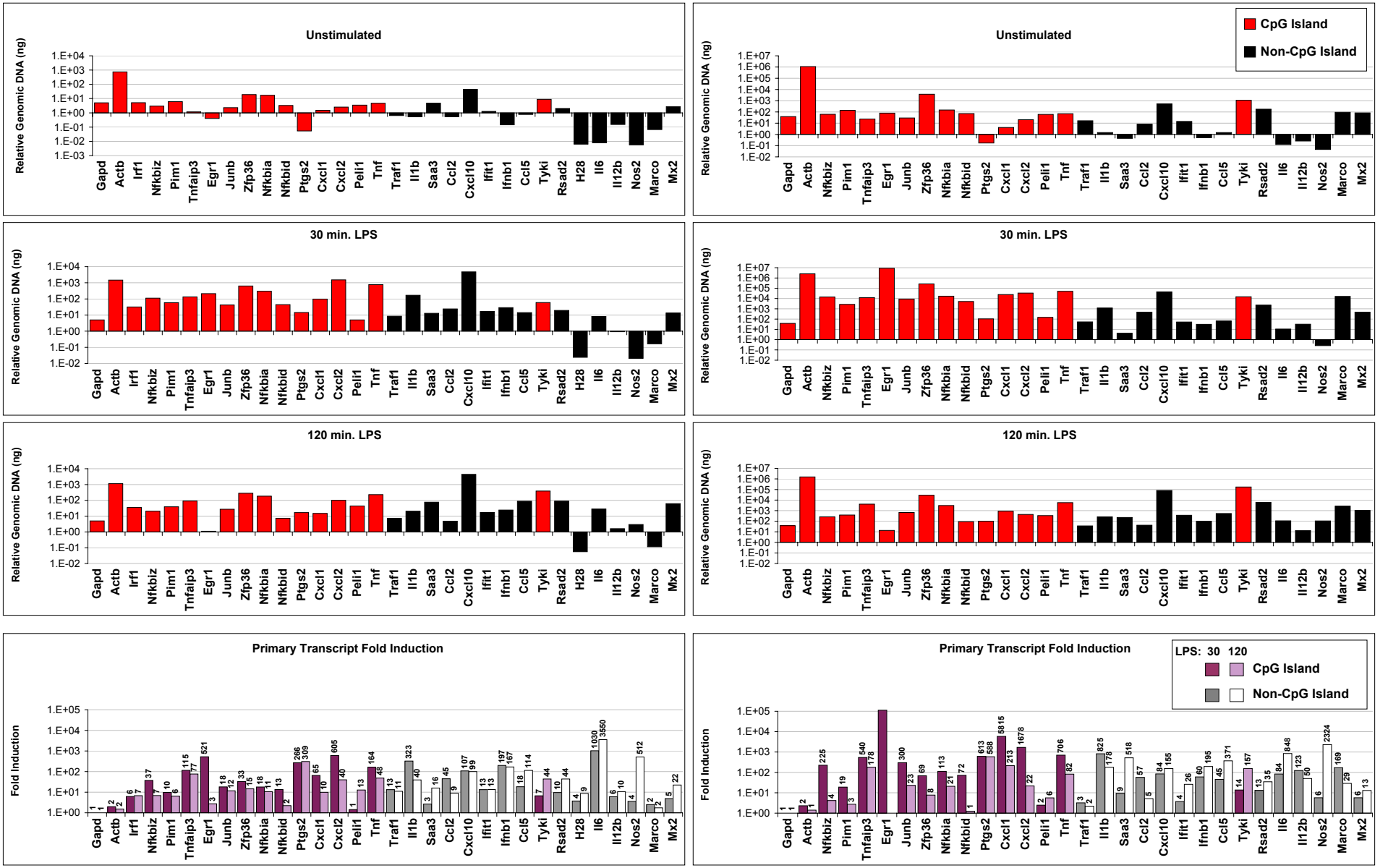
The data in Figure 5 of the main manuscript compare mRNA levels for 61 primary and secondary response genes following stimulation of bone marrow-derived macrophages with 5 different stimuli. In Figure 5, mRNA levels are shown as a percentage of the highest level found following induction by any of the stimuli (set at 100%). In this figure, the data are instead presented as fold-inductions at the 30-min, 1-hr, and 2-hr time points relative to the mRNA

levels observed in unstimulated cells. The results show the same trends that are apparent in Figure 5, with preferential activation of Class C and Class D genes by IFNβ and preferential activation of Class A genes by TNFα. However, these fold-induction values provide additional insights. For example, although TNFα activates Class A genes more strongly than genes in the other classes, especially when compared to the level of activation observed with the TLR stimuli, it can activate several genes in the other classes several fold above background. However, the expression levels remain very low relative to the levels achieved with the TLR stimuli. Furthermore, TLR2 activates Class D genes to a significant extent above background, despite the fact that TLR2 is known to be incapable of activating IRF3. Nevertheless, the level of induction of these genes achieved by TLR2 remains well below the level of induction achieved by TLR3 and TLR4, whereas genes in the other classes were induced similarly by TLR2, TLR3, and TLR4.

