

SUPPLEMENTARY INFORMATION

Systems Biological Approaches Reveal Non-additive Responses and Multiple Crosstalk Mechanisms between TLR and GPCR Signaling

Jayalakshmi Krishnan, Sangdun Choi*

Department of Molecular Science and Technology, Ajou University, Suwon 443-749, Korea

Supplementary Table 1. The genes up- and down-regulated during KDO treatment in RAW 267.4 cells at 60 min and 120 min as analyzed by MeV

Function and GenBank identification	Gene name	60 min	120 min
Inflammation/Immune response			
NM_008361	Interleukin 1 beta ³	1.7	4.8
	Interleukin 1 beta ³	2.6	5.4
NM_010554	Interleukin 1 alpha	1.8	4.0
NM_013693	Tumor necrosis factor ³	NA	3.2
	Tumor necrosis factor ³	2.6	3.4
NM_013652	Chemokine ligand(c-x-c) 4 ³	1.0	3.3
	Chemokine ligand(c-x-c) 4 ³	1.2	3.3
XM_127883	Immunoresponsive gene 1	1.9	4.2
NM_021274	Chemokine (c-x-c) motif ligand 10 ³	NA	2.0
	Chemokine (c-x-c) motif ligand 10 ³	1.7	NA
NM_009845	CD22 antigen	1.7	2.0
NM_009404	Tumor necrosis factor (ligand) superfamily, member 9	1.4	2.1
NM_010510	Interferon beta 1, fibroblast	NA	1.5
NM_007795	Cardiotropin 1	NA	1.3
NM_019948	C-type lectin domain family 4, member	1.4	2.3
NM_010501	Interferon-induced protein with tetratricopeptide repeats 3	3.1	3.9
NM_138648	Oxidized low density lipoprotein	NA	1.8
NM_031167	Interleukin 1 receptor antagonist	NA	1.5
NM_009137	Chemokine (C-C motif) ligand 22	NA	2.2
Transcription factor			
NM_010755	v-maf musculoaponeurotic fibrosarcoma oncogene family, protein F (avian) ³	1.1	1.8
	v-maf musculoaponeurotic fibrosarcoma oncogene family, protein F (avian) ³	NA	-1.1
NM_008690	Nuclear factor of kappa light polypeptide gene enhancer in B cells inhibitor epsilon	NA	1.7
NM_010118	Early growth response 2	3.2	3.3
NM_030612	Nuclear factor of kappa light polypeptide gene enhancer in B-cells inhibitor, zeta	2.1	2.6
NM_018781	Early growth response 3	1.5	1.8
NM_008037	Fos like antigen 1	NA	1.6
NM_007625	Chromobox homolog 4 (Drosophila Pc class)	NA	-1.3
NM_010448	Heterogeneous nuclear ribonucleoprotein A/B	-1.3	-2.7
Signaling			
NM_013642	Dual specificity phosphatase 1 ³	2.0	1.3
	Dual specificity phosphatase 1 ³	2.0	1.3
NM_010090	Dual specificity phosphatase 2 ³	2.4	3.4
	Dual specificity phosphatase 2 ³	1.3	3.4
NM_011267	Phosphodiesterase 4 B cAMP ³	1.6	3.4
	Phosphodiesterase 4 B cAMP ³	1.4	2.8
NM_009421	Regulators of G protein signaling 16 ³	NA	1.9
	Regulators of G protein signaling 16 ³	NA	1.9
NM_031168	Radical S adenosyl methionine domain containing 2	NA	1.2
NM_007746	Mitogen activated protein kinase kinase kinase 8	1.3	1.3
NM_029466	ADP-ribosylation factor-like 5B	NA	1.3
NM_011113	Plasminogen activator, urokinase receptor	NA	1.8
NM_019654	Suppressor of cytokine signaling 5	NA	1.2
NM_009895	Cytokine inducible SH2-containing protein	NA	1.8
NM_009913	Chemokine (C-C motif) receptor 9	NA	1.8
NM_170701	CD40 antigen	NA	2.3
Apoptosis			
NM_009421	TNF receptor associated factor 1 ³	1.5	2.8
	TNF receptor associated factor 1 ³	1.5	3.0
NM_207652	TSC22 domain family, member 1	NA	2.3
NM_009344	Pleckstrin homology-like domain, family A, member 1	1.9	3.7
NM_010849	Myelocytomatosis oncogene	1.6	2.6
NM_009397	Tumor necrosis factor, alpha-induced protein 3 ³	2.0	1.8
	Tumor necrosis factor, alpha-induced protein 3 ³	1.6	1.5
NM_007987	Fas (TNF receptor superfamily member)	NA	2.0
Surface markers			
NM_009856	CD 83 antigen ³	1.8	3.1
	CD 83 antigen ³	1.8	3.0
Cell adhesion			
NM_008485	Laminin gamma 2	1.0	2.0
Cell cycle			
NM_007836	Growth arrest and DNA damage inducible 45 alpha ³	1.0	2.7
	Growth arrest and DNA damage inducible 45 alpha ³	1.2	3.2
NM_011121	Polo-like kinase 1 (Drosophila) ³	1.0	1.0
	Polo-like kinase 1 (Drosophila) ³	1.0	1.0
Cell proliferation (-ve)			
NM_011408	Schlafen 2	NA	2.0
Cell differentiation			
NM_008654	Myeloid differentiation primary response gene 116	1.4	NA
Transport			
NM_015747	Solute carrier family 20, member 1	NA	1.2
NM_010119	EH-domain containing 1 ³	NA	1.5
	EH-domain containing 1 ³	NA	1.5
NM_010215	Interleukin 4 induced 1	NA	2.7
Metabolism			
NM_013614	Ornithine decarboxylase, structural 1	NA	1.7

KDO, 2-keto-3-deoxyoctonic acid; MeV, Multiple Experiment Viewer; NA, not available.