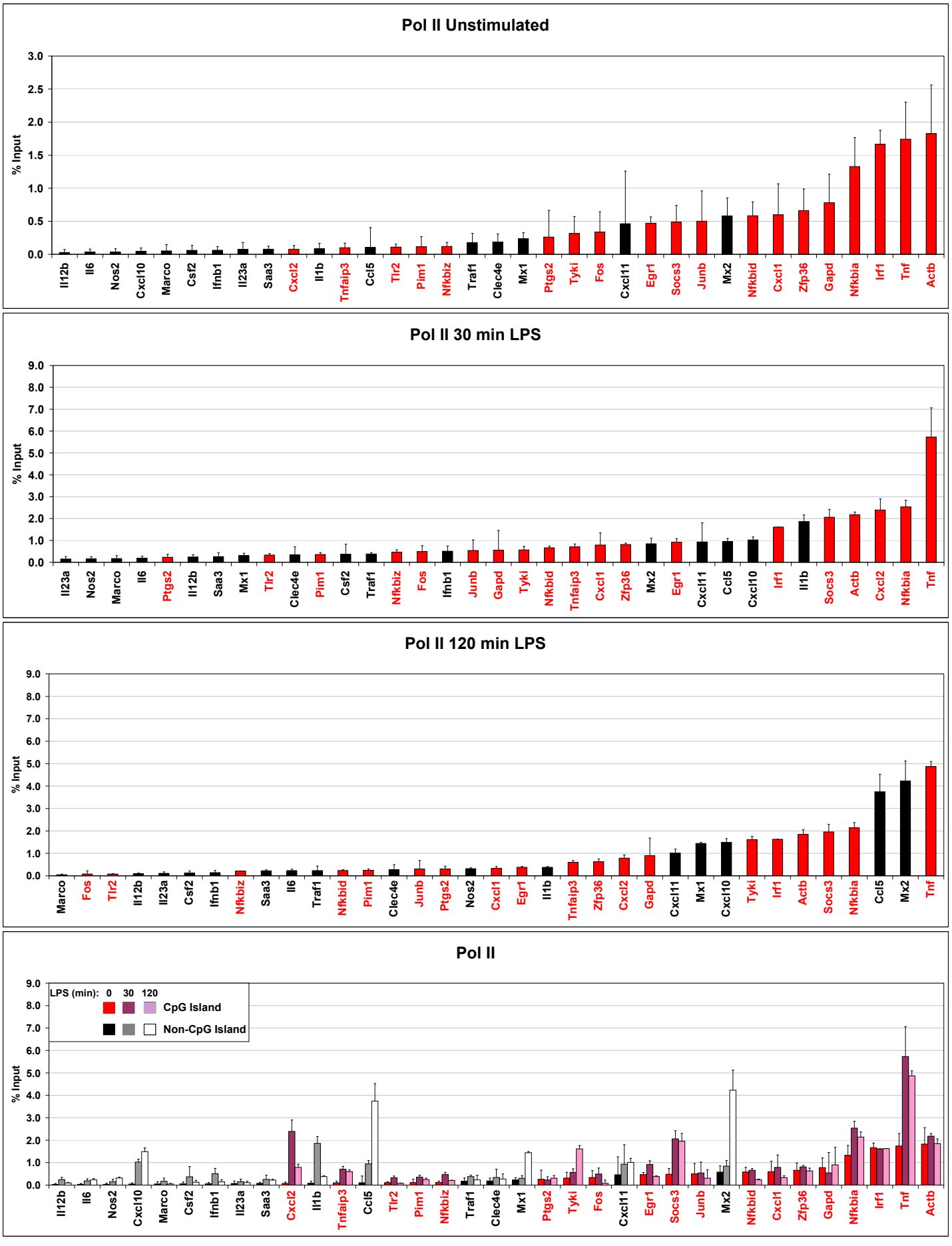
Supplemental Figure 1



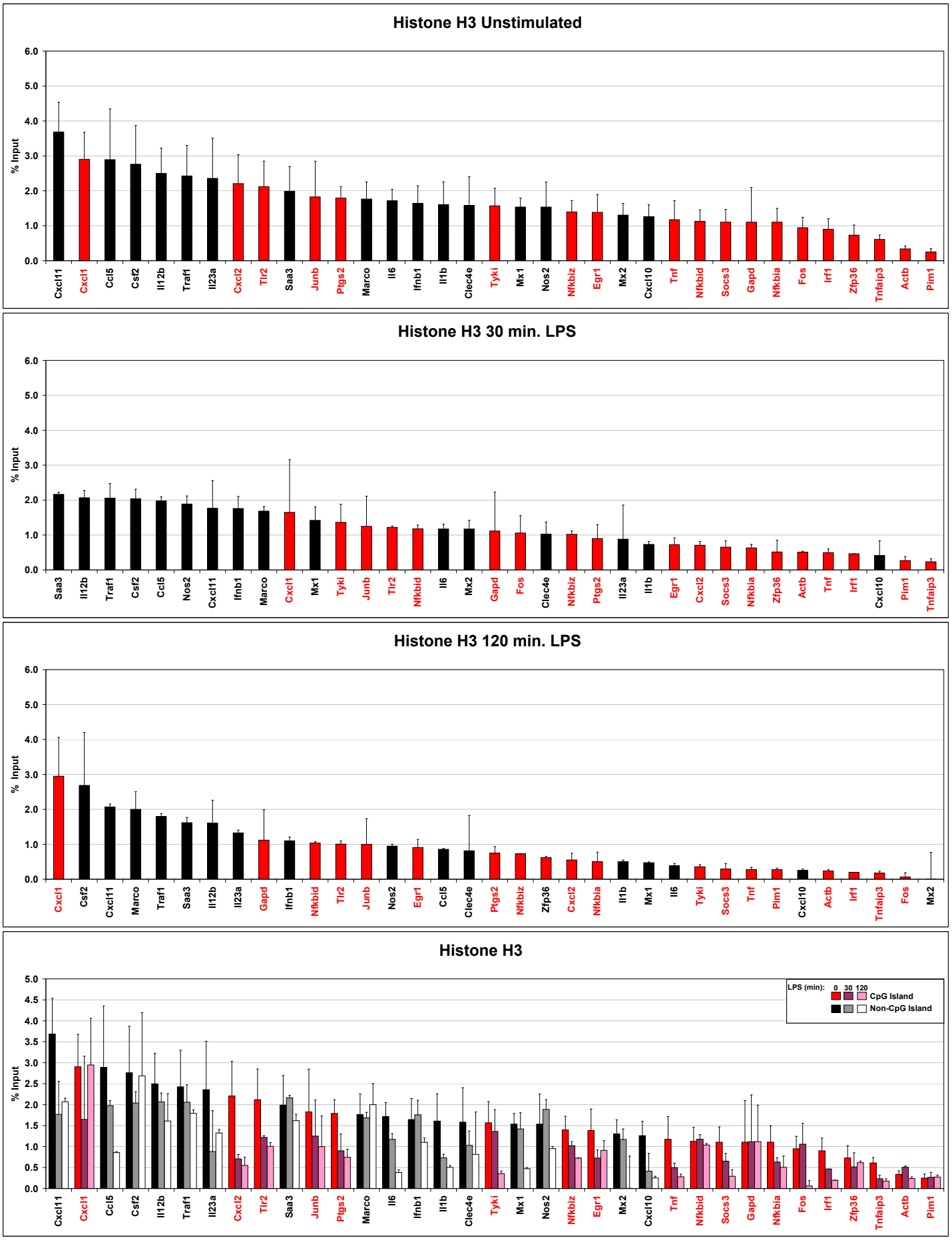
Supplemental Figure 2



Supplemental Figure 3



Supplemental Figure 4



Supplemental Figure 5

A

Class	Gene	CpG / Expected -200 / 1	DNaseI HS	Maximum DNaseI HS Score
Primary Response Genes				
A	*Cd83*	2.3	Yes	6.09
	Nr4a1	2.0	Yes	4.99
	Ccrn4l	2.0	Yes	3.62
	Irf1	1.9	Yes	3.96
	Nfkbiz	1.9	Yes	3.32
	Sod2	1.8	Yes	3.13
	Pim1	1.8	Yes	11.45
	Socs3	1.7	Yes	5.06
	Tnfaip3	1.5	Yes	4.68
	Egr1	1.5	Yes	5.60
	Junb	1.5	Yes	2.65
	Marcksl1	1.4	Yes	1.05
	Bcl3	1.3	Yes	3.58
	Icam1	1.3	Yes	1.79
	Zfp36	1.2	Yes	2.62
	Tnfsf9	1.1	Yes	1.55
	Nfkbia	1.1	Yes	4.39
	Fosb	1.1	Yes	3.48
	Egr2	1.1	Yes	5.94
	Fos	1.0	Yes	4.42
	Nfkbid	0.8	Yes	2.79
	Ptgs2	0.8	No	