³Transcription factors and genes involved in inflammation, immune responses, signal transduction, cell cycle regulation, cell proliferation, differentiation, metabolism, transport, cell adhesion, and apoptosis.

Supplementary Table 2. The genes up-regulated during IFN- β treatment in RAW 267.4 cells at 60 min and 120 min as analyzed by MeV

Function and GenBank identification	Gene name	60 min	120 min
Inflammation/Immune response			
NM_021274	Chemokine ligand (c-x-c) motif 10	3.0	5.0
NM_008331	Interferon induced protein with tetratricopeptide repeats 1	2.6	3.5
NM_021792	Interferon inducible GTPase 1	2.6	4.2
NM_010259	Guanylate nucleotide binding protein 1	2.7	4.5
NM_010260	Guanylate nucleotide binding protein 2	2.2	4.2
NM_008332	Interferon-induced protein with tetratricopeptide repeats 2 ^a	2.2	3.1
	Interferon-induced protein with tetratricopeptide repeats 2 ^a	2.6	2.0
NM_015783	ISG15 Ubiquitin like modifier ^a	NA	2.2
	ISG15 Ubiquitin like modifier ^a	1.1	2.3
NM_126166	Toll like receptor 3	1.2	1.9
NM_020557	Thymidylate kinase family, LPS inducible member	1.9	3.5
NM_008326	Immunity-related GTPase family, M	2.0	NA
NM_018738	Interferon gamma induced GTPase	1.8	3.0
NM_008330	Interferon gamma inducible protein 47	1.8	2.9
NM_013606	Myxovirus (influenza virus) resistance 2	2.4	2.9
NM_013655	Chemokine (c-x-c motif) ligand 12	1.6	NA
NM_011579	T cell specific GTPase ^a	1.4	2.6
	T cell specific GTPase ^a	2.7	4.3
NM_008327	Interferon activated gene 202B ^a	1.3	2.2
	Interferon activated gene 202B ^a	1.5	2.3
NM_153564	Guanylate nucleotide binding protein 5	2.4	3.9
NM_019494	Chemokine (c-x-c) motif ligand 11	2.1	3.2
NM_008328	Interferon activated gene 203	1.1	1.5
NM_008355	Interleukin 13	1.0	NA
NM_009735	Beta 2 microglobulin	1.1	1.7
NM_011163	Eukaryotic translation initiation factor 2-alpha kinase 2 ^a	NA	1.4
	Eukaryotic translation initiation factor 2-alpha kinase 2 ^a	NA	1.1
NM_013683	Transporter 1,ATP-binding cassette, subfamily B (MDR/TAP) ^a	1.0	1.5
	Transporter 1,ATP-binding cassette, subfamily B (MDR/TAP) ^a	NA	1.5
NM_008599	Chemokine (c-x-c) motif ligand 9	NA	1.5
NM_007778	Colony stimulating factor 1 (macrophage)	NA	1.3
NM_008198	Complement factor B	NA	1.9
NM_010501	Interferon-induced protein with tetratricopeptide repeats 3	1.4	3.5
NM_010531	Interleukin 18 binding protein	NA	1.3
NM_010186	Fc receptor, IgG, high affinity 1	NA	1.7
NM_170701	CD40 antigen	1.4	2.4
Apoptosis			
NM_007609	Caspase 4, apoptosis-related cysteine peptidase	1.0	1.2
NM_007987	FAS (TNF receptor superfamily member)	NA	1.7
NM_027835	Interferon induced with helicase C domain 1	NA	1.9
Transport			
NM_015751	ATP-binding cassette, sub-family E (OABP), member 1	NA	-1.5
Metabolism			
NM_013632	Purine-nucleoside phosphorylase ^a	NA	1.1
	Purine-nucleoside phosphorylase ^a	NA	1.1
Nuclease			
NM_011637	Three prime repair exonuclease 1	1.3	2.1
NM_030098	Ribonuclease, RNase A family, 6	NA	1.1
Protease related			
NM_011909	Ubiquitin specific peptidase 18	1.1	2.4
Cell differentiation			
NM_010612	Kinase insert domain protein receptor	NA	2.1
Signaling			
NM_009896	Suppressor of cytokine signaling 1 ^a	2.7	3.4
	Suppressor of cytokine signaling 1 ^a	2.3	2.7
NM_010175	FAS death domain associated protein	1.4	2.1
NM_031168	Radical S adenosyl methionine domain containing 2	3.2	3.8
NM_008519	Leukotriene B4 receptor 1	NA	1.3
NM_031874	RAB3D, member RAS oncogene family	NA	1.1
NM_009058	Ral guanine nucleotide dissociation stimulator	NA	1.1
Transcription			
NM_019963	Signal transducer and activator of transcription 2 ^a	1.2	2.3
	Signal transducer and activator of transcription 2 ^a	1.2	2.3
NM_009826	RB1-inducible coiled-coil 1	1.2	2.3
NM_028967	Basic leucine zipper transcription factor, ATF like 2	1.2	2.3
NM_010757	V-maf maculo aponeurotic fibrosarcoma oncogene Family, protein K (avian)	NA	1.1
NM_017405	Lipolysis stimulated lipoprotein receptor	NA	1.0
NM_002198	Interferon regulatory factor 1	1.0	2.7
NM_017375	Osteoclast stimulating factor 1	NA	3.8

IFN- β , interferon- β ; MeV, Multiple Experiment Viewer; NA, not available.^aGenes involved in inflammatory/immune responses, transcription, signaling, and apoptosis.