	Tlr2	0.7	Yes	2.25
	Cxcl1	0.7	No	
	Cxcl2	0.6	No	
	Tnf	0.6	Yes	1.75
B	*Ccrl2*	0.5	Yes	1.37
	Traf1	0.2	Yes	1.64
	Cxcl11	0.2	No	
	Clec4e	0.2	No	
	Il1a	0.2	No	
	Csf2	0.2	No	
	Il23a	0.2	Yes	2.27
	Ccl3	0.2	No	
	Gbp2	0.0	Yes	1.75
	Gbp1	0.1	Yes	1.64
	Il1b	0.0	No	
C	*Map3k8*	1.0	Yes	2.53
	Serpine1	0.5	Yes	0.34
	Arhgef3	0.2	Yes	3.06
	Vcam1	0.2	No	
	Saa3	0.2	Yes	0.91
	Ccl2	0.2	No	
	Il10	0.2	No	
	Ikbke	0.2	Yes	2.02
D	*Peli1*	1.0	No	
	Ifit2	0.5	Yes	2.04
	Cxcl10	0.4	Yes	0.18
	Ifit1	0.2	Yes	2.30
	Ifnb1	0.1	No	
	Mmp13	0.1	No	
	Ccl5	0.0	No	
	Ifit3	0.0	Yes	1.32
Secondary				
E	*Tyki*	1.4	Yes	3.46
	Rsad2	0.2	Yes	1.81
F	*Irf7*	0.2	Yes	3.76
	Il6	0.2	Yes	0.70
	Il12b	0.2	No	
	Nos2	0.2	No	
	Lcn2	0.2	No	
	Marco	0.2	No	
	Mx1	0.2	Yes	3.04
	Mx2	0.1	Yes	2.32
	Serpinb3b	0.0	No	

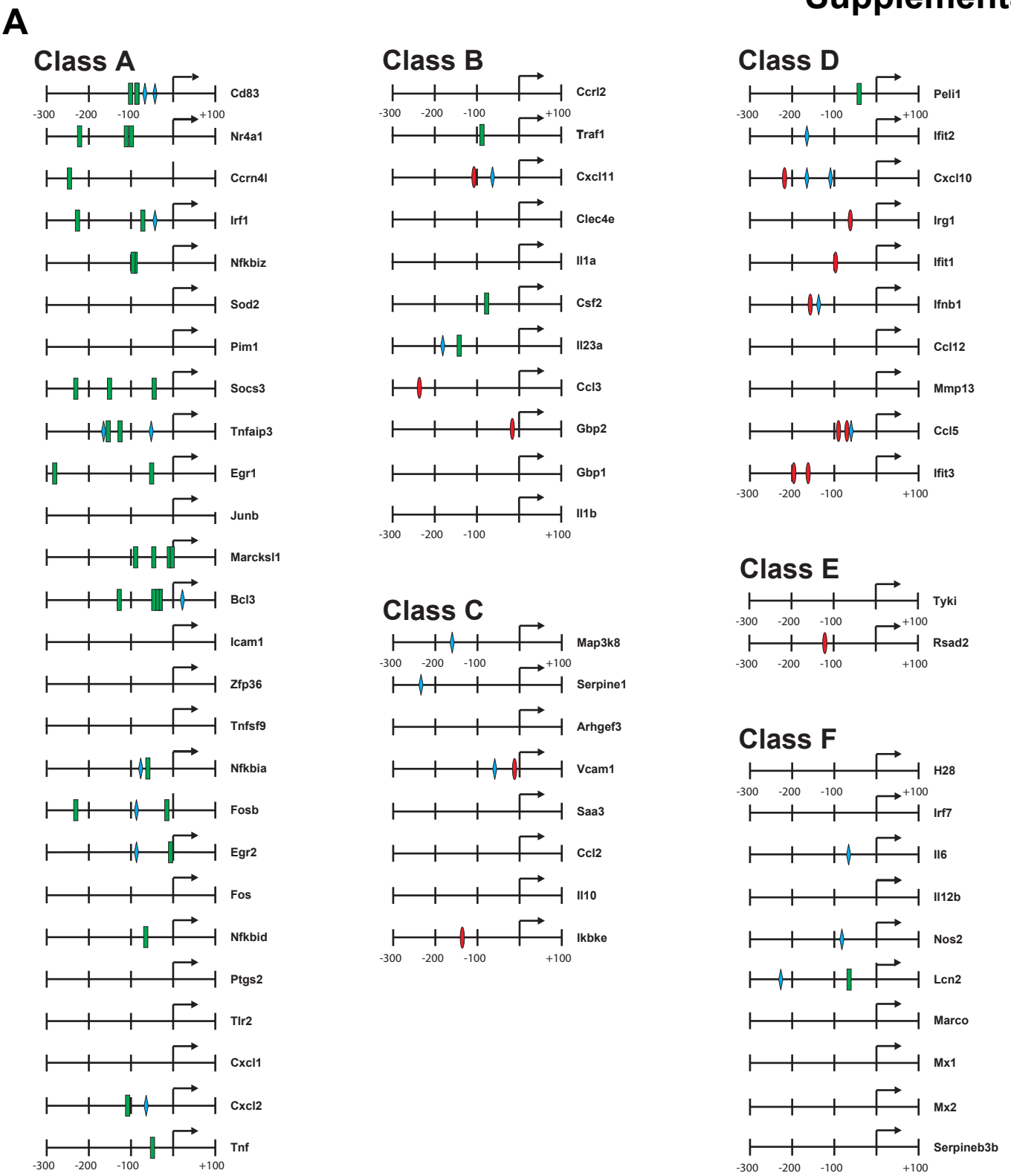
Maximum DNaseI HS Score
- ≥ 2.5
- < 2.5

CpG / Expected
- > 0.6
- 0.6
- < 0.6

B

Gene	CpG Island	DNaseI HS	Maximum DNaseI HS Score
Il2	No	Yes	1.44
Ccl4	No	No	
Ccl3	No	No	
Cxcl9	No	No	
Ifng	No	Yes	0.84
Csf2	No	No	
Xcl2	No	No	

Supplemental Figure 6

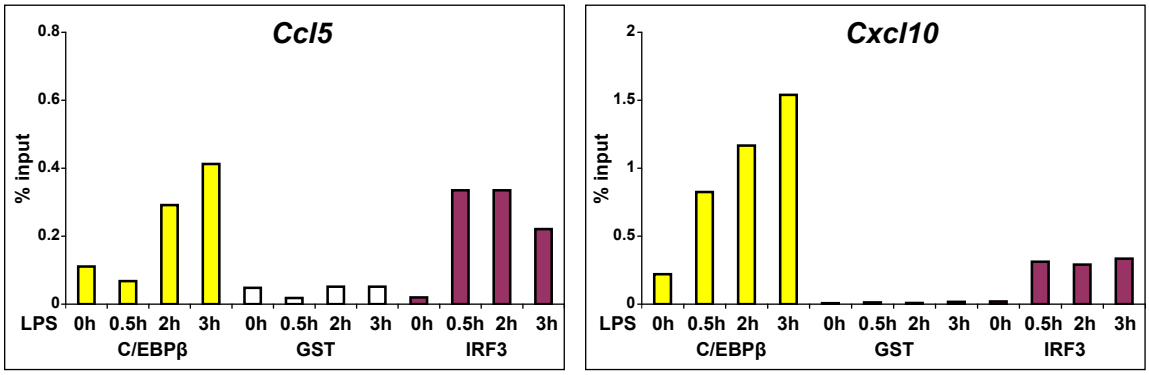


B

Class	Potential IRF3 Binding Sites
A	0 / 26
B	3 / 11
C	2 / 8
D	6 / 10
E	1 / 2
F	0 / 10