Supplementary Table 3. RAW 267.4 cells were treated with 8-Br for 60 min and 120 min and their gene expression levels were analyzed by oligonucleotide arrays

Function and GenBank identification	Gene name	60 min	120 min
Apoptosis			
NM_029083	DNA damage inducible transcript 4	4.1	3.9
NM_028778	NUAK family, SNF1-like kinase, 2	1.6	2.4
NM_010588	Jagged 2	NA	1.3
NM_013863	Bcl2 associated athnogene 3	1.0	1.0
NM_009421	Tnf receptor-associated factor 1	NA	2.4
NM_009344	Pleckstrin homology-like domain, family A, member 1	3.0	2.9
NM_010849	Myelocytomatosis oncogene	NA	-2.5
Metabolism			
NM_018757	Expressed in non-metastatic cells 6, protein	1.6	1.7
NM_013614	Ornithine decarboxylase, structural 1	1.3	2.1
NM_008517	Leukotriene A4 hydrolase	NA	1.2
NM_025907	Methyltransferase like 6	NA	2.2
NM_031159	Apolipoprotein B editing complex 1	-1.0	-1.2
Proteolysis			
NM_009373	Transglutaminase 2, C polypeptide ^a	1.4	3.0
	Transglutaminase 2, C polypeptide ^a	1.3	2.7
Immune response			
NM_019948	C-type lectin domain family 4, member e	NA	1.6
NM_010501	Interferon-induced protein with tetratricopeptide repeats 3	2.0	1.7
NM_138648	Oxidized low density lipoprotein (lectin-like) receptor 1	NA	1.3
NM_009704	Amphiregulin	4.7	NA
NM_008361	Interleukin 1 beta ^a	2.5	3.3
	Interleukin 1 beta ^a	3.6	4.1
NM_010554	Interleukin 1 alpha	NA	NA
NM_013693	Tumor necrosis factor ^a	-1.6	-1.7
	Tumor necrosis factor ^a	-1.6	-1.8
NM_013652	Chemokine ligand (C-X-C) 4 ^a	NA	-1.4
	Chemokine ligand (C-X-C) 4 ^a	NA	-1.9
NM_009404	Tumor necrosis factor (ligand) superfamily, member 9	NA	-1.3
Transport			
NM_015751	ATP-binding cassette, sub-family E (OABP), member 1	NA	1.7
Transcription factor			
NM_017373	Nuclear factor, interleukin 3, regulated	1.5	NA
NM_207652	TSC22 domain family, member 1	2.0	3.2
NM_010755	v-maf musculoaponeurotic fibrosarcoma oncogene family, protein F (avian)	2.3	2.0
NM_031494	Zinc finger protein 275	-1.7	-1.3
NM_010234	FOS B	3.3	3.5
NM_013498	cAMP responsive element modulator	2.5	3.9
NM_013613	Nuclear receptor subfamily 4, group A, member 2	4.3	4.4
NM_008690	Nuclear factor of kappa light polypeptide gene enhancer in B cells inhibitor, epsilon ^a	2.8	2.8
	Nuclear factor of kappa light polypeptide gene enhancer in B cells inhibitor, epsilon ^a	1.3	2.8
NM_010495	Inhibitor of DNA binding 1	1.8	NA
NM_013745	Nuclear fragile X mental retardation protein interacting protein 1	1.2	1.7
NM_009883	CCAAAT/enhancer binding protein (C/EBP) beta ^a	1.3	1.4
	CCAAAT/enhancer binding protein (C/EBP) beta ^a		
NM_010637	Kruppel-like factor 4 (gut)	2.0	2.2
NM_008037	Fos-like antigen 2	1.5	NA
NM_007625	Chromobox homolog 4 (Drosophila Pc class)	1.5	1.5
NM_010235	Fos like antigen 1	1.0	1.0
NM_008452	Kruppel like factor (lung)		
NM_009046	Avian reticuloendotheliosis viral (v-rel) oncogene related B	NA	2.2
Cell cycle			
NM_010831	SNF1-like kinase	1.4	1.3
NM_011121	Polo-like kinase 1 (Drosophila) ^a	NA	-1.3
	Polo-like kinase 1 (Drosophila) ^a	-1.0	-1.9
Surface markers			
NM_009856	CD 83 antigen ^a	2.4	3.1
	CD 83 antigen ^a	2.3	3.0
Signal transduction			
NM_019518	GRP1 (general receptor for phosphoinositides 1) associated scaffold protein	2.2	2.0
NM_011180	Pleckstrin homology, Sec7 and coiled-coil domains 1	1.7	1.6
NM_009217	Somatostatin receptor 2	NA	1.5
NM_010139	Eph receptor A2	NA	1.3
NM_029466	ADP-ribosylation factor-like 5B	1.3	1.6
NM_010555	Interleukin 1 receptor, type II	NA	2.1
NM_027897	Rhopilin, Rho GTPase binding protein 2	NA	2.2
NM_013642	Dual specificity phosphatase 1 ^a	3.3	2.2
	Dual specificity phosphatase 1 ^a	3.4	2.4
NM_010090	Dual specificity phosphatase 2 ^a	2.8	1.9
	Dual specificity phosphatase 2 ^a	2.8	2.0
NM_019840	Phosphodiesterase 4 B cAMP ^a	2.7	3.8
	Phosphodiesterase 4 B cAMP ^a	2.0	3.4
NM_011267	Regulators of G protein signaling 16 ^a	-2.2	-1.0
	Regulators of G protein signaling 16 ^a	-2.6	-1.2
NM_009421	TNF receptor associated factor 1	NA	2.4
NM_007746	Mitogen activated protein kinase kinase kinase 8	-1.0	-1.3
NM_008965	Prostaglandin E receptor 4 (subtype EP4)	-1.3	-1.9

8-Br, 8-bromoadenosine-3',5'-cyclic monophosphate; NA, not available.

^aGenes involved in transcription, signaling, inflammation, apoptosis, metabolism, proteolysis, transport, and cell cycle regulation.

Supplementary Table 4. Genes regulated upon KDO + IFN- β treatment in RAW 267.4 cells were grouped based on their functions

Function and GenBank identification	Gene name	60 min	120 min
Immune response/Inflammation			
NM_153564	Guanylate nucleotide binding protein 5	3.7	5.6
NM_019494	Chemokine (c-x-c) motif ligand 11	4.6	5.7
	Chemokine (c-x-c) motif ligand 11	4.4	NA
NM_010259	Guanylate nucleotide binding protein 1	4.1	5.7
	Guanylate nucleotide binding protein 1	4.3	6.1
NM_011579	T cell specific GTPase	3.7	5.8
	T cell specific GTPase	2.2	2.7
NM_008599	Chemokine (c-x-c) motif ligand 9	4.2	5.0
NM_021792	Interferon inducible GTPase 1	4.2	4.8
NM_008331	Interferon induced protein with tetratricopeptide repeats 1	3.7	4.7
NM_013655	Chemokine (c-x-c motif) ligand 12	3.3	4.6
	Chemokine (c-x-c motif) ligand 12	3.2	3.9
NM_008332	Interferon induced protein with tetratricopeptide repeats 2	2.9	5.0
	Interferon induced protein with tetratricopeptide repeats 2	3.5	3.5
NM_020557	Thymidylate kinase family,LPS inducible member	3.0	5.3
NM_021274	Chemokine (c-x-c motif) ligand 10	4.3	NA
NM_008328	Interferon activated gene 203	1.2	2.3
NM_009735	Beta 2 microglobulin	2.3	2.1
NM_008326	Immunity related GTPase	2.0	NA
NM_018738	Interferon gamma induced GTPase	2.0	3.7
NM_008330	Interferon gamma inducible GTPase 47	2.3	3.9
NM_013683	Transporter 1,ATP-binding cassette, subfamily B (MDR/TAP)	1.8	2.5
	Transporter 1,ATP-binding cassette, subfamily B (MDR/TAP)	1.1	1.9
NM_007778	Colony stimulating factor 1 (macrophage)	NA	1.7
NM_008361	Interleukin 1 beta	1.6	5.0
	Interleukin 1 beta	2.8	5.6
NM_019840	Phosphodiesterase 4B, cAMP specific	1.9	2.6
NM_010501	Interferon induced protein with tetratricopeptide repeats 3	3.6	3.6
NM_010548	Interleukin 10	NA	3.1
	Interleukin 10	NA	2.9
NM_010260	Guanylate nucleotide binding protein 2	1.4	2.9
NM_009971	Colony stimulating factor 3 (granulocyte)	NA	2.7
NM_009778	Complement component 3	NA	2.7
NM_010846	Myxovirus (influenza virus) resistance 1	2.5	4.6
NM_027835	Interferon induced with helicase C domain 1	NA	3.5
NM_008360	Interleukin 18	NA	1.9
NM_010171	Coagulation factor III	3.7	4.3
NM_170701	CD 40 antigen	2.9	4.7
NM_009856	CD 83 antigen	2.6	3.7
NM_008327	Interferon activated gene 202B	1.3	3.5
	Interferon activated gene 202B	1.3	3.6
NM_031168	Interleukin 6	2.9	4.4
NM_008355	Interleukin 13	1.4	1.5
NM_019583	Interleukin 17receptor B	NA	2.4
NM_015783	ISG 15 ubiquitination like modifier	1.3	3.2
NM_013652	Chemokine (C-C) motif ligand 4	2.9	4.3
NM_010554	Interleukin 1 alpha	2.9	4.6
NM_013693	Tumor necrosis factor	3.3	3.4
Transcription factor			
NM_008320	Interferon regulatory factor 8	1.6	2.4
NM_019963	Signal transducer and activator of transcription 2	1.5	2.6
NM_028967	Basic leucine zipper transcription factor, ATF like 2	2.7	3.2
NM_010757	V-maf maculo aponeurotic fibrosarcoma oncogene Family, protein K (avian) C22:C54	1.3	1.8
	V-maf maculo aponeurotic fibrosarcoma oncogene Family, protein K (avian)	2.1	1.8
NM_018781	Early growth response 3	1.1	1.3
NM_013692	Kruppel-like factor 10	NA	1.7
NM_013674	Interferon regulatory factor 4	NA	1.5
NM_008689	Nuclear factor of kappa light chain gene enhancer in B-cells 1, p105	NA	1.3
NM_007498	Activating transcription factor 3	2.3	2.5
NM_011141	POU domain, class 3, transcription factor 1	NA	1.7
NM_213659	Signal transducer and activator of transcription 3	NA	1.6
NM_002198	Interferon regulatory factor 1	4.1	NA
NM_008036	FBJ osteosarcoma Oncogene B	NA	1.1
NM_008690	Nuclear factor of Kappa light polypeptide gene enhancer In B cells inhibitor , epsilon	NA	1.5
NM_010849	Myelocytomatosis oncogene	NA	1.4
NM_013498	cAMP responsive element Modulator	NA	1.8
Signaling			
NM_007746	Mitogen activated protein kinase kinase kinase 8	3.6	2.7
NM_009896	Suppressor of cytokine signaling 1	4.4	3.3
	Suppressor of cytokine signaling 1	3.7	2.2
NM_031168	Radical S adenosyl methionine domain containing 2	4.5	NA
NM_029499	Membrane spanning 4 domains, subfamily A, member C	1.6	2.6
NM_010175	FAS death domain associated protein	1.9	3.2
NM_008519	Leukotriene B4 receptor 1	1.7	1.2
NM_009895	Cytokine inducible SH2 containing protein	1.3	1.7
NM_029466	ADP-ribosylation factor-like 5B	NA	1.6
NM_025658	Membrane-spanning 4-domains, subfamily A, member 4D	NA	1.9
NM_021454	CDC42 effector protein (Rho GTPase binding) 5	1.4	NA