IRF3	A/TAANNGAAA
NFκB	GGGRNNYYCC
Sp1	GGCGGG

C



Supplemental Figure 7

Class	Gene	CpG / Expected -200/-1	TLR2 30'	TLR2 1h	TLR2 2h	TLR3 30'	TLR3 1h	TLR3 2h	TLR4 30'	TLR4 1h	TLR4 2h	IFNβ 30'	IFNβ 1h	IFNβ 2h	TNFα 30'	TNFα 1h	TNFα 2h	% Induction Brg1/Brm RNAi	% Induction IRF3-/- w/ CHX
Primary Response Genes																			
A	*Cd83*	2.3	5	14	11	2	13	12	9	19	17	1	2	2	8	13	6	167	422
	Nr4a1	2.0	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	200	246
	Ccrn4l	2.0	3	14	33	2	12	36	6	35	62	2	6	7	4	13	7	167	349
	Irf1	1.9	7	8	4	3	9	11	8	6	18	5	12	11	7	6	5	333	98
	Nfkbiz	1.9	6	13	7	3	11	3	11	22	12	1	2	1	5	2	2	71	323
	Sod2	1.8	2	4	10	1	4	9	2	5	13	1	2	2	2	4	6	167	152
	Pim1	1.8	5	11	10	3	12	8	8	19	13	3	4	6	7	7	6	100	120
	Socs3	1.7	18	48	67	7	47	64	24	74	118	20	37	11	24	11	11	200	19
	Tnfaip3	1.5	16	37	25	6	49	40	28	87	88	1	2	2	26	11	9	125	129
	Egr1	1.5	3	1	1	2	3	1	8	1	1	1	1	1	2	0	2	111	1022
	Junb	1.5	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	143	220
	Marcksl1	1.4	4	9	11	2	12	16	7	13	23	1	1	1	4	7	7	57	163
	Bcl3	1.3	2	4	10	1	4	8	2	3	11	1	1	1	2	4	5	167	100
	Icam1	1.3	3	13	15	2	9	16	4	13	21	1	1	2	3	9	7	91	211
	Zfp36	1.2	13	9	6	4	19	4	17	25	8	2	2	2	13	3	4	100	226
	Tnfsf9	1.1	46	118	35	16	158	54	82	266	89	2	2	1	66	31	18	250	25
	Nfkbia	1.1	9	12	13	5	16	10	17	18	20	1	1	1	13	6	8	200	87
	Fosb	1.1	15	17	2	6	12	2	43	48	9	1	1	1	27	16	1	500	175
	Egr2	1.1	9	17	3	4	17	4	18	27	4	2	2	1	9	7	3	1000	56
	Fos	1.0	12	2	1	4	11	1	16	8	1	1	0	1	9	1	1	100	298
	Nfkbid	0.8	11	8	2	6	12	1	20	9	2	0	0	0	10	4	4	100	37
	Ptgs2	0.8	109	194	404	29	230	149	170	531	404	2	1	5	68	15	33	91	220
	Tlr2	0.7	3	7	9	1	5	8	2	6	10	1	1	0	3	5	4	71	41
	Cxcl1	0.7	121	117	55	24	149	13	121	228	54	1	1	1	62	7	19	91	67
	Cxcl2	0.6	158	319	113	53	371	40	219	552	91	1	0	0	207	40	37	71	231
	Tnf	0.6	92	171	72	41	292	45	163	329	113	1	1	2	77	43	51	77	50
B	*Ccrl2*	0.5	12	25	16	5	35	43	28	65	75	2	2	8	16	18	7	250	68
	Traf1	0.2	2	9	17	2	7	17	3	15	29	1	1	1	2	9	8	83	290
	Cxcl11	0.2	7	12	38	4	66	921	33	247	2216	7	71	247	5	5	8	125	0
	Clec4e	0.2	6	14	21	3	14	19	9	13	22	1	1	1	8	10	16	48	348
	Il1a	0.2	10	76	34	7	91	37	36	364	198	3	3	2	19	12	1	67	177
	Csf2	0.2	105	694	103	43	790	37	560	3022	206	2	0	1	99	50	6	180	1746
	Il23a	0.2	1	1	1	1	2	1	3	13	2	1	1	1	1	1	1	1000	480
	Ccl3	0.2	5	10	12	3	10	12	9	23	23	4	7	5	6	3	1	200	81
	Gbp2	0.0	2	3	7	1	3	20	2	5	31	4	15	46	2	2	5	133	15
	Gbp1	0.1	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	200	ND
	Il1b	0.0	49	205	160	20	175	118	98	385	274	1	1	1	54	32	10	77	54
C	*Map3k8*	1.0	2	2	2	1	2	2	1	1	5	2	4	3	1	1	1	29	198
	Serpine1	0.5	2	11	66	1	8	55	4	56	559	1	1	1	2	6	8	13	613
	Arhgef3	0.2	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	30	130
	Vcam1	0.2	1	10	15	1	7	19	2	21	38	1	1	1	2	7	11	26	541
	Saa3	0.2	2	12	116	2	6	65	2	8	86	1	2	2	2	4	12	7	398
	Ccl2	0.2	7	23	8	4	34	17	15	81	51	18	21	15	7	5	1	26	185
	Il10	0.2	9	3	5	4	9	5	16	11	11	2	4	8	6	1	1	19	227
	Ikbke	0.2	1	2	3	1	1	3	1	1	4	1	1	1	1	2	2	22	85
D	*Peli1*	1.0	1	1	2	1	6	7	2	10	8	1	2	3	1	1	1	29	7
	Ifit2	0.5	1	1	2	1	1	10	1	2	14	3	9	29	1	1	1	34	1
	Cxcl10	0.4	12	9	21	9	71	206	54	111	221	49	140	176	10	2	5	43	1
	Irg1	0.2	5	29	69	5	60	188	26	117	234	9	19	34	6	17	29	24	5
	Ifit1	0.2	3	19	58	2	21	148	6	60	213	20	43	52	3	12	27	27	0
	Ifnb1	0.1	2	3	4	4	109	100	48	279	98	1	2	3	1	1	2	15	0
	Ccl12	0.1	7	14	15	5	21	38	13	41	32	13	31	62	7	6	5	6	9
	Mmp13	0.1	1	4	19	1	3	45	2	13	173	6	50	191	2	5	5	8	0
	Ccl5	0.0	1	3	21	1	4	40	1	14	151	1	19	76	1	3	6	20	2
	Ifit3	0.0	1	1	1	1	1	5	1	1	5	2	7	17	1	1	1	30	1
Secondary																			
E	*Tyki*	1.4	1	1	3	1	2	25	2	5	36	8	17	38	1	1	1	43	
	Rsad2	0.2	2	1	2	1	2	22	2	6	35	7	16	51	1	1	1	77	
F	*H28*	0.2	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	5	
	Irf7	0.2	1	1	1	1	1	2	1	1	2	2	3	7	1	1	1	26	
	Il6	0.2	7	14	20	3	33	97	16	140	274	8	15	18	5	2	2	6	
	Il12b	0.2	2	17	13	2	34	53	6	74	93	1	1	1	3	3	2	6	
	Nos2	0.2	1	5	233	1	3	202	1	8	386	1	2	20	1	2	10	18	
	Lcn2	0.2	1	2	11	1	2	5	1	2	6	1	1	1	1	1	1	2	
	Marco	0.2	1	4	15	1	2	4	1	2	5	1	1	1	2	2	3	18	
	Mx1	0.2	2	1	4	1	1	27	1	1	32	6	26	63	1	1	1	2	
	Mx2	0.1	1	1	4	1	2	22	1	5	25	12	11	13	1	1	2	7	
	Serpinb3b	0.0																8	