NM_018851	SAM domain and HD domain, 1	NA	1.6
NM_023622	Ral guanine nucleotide dissociation stimulator like 1	NA	1.5
NM_010090	Dual specificity phosphatase 2	NA	1.7
NM_011227	RAB20, member RAS oncogene family	1.1	1.5
NM_013642	Dual specificity phosphatase 1	1.4	1.8
NM_030701	G protein coupled receptor 109A	1.9	3.0
NM_008011	Fibroblast growth factor receptor 4	NA	1.5
NM_017466	Chemokine (c-c motif) receptor like 2	4.0	4.8
Apoptosis			
NM_007987	Fas (TNF receptor superfamily member)	NA	3.2
NM_007536	B-cell leukemia/lymphoma 2 related protein A1d	NA	1.5
NM_009397	Tumor necrosis factor, alpha-induced protein 3	3.0	1.8
NM_008655	Growth arrest and DNA damage inducible 45beta	1.7	1.9
NM_007464	Baculoviral IAP repeat containing 3	NA	1.8
NM_009805	CASP 8 and FADD like apoptosis regulator	1.2	2.4
NM_009174	Seven in absentia 2	NA	1.4
Stress			
NM_016737	Stress induced phosphoprotein 1	NA	3.9
Cell adhesion			
NM_011521	Syndecan 4	NA	1.5
NM_010612	Kinase insert domain protein receptor	NA	1.7
Proteolysis			
NM_008608	Matrix metalloproteinase 14 (membrane-inserted)	NA	1.4
Cell cycle arrest			
NM_007669	Cyclin-dependent kinase inhibitor 1A	1.3	1.4
Metabolism			
NM_013632	Purine nucleoside phosphorylase	NA	1.8
NM_008384	Inositol polyphosphate-1-phosphatase	NA	2.1
NM_021542	Potassium channel subfamily k member 5	NA	2.1
Cell differentiation			
NM_008654	Myeloid differentiation primary response gene 116	2.4	1.3
Transport			
NM_145983	Potassium voltage-gated channel, shaker-related Subfamily member 5	NA	3.8

KDO, 2-keto-3-deoxyoctonic acid; IFN- β , interferon- β ; NA, not available.

Supplementary Table 5. Genes regulated upon KDO + 8-Br treatment in RAW 267.4 cells were grouped based on their functions

Function and GenBank identification	Gene name	60 min	120 min
Transcription factor			
NM_008690	Nuclear factor of kappa light Polypeptide gene enhancer in B cells Inhibitor , epsilon	3.3	3.5
NM_008036	FBJ osteosarcoma oncogene B	4.1	3.7
NM_013613	Nuclear receptor factor, Subfamily 4, Group A, member 2	4.5	3.9
NM_013498	cAMP responsive element modulator	3.0	3.6
NM_015743	Nuclear receptor factor, Subfamily 4, Group A, member 3	NA	1.1
NM_007548	PR domain containing 1, with ZNF domain	1.6	NA
	PR domain containing 1, with ZNF domain	1.9	2.9
NM_009883	CCAAAC/enhancer binding protein (c/EBP) beta	1.3	1.9
NM_008723	Nucleoplasmin 3	1.4	NA
NM_018781	Early growth response 3	1.5	NA
NM_013674	Interferon regulatory factor 4	NA	2.1
NM_010757	V maf maculoaponeurotic fibrosarcoma oncogene family , Protein F avain	2.8	3.3
NM_007498	Activating transcription factor 3	NA	-1.9
NM_008321	Inhibitor of DNA binding 3	1.5	NA
NM_010637	Kruppel like factor 4 (gut)	1.6	1.8
NM_011809	E26 avian leukemia oncogene 2, 3' domain	1.8	1.4
NM_024406	Fatty acid binding protein 4, adipocyte	NA	2.0
Signaling			
NM_019819	Dual specificity phosphatase 14	2.4	2.4
NM_019925	G protein coupled receptor 132	2.8	2.8
NM_009962	G protein coupled receptor 44	NA	NA
NM_007539	Bradykinin receptor, beta 1	NA	NA
NM_011180	Pleckstrin homology, sec 7, coiled coil domains 1	1.4	1.4
NM_019518	GRP1 (general factor for phosphoinositides 1) associated scfold protein	1.8	1.8
NM_011065	Period homolog 1 (Drosophila)	1.3	1.3
NM_008152	G-protein coupled receptor 65	NA	NA
NM_011267	Phosphodiesterase 4B, cAMP specific	3.5	3.5
NM_010098	Opsin (encephalopsin)	NA	NA
NM_021454	CD42 effector protein (Rho GTPase binding) 2	NA	NA
NM_018882	G protein coupled receptor 56	1.2	1.2
	G protein coupled receptor 56	1.2	1.2
NM_010090	Dual specificity phosphatase 2	2.2	2.2
NM_010317	Guanine nucleotide binding protein (G protein gamma 4 subunit)	NA	NA
NM_011227	RAB 20, member RAS oncogene family	1.5	1.5
NM_008011	Fibroblast growth factor receptor 4	NA	NA
Apoptosis			
NM_029083	DNA damage inducible transcript 4	4.0	4.4
NM_013863	Bcl 2 associated athanogene 3	1.1	NA
NM_007987	Fas (TNF receptor super family member)	NA	1.2
NM_007536	B cell leukemia/lymphoma 2 related protein A1d	NA	1.3
XM_887085	TNF receptor associated factor 5	NA	1.0
NM_019952	Cardiotrophin-like cytokine factor 1	NA	1.2
NM_009397	Tumor necrosis factor alpha induced protein 3	3.1	2.2
NM_008655	Growth arrest and DNA-damage-inducible 45 beta	1.3	2.0
NM_007464	Baculoviral IAP repeat-containing 3	1.2	1.8
NM_009805	CASP 8 and FADD like apoptosis regulator	NA	1.1
Inflammation/ Immune response			
NM_008361	Interleukin 1 beta	5.6	5.5
	Interleukin 1 beta	6.2	5.7
NM_010501	Interferon induced protein with tetratricopeptide repeats 3	4.0	4.4
NM_010548	Interleukin 10	3.3	3.2
	Interleukin 10	3.0	3.0
NM_009855	CD80 antigen	1.7	1.7
NM_138648	Oxidized low density lipoprotein (lectin like) receptor 1	NA	2.1
NM_009971	Colony stimulating factor 3	4.3	5.0
NM_031168	Interleukin 6	2.1	3.5
NM_010171	Coagulation factor III	2.7	NA
NM_009856	CD83 antigen	3.6	3.9
NM_013654	Chemokine ligand 7	1.9	2.4
NM_019583	Interleukin 17 receptor B	NA	2.0
NM_013693	Tumor necrosis factor	2.2	1.7
NM_010554	Interleukin 1 alpha	4.5	5.3
Coagulation			
NM_009378	Chemokine (C-X-C motif) ligand 4	3.5	4.9
NM_011171	Protein C receptor, endothelial	2.3	3.7
	Protein C receptor, endothelial	2.2	3.3

Proteolysis			
NM_009373	Transglutaminase 2, C polypeptide	2.7	2.5
NM_008517	Leukotriene A4 hydrolase	1.2	1.6
Defence response			
NM_028071	Chemokine (c-x-c motif) ligand 12	1.1	NA
	Chemokine (c-x-c motif) ligand 12	1.0	NA
Transport			
NM_133977	Transferrin	1.1	1.3
NM_175933	Peroxisome biogenesis factor 5	2.0	3.5
NM_010329	Podoplanin		
Protein folding			
NM_013559	Heat shock protein 110	1.4	1.5
Cell cycle			
NM_007669	Cyclin dependent kinase inhibitor 1A p21	1.3	1.6
NM_013562	Interferon related developmental regulator 1	1.8	2.2
Cell growth			
NM_001025257	Vascular endothelial growth factor	3.4	3.9
Protease			
NM_008608	Matrix metalloproteinase 14 (membrane related)	NA	1.4
Metabolism			
NM_013614	Ornithine decarboxylase structural 1	1.9	3.0
NM_008384	Inositol polyphosphate-1-phosphatase	NA	2.9
NM_021542	Potassium channel, subfamily K member 5	NA	2.9
NM_025907	Methyltransferase like 6	NA	2.8
NM_080633	Aconitase 2, mitochondrial	NA	3.5

KDO, 2-keto-3-deoxyoctonic acid; 8-Br, 8-bromoadenosine-3',5'-cyclic monophosphate; NA, not available.

Supplementary Table 6. Triple ligand stimulation (KDO + IFN- β + 8-Br) of RAW 264.7