Induction: highest; 2nd highest; 3rd highest

CpG / Expected: > 0.6; 0.6; < 0.6

% Induction: ≥ 50; 33 - 49%; 0 - 33%

A

Class	Gene	Forward Primer	Reverse Primer
HK	*Gapd*	TGGTGAAGGTCGGTGTGAAC	CCATGTAGTTGAGGTCAATGAAGG
	Actb	AGAGGGAAATCGTGCGTGAC	CAATAGTGATGACCTGGCCGT
A	*Cd83*	CATCCTCAGATGGCAACCTT	TGCTCAAGACCCTGTGTCAG
	Nr4a1	CTGTCCGCTCTGGTCCTCATC	GGTCTCCTGCCACGGTAGC
	Ccrn4l	AAAGATCCCCCTGATCGTCT	GGGACTCAGCAGCTTGTAGG
	Irf1	TCCAAGTCCAGCCGAGACA	TGCTGAGTCCATCAGAGAAAGTGT
	Nfkbiz	GTGGCAGGTAGAGCAGGAAG	CCTTGGGCAACAGCAATATG
	Sod2	CGGCCTACGTGAACAATCTC	TGAACTTCAGTGCAGGCTGA
	Pim1	TCAAGGACACAGTCTACACGG	AGCGATGGTAGCGAATCC
	Socs3	GCTCCAAAAGCGAGTACCAGC	AGTAGAATCCGCTCTCCTGCA
	Tnfaip3	GGCAGCTGGAATCTCTGAAA	CTGCAGGTGTGTCTGCTGAT
	Egr1	CCTGGCCTGGATAAAAGTCA	GCGGAACCCTGATTGTTCTA
	Junb	TACTTTTCGGGTCAGGGATCA	GCGCTCCAGTTCCGTGG
	Marcksl1	CAAATTGAGTGGCCTGTCCT	TGCTCCTGCTCTTCCTCTGT
	Bcl3	GCGAAGTAGACGTCCATAACAACC	ACCAAGAGCCGGACCATGT
	Icam1	TGTCAGCCACTGCCTTGGTA	CAGGATCTGGTCCGCTAGCT
	Zfp36	CCCTCTGCAACTCTGGTCTC	GACCACCGGACACTGAACTT
	Tnfsf9	GCCCCAACACTACACAACAG	GCTGTGCCAGTTCAGAGTTG
	Nfkbia	CCTGGCCAGTGTAGCAGTCT	AGAGGCTAGGTGCAGACACG
	Fosb	TGCGGGCCTTTGTTAATATG	CAACAGGTGCCGCACAAT
	Egr2	CTCCTCCTGGACGAATTGA	CAGTCAGATAGGGGTTCACATT
	Fos	GTGCCAGCTGCTATCCAGAAG	GGCTGTGGTTGAAGCTGGAG
	Nfkbid	GTGACAGCCATCTCCACCA	GTGGGGAGTCCGTAAGGATG
	Ptgs2	CCCCTTCCTGCGAAGTTTA	GAGAAGGCTTCCCAGCTTTT
	Tlr2	TTTGCTGGGCTGACTTCTCT	TCGCGGATCGACTTTAGACT
	Cxcl1	TGTCAGTGCCTGCAGACCAT	GTGGCTATGACTTCGGTTTGG
	Cxcl2	GCCAAGGGTTGACTTCAAGA	ACTTTTTGACCGCCCTTGAG
	Tnf	CCCCAAAGGGATGAGAAGTT	TGGGCTACAGGCTTGTCACT
B	*Ccrl2*	TTCCAACATCCTCCTCCTTG	GATGCACGCAACAATACCAC
	Traf1	TGTGTGGCCGGACTGTCA	AGCGCAGGCACAACTTGTAAC
	Cxcl11	AGTAACGGCTGCGACAAAGT	CTGCATTATGAGGCGAGCTT
	Clec4e	CTGTGCCACCATAAGGGACT	TCTGGCATCTCACAAATCCA
	Il1a	AGCAGCCTTATTTCGGGAGT	GTGCAAGTGACTCAGGGTGA
	Csf2	TTTTGTGCCTGCGTAATGAG	CAGCGTTTTCAGAGGGCTAT
	Il23a	GGTGCTTATAAAAAGCCAGACC	AATAATGTGCCCCGTATCCA
	Ccl3	AGATTCCACGCCAATTCATC	CCCAGGTCTCTTTGGAGTCA
	Gbp2	CTCTACCGCACAGGCAAATC	GATGCCCTTGGTGTGAGACT
	Gbp1	ATCATATCCCTTAAACTTCAGGAACAG	GTGGAAACAGGGTAGAGAGCTTTAGT
	Il1b	GCTGAAAGCTCTCCACCTCA	AGGCCACAGGTATTTTGTCG
C	*Map3k8*	TCCAAGAAAGTGATCCACCA	CACTCAGGCCAAAATCTACCA
	Serpine1	CCGATCCTTTCTCTTTGTGG	CAAATGAAGGCGTCTCTTCC
	Arhgef3	AAGACCTGCAGGATGGAGAA	ACCGAGTCTGCTACCCACAC
	Vcam1	GCTGTGACCTGTCTGCAAAG	GTCTCCCATGCACAAGTGG
	Saa3	CCTTCCATTGCCATCATTCT	AGTAGGCTCGCCACATGTCT
	Ccl2	GGGCCTGCTGTTCACAGTT	GGGATCATCTTGCTGGTGAA
	Il10	AAGGACCAGCTGGACAACAT	TCATTTCCGATAAGGCTTGG
	Irg1	CACAGAGAGCTTTGCTGGTATGA	TGCTCCTCCGAATGATACCAT
	Ikbke	AGCTATTCGCAGTGGAGGAA	CGAACGTGTTCTCAGGGTCT
	Ccl12	GTCCGGAAGCTGAAGAGCTA	GGGTCAGCACAGATCTCCTT
	Mmp13	GTTCAAGGAATTCAGTTTCTTTATGGT	GGTAATGGCATCAAGGGATAGG
D	*Peli1*	CTTCCAAAGCCCCAGTAAAA	ACCCAGAGACCAAAATGAGC
	Ifit2	AGAAATGCCAGGAAGACAGC	GGTGTGACACATTTCACATGG
	Cxcl10	CCTGCCCACGTGTTGAGAT	GAGTCACAGACCCGTCCCTA
	Ifit1	TCCATTTCTGGCCATTTCTC	TGAAGCAGATTCTCCATGACC
	Ifnb1	AGCTCCAAGAAAGGACGAACAT	GCCCTGTAGGTGAGGTTGATCT
	Ccl5	GTGCCCACGTCAAGGAGTAT	CCCACTTCTTCTCTGGGTTG
	Ifit3	AGTGAGGTCAACCGGGAATCT	TCTAGGTGCTTTATGTAGGCCA
E	*Tyki*	GGCAATTATCTCGTGGCTTC	GGCCTCCACTCACCTCAGTA
	Rsad2	AACCCCCGTGAGTGTCAACTA	AACCAGCCTGTTTGAGCAGAA
F	*H28*	ATGGTGGCAAAGCTGAAAAA	CCGCTCCTTCTGTAGTGTCC
	Irf7	TCTTCGCTCTCTTCGCTCA	GGTCGTAGGGATCTGGATGA
	Il6	GTTCTCTGGGAAATCGTGGA	TTTCTGCAAGTGCATCATCG
	Il12b	AGCCACTCACATCTGCTGCT	AACCGTCCGGAGTAATTTGG
	Nos2	CAGCTGGGCTGTACAAACCTT	CATTGGAAGTGAAGCGTTTCG
	Lcn2	TTCCGGAGCGATCAGTTCC	TGACCAGGATGGAGGTGACA
	Marco	ATCCTGCTCACGGCAGGTACT	GCACATCTCTAGCATCTGGAGCT
	Mx1	AAACCTGATCCGACTTCACTTCC	TGATCGTCTTCAAGGTTTCCTTGT
	Mx2	AGGGTACACGTGTCCTTTCG	TAGGGCAGTGATGTCCTGGT
	Serpinb3b	GCTGTCCTATGTCTCCCAGAA	ATCCCCCAGAAAGCTGAAGT

Primary Response Genes

Secondary

B

Class	Gene	Forward Primer	Reverse Primer
HK	*Gapd*	GGTCCAAAGAGAGGGAGGAG	GCCCTGCTTATCCAGTCCTA
	Actb	GAGGGGAGAGGGGGTAAAA	TCGAGCCATAAAAGGCAACT
A	*Irf1*	TTTCCCCGAAATGATGAGG	GCCGCGAAGAAATCTAAACA
	Nfkbiz	GGCCTTTGAGGTCACAATGA	AGGACTCCTGTCCCAGTGTC
	Pim1	CGTTAGCGACCATTCTGACC	GCTTCAGCCAACCAGAAGAC
	Socs3	CACAGCCTTTCAGTGCAGAG	GGGTATTTACCCGGCCAGT
	Tnfaip3	GCGGGACCTAGGAGTTTCTC	TTGCCAACAGGGGGATTT
	Egr1	GGCCGGTCCTTCCATATTAG	CGAATCGGCCTCTATTTCAA
	Junb	GTGTGTCTCTGTCTCCACAGC	TCGCGTCACTGTCAGGAA
	Zfp36	CGCTACCATCACCTCCAGTT	CATGCAAAATGTGCCTGAAC
	Nfkbia	GCTTCTCAGTGGAGGACGAG	CTGGCAGGGGATTTCTCAG
	Nfkbid	TACGGTGGTGGAAAGTTGGT	CGAGCGAGCTGGAGAACTAC
	Fos	GGGCGTAGAGTTGACGACAG	TGGATGGACTTCCTACGTCAC
	Ptgs2	CGCAACTCACTGAAGCAGAG	CCACGTGACGTAGTGGTGAC
	Tlr2	GCTGGAGCATTCCAATAACC	CTCGCCTTTTGGCTGAGTT
	Cxcl1	ACCCTGTACTCCGGGAATTT	GGAGTCTGGAGTGCTGGAAC
	Cxcl2	GGGCTCTGTGCTTCCTGAT	TCCCGAGAGCTCCTTTTATG
	Tnf	GATTCCTTGATGCCTGGGTGTC	GAGCTTCTGCTGGCTGGCTGT
B	*Traf1*	ACCACTTCCCTGCTTCACC	CCCGGTAAACTCTGCCAAT
	Cxcl11	GCTGAGTGCTTTCACCTTCC	CGTAGCTTTCTTGCCTCCTG
	Clec4e	AAGGAAAATCGGGACCAAGT	GCATCAAGAGAAATGGCAGAG
	Csf2	CCAGTTCTTGGAAGGGCTTA	GGAATCTCCTGGCCCTTATC
	Il23a	GCCTCTAGCCACAACAACCTC	ATTCCCCTCCCTACATCATCTC
	Il1b	CCCACCCTTCAGTTTTGTTG	CTTGTTTTCCCTCCCTTGTTT
C	*Saa3*	CGCAATCTGGGGAAAGAAG	AATGGAGCAATCCCTGTTTG
D	*Peli1*	CCTGACCAGTGAACAGAGATTTGA	GGGCTTTGGAAGGATGATTTTCT
	Cxcl10	TCCAAGTTCATGGGTCACAA	TGATTGGCTGACTTTGGAGA
	Ifit1	AGTGCGTCTCCCTGGATAAA	TTAAAGGGCTGGGTGAGCTA
	Ifnb1	GCCAGGAGCTTGAATAAAATG	GATGGTCCTTTCTGCCTCAG
	Ccl5	CTGCTACCCTGGCTCCCTAT	TGGGAGATGCATGTGCTGT
	Ifit3	GGACTGTCAGGCTGGAGGAAAT	TGTCCTGGCCACAGCATTG
E	*Tyki*	GCTGCCTTCACTTTCGTTTC	TTATTAGGGCCATGGGTGTC
F	*Il6*	AATGTGGGATTTTCCCATGA	GCTCCAGAGCAGAATGAGCTA
	Il12b	GGGGAGGGAGGAACTTCTTA	CTTTCTGATGGAAACCCAAAG
	Nos2	CCCTTTGGGAACAGTTATGC	GGGGCCAGAGTCTCAGTCTT
	Lcn2	GGGGAGAGAGGGACAGAAAT	CCTTTACCAAGTCCAGGAAGC
	Marco	GGAGGGTCTTTCCCAAACTT	CCGGCTGCTCCTTAGTGTAG
	Mx1	TCCCAACCTCAGTACCAAGC	GAAACCGTGAAAAAGCCTGA
	Mx2	GCAGCTGCACACTCTGTCTC	TGCCCTGCTGTACTTACCAGT