Function and GenBank identification	Gene name	60 min	120 min
Signaling			
NM_009896	Suppressor of cytokine signaling 1	4.3	3.8
	Suppressor of cytokine signaling 1	3.7	2.9
NM_008519	Leukotriene B4 receptor 1	1.9	1.6
NM_029499	Membrane spanning 4 domains, Subfamily A, member 4c	1.7	2.6
NM_007746	Mitogen activated protein kinase kinase 8	3.3	2.5
NM_011065	Period homolog 1, drosophila	1.1	1.4
NM_011267	Regulators of G protein signaling 16	-2.6	NA
	Regulators of G protein signaling 16	-2.1	NA
NM_007577	Complement component 5a, receptor 1	NA	1.2
NM_009912	Chemokine (c-c motif) receptor 1	1.9	2.1
NM_012051	Ets variant gene 3	1.1	NA
NM_019840	Phosphodiesterase 4 B cAMP specific	3.8	3.8
	Phosphodiesterase 4 B cAMP specific	4.2	4.5
NM_017466	Chemokine (C-C motif) receptor-like 2	5.5	5.2
NM_011113	Plasminogen activator, urokinase receptor	1.1	1.9
NM_029035	SplA/ryanodine receptor domain and SOCS box containing 1	1.2	1.5
NM_009058	Ral guanine nucleotide dissociation stimulator	NA	2.0
NM_010025	Doublecortin	1.0	1.3
NM_009372	TG interacting factor	1.2	1.0
NM_010186	Fc receptor, IgG, high affinity I	1.0	2.3
NM_019777	Inhibitor of kappa B kinase epsilon	NA	1.6
NM_023764	Toll interacting protein	NA	1.7
NM_029466	ADP-ribosylation factor-like 5B	1.5	3.0
NM_019925	G protein coupled receptor 132	2.7	2.8
NM_018782	Calcitonin receptor-like	1.4	NA
NM_008152	G protein coupled receptor 65	NA	2.5
NM_018851	SAM domain and HD domain, 1	NA	1.6
NM_010090	Dual specificity phosphatase 2	3.7	3.1
	Dual specificity phosphatase 2	3.8	3.4
NM_010415	Heparin binding EGF like growth factor	2.5	2.4
NM_028544	Ras interacting protein 1	NA	1.7
NM_008413	Janus kinase 2	NA	2.3
NM_010583	IL2-inducible T-cell kinase	NA	1.2
Calcium binding			
NM_010807	MARCKS-like 1	1.4	1.8
NM_007493	Asialoglycoprotein receptor 2	NA	1.0
Complement			
NM_011113	Plasminogen activator, urokinase receptor	1.3	1.6
NM_008871	Serine (or cysteine) peptidase inhibitor, clade E, member 1	1.5	3.4
NM_009971	Coagulation factor III	5.9	4.9
Defence response			
NM_207648	Histocompatibility 2, Q region, Locus 6	1.6	1.5
NM_010393	Histocompatibility 2, Q region, Locus 5	1.9	1.9
Cell proliferation			
NM_027514	Poliovirus receptor	1.5	NA
	Poliovirus receptor	1.7	1.4
Proteolysis			
NM_009373	Transglutaminase 2,C polypeptide	2.8	3.3
	Transglutaminase 2,C polypeptide	2.5	3.5
Metabolism			
NM_013614	Ornithine decarboxylase, structural 1	1.7	2.9
NM_027442	D-aspartate oxidase	NA	2.2
NM_021319	Peptidoglycan recognition protein 2	1.4	1.4
NM_011673	UDP-glucose ceramide glucosyltransferase	1.1	1.1
NM_008317	Hyaluronoglucosaminidase 1	-1.4	-2.2
NM_026428	Dicarbonyl L-xylulose reductase	NA	1.7
NM_007494	Argininosuccinate synthetase 1	NA	1.4
NM_025907	Methyltransferase like 6	NA	1.7
NM_023475	Serine hydrolase like	NA	1.4
NM_013529	Glutamine fructose-6-phosphate transaminase 2	1.2	NA
NM_013820	Hexokinae 2	NA	1.6
NM_009698	Adenine phosphoribosyl transferase	-1.4	NA
NM_008384	Inositol polyphosphate-1-phosphatase	NA	4.2
NM_011198	Prostaglandin-endoperoxide synthase 2	3.9	NA
NM_008847	Phosphatidylinositol-4-phosphate 5-kinase, type 1 beta	1.4	1.1
NM_134102	Phospholipase A1 member A	NA	1.4
Anti-apoptosis			
NM_007464	Baculoviral IAP repeat-containing 3	1.5	2.1
NM_008654	Myeloid differentiation primary response gene 116	2.9	1.6
others			
NM_008538	Myristoylated alanine rich protein kinase C substrate	NA	1.9
NM_008842	Proviral integration site 1	2.2	3.5
	Proviral integration site 1	2.7	3.8
NM_028800	Serine/therionine kinase40	NA	2.2