Primary Response Genes

Secondary

C

Class	Gene	Forward Primer	Reverse Primer
HK	*Gapd*	TGGTGAAGGTCGGTGTGAAC	TCATCCACCTCCCCACAGTA
	Actb	AGAGGGAAATCGTGCGTGAC	CAATAGTGATGACCTGGCCGT
A	*Irf1*	GTAGGTAGGGTGGGCAGTGA	TGCTGAGTCCATCAGAGAAAGTGT
	Nfkbiz	GTGGCAGGTAGAGCAGGAAG	CAAGCGTGAAAGGACATGAA
	Pim1	TCAAGGACACAGTCTACACGG	GGCCCAATGACCTTTACAA
	Tnfaip3	GGCAGCTGGAATCTCTGAAA	ATAGGGCTGCCACACAAAAG
	Egr1	GCGCTCTCGGAGCTGCAGTA	GGACCTGCTCCAGTGGCAGA
	Junb	AGCAGGGACCCATGGAAGTT	ATGCACAAGCAAAGGTCATCTTT
	Zfp36	CCCTCTGCAACTCTGGTCTC	GACCACCGGACACTGAACTT
	Nfkbia	CCTGGCCAGTGTAGCAGTCT	TGATGTTTGTGGTTGTCCTCA
	Nfkbid	TGCGGGCCTTTGTTAATATG	GGGTGCTAACAGCCATCTTG
	Ptgs2	CCCCTTCCTGCGAAGTTTA	TTGGGGTTGGGTTTCTAGTG
	Cxcl1	TGTCAGTGCCTGCAGACCAT	AGTCTGGAAGTGGCAGAAGC
	Cxcl2	GAACATCCAGAGCTTGAGTGTGA	CATGGCAGAGGCTATCCAG
	Trf	GGGAAGCCTAAAAGGCTCAT	TGGGCTACAGGCTTGTCACT
B	*Traf1*	TGTGTGGCCGGACTGTCA	AGCTGACCTAGCATGGCTGT
	Il1b	TGTTCCCTTGACCCAGACTC	AGGCCACAGGTATTTTGTCG
C	*Saa3*	CAGGATGAAGCCTTCCATTG	TCATGAACTGGACCCATCTTT
	Ccl2	GGGCCTGCTGTTCACAGTT	TGCTATCACAATCCACGACAA
D	*Peli1*	CTTCCAAAGCCCCAGTAAAA	ACCCAGAGACCAAAATGAGC
	Cxcl10	CCTGCCCACGTGTTGAGAT	GAGTCACAGACCCGTCCCTA
	Ifit1	TCCATTTCTGGCCATTTCTC	TGAAGCAGATTCTCCATGACC
	Ifnb1	AGCTCCAAGAAAGGACGAACAT	GCCCTGTAGGTGAGGTTGATCT
	Ccl5	TGGGTATCAAGATGGGTTGG	CTCACCATCATCCTCACTGC
E	*Tyki*	GGCAATTATCTCGTGGCTTC	TTAGGGAGAAGGCATCTTGC
	Rsad2	AACCCCCGTGAGTGTCAACTA	CCAGGAACTTACCAGCCTGT
F	*H28*	ATGGTGGCAAAGCTGAAAAA	CCGCTCCTTCTGTAGTGTCC
	Il6	GCCCTCTAGTGGTGCTTGTT	TTTCTGCAAGTGCATCATCG
	Il12b	AGCCACTCACATCTGCTGCT	ACTGTCAAGGGGGATGGAAT
	Nos2	CAGCTGGGCTGTACAAACCTT	CCTAGCAGAGGCCATGACAC
	Marco	ATCCTGCTCACGGCAGGTACT	GCAGCCTGAACCTTACACCA
	Mx2	AGGGTACACGTGTCCTTTCG	TAGGGCAGTGATGTCCTGGT

Primary Response Genes

Secondary