Growth arrest			
NM_008380	Inhibin beta-A	NA	1.3
NM_007609	Schflan 3	1.7	2.6
Apoptosis			
NM_009805	CASP8 and FADD-like apoptosis regulator	NA	1.8
NM_008655	Growth arrest and DNA damage inducible 45 beta	2.0	2.4
	Growth arrest and DNA damage inducible 45 beta	1.9	2.3
NM_007536	B-cell leukemia/lymphoma 2 related protein A1d	NA	1.4
	B-cell leukemia/lymphoma 2 related protein A1d	NA	1.7
NM_007987	FAS (TNF receptor superfamily member)	NA	2.4
NM_013464	Aryl hydrocarbon receptor		
NM_008842	Proviral integration site 1	2.6	3.5
NM_007570	B-cell translocation gene 2, anti-proliferative	5.1	NA
Surface markers			
NM_009856	CD 83 antigen	3.7	4.3
	CD 83 antigen	3.6	4.0
NM_008536	Transmembrane 4 superfamily member 1	1.6	2.2
Transcription			
NM_013613	Nuclear receptor factor, Subfamily 4, Group A, member 2	3.7	3.1
NM_013498	cAMP responsive element modulator	3.1	3.7
NM_007498	Activating transcription factor 3	1.4	NA
NM_008320	Interferon regulatory factor 8	1.2	1.8
NM_007829	FAS death domain associated protein	1.7	3.0
NM_016767	Basic leucine zipper transcription factor , ATF like 2	2.6	2.8
NM_033601	B-cell leukemia/lymphoma 3	1.2	1.9
NM_019408	Nuclear factor of kappa light polypeptide gene enhancer in B cells 2, p49/p100	NA	1.6
NM_010118	Early growth response 2	3.6	1.1
NM_007913	Early growth response 1	4.4	2.6
NM_030612	Nuclear factor of kappa light polypeptide gene enhancer in B-cells inhibitor, zeta	2.7	2.7
NM_010755	v-maf musculoaponeurotic fibrosarcoma oncogene family, protein F (avian)	3.0	2.7
NM_009895	Cytokine inducible SH2-containing protein	2.5	2.7
NM_019980	LPS-induced TN factor	NA	1.3
NM_021397	Zinc finger and BTB domain containing 32	NA	1.3
NM_010235	FOS like antigen 1	NA	1.8
NM_008535	Lymphoblastomic leukemia	NA	-1.6
NM_016850	Interferon regulatory factor 7	NA	1.6
NM_030887	Jun dimerization protein 2	NA	2.3
NM_010751	MAX dimerization protein 1	1.7	3.1
NM_213659	Signal transducer and activator of transcription 3	NA	1.8
	Signal transducer and activator of transcription 3	1.0	1.8
NM_019963	Signal transducer and activator of transcription 2	NA	2.8
NM_002198	Interferon regulatory factor 1	3.9	1.2
NM_009046	Avian reticuloendotheliosis viral (v-rel) oncogene related B	NA	1.9
NM_017373	Nuclear factor, interleukin 3, regulated	2.7	NA
NM_007614	Catenin (cadherin associated protein), beta 1	1.0	1.9
NM_010234	FOS B	3.2	3.3
NM_183091	Nuclear factor of kappa light polypeptide gene enhancer in B-cells inhibitor, epsilon	2.5	3.3
Nuclease			
NM_011637	Three prime repair exonuclease 1	1.9	2.7
Proteases			
NM_013640	Proteasome (prosome, macropain) subunit, beta type 10	NA	1.1
	Proteasome (prosome, macropain) subunit, beta type 10	NA	1.2
NM_011046	Furin (paired basic amino acid cleaving enzyme)	NA	1.3
Transport			
NM_015747	Solute carrier family 20, member 1	NA	1.1
NM_021450	Transient receptor potential cation channel, subfamily M, member 7	NA	4.1
NM_026936	Oxidase assembly 1-like	NA	3.3
NM_181411	Aftiphilin	NA	1.2
Cell cycle arrest			
NM_021878	Jumonji, AT rich interactive domain 2	NA	2.3
Immunity/Inflammation			
NM_019932	Chemokine (C-X-C motif) ligand 4	2.5	3.1
	Chemokine (C-X-C motif) ligand 4	2.4	3.2
NM_013693	Tumor necrosis factor	2.4	3.9
NM_031168	Interleukin 6	6.1	5.9
NM_021792	Interferon inducible GTPase 1	4.0	4.3
NM_008599	Chemokine (c-x-c) motif ligand 9	4.3	4.0
NM_013655	Chemokine (c-x-c motif) ligand 12	3.2	3.2
	Chemokine (c-x-c motif) ligand 12	2.0	2.3
NM_009855	CD80 antigen	1.4	1.6
NM_008328	Interferon activated gene 203	1.2	1.7
NM_008355	Interleukin 13	1.5	1.5
NM_013683	Transporter 1, ATP binding cassette, subfamily B (MDR/TAP)	3.1	4.8
NM_011579	T cell specific GTPase	3.1	4.8
NM_008326	Immunity related GTPase family, M	2.3	NA
NM_008332	Interferon induced protein with tetratricopeptide repeats 2	2.7	3.9
NM_029000	GTPase, very large interferon inducible 1	1.4	2.7
NM_010501	Interferon induced protein with tetratricopeptide repeats 3	3.6	NA

NM_008331	Interferon induced protein with tetratricopeptide repeats 1	3.4	4.4
NM_020557	Thymidylate kinase family, LPS inducible member	3.3	5.3
NM_153564	Guanylate nucleotide binding protein 5	3.6	5.4
NM_010259	Guanylate nucleotide binding protein 1	4.6	6.6
NM_010260	Guanylate nucleotide binding protein 2	3.9	5.1
NM_019494	Chemokine (c-x-c) motif ligand 11	5.2	5.3
	Chemokine (c-x-c) motif ligand 11	4.5	5.0
NM_008361	Interleukin 1 beta	6.1	6.1
	Interleukin 1 beta	5.4	NA
NM_010554	Interleukin 1 alpha	5.3	5.4
NM_009397	Tumor necrosis factor, alpha-induced protein 3	1.9	1.9
NM_011157	Interleukin 20	2.0	2.3
NM_008501	Leukimia inhibitory factor	2.3	3.9
	Leukimia inhibitory factor	3.9	4.1
NM_008348	Interleukin 10 receptor, alpha	2.2	2.0
NM_007778	Colony stimulating factor 1(macrophage)	NA	1.2
XM_127883	Immunoresponsive gene 1	2.9	4.8
NM_010548	Interleukin 10	3.2	3.0
NM_008360	Interleukin 18	NA	1.9
NM_008357	Interleukin 15	1.5	4.1
NM_011610	Tumor necrosis factor receptor superfamily, member 1b	NA	1.9
NM_010740	CD93 antigen	NA	3.3
NM_013606	Myxovirus (influenza virus) resistance 2	3.5	3.5
NM_019583	Interleukin 17 receptor B	NA	2.4
NM_001008700	Interleukin 4 receptor alpha	NA	1.7

KDO, 2-keto-3-deoxyoctonic acid; IFN- β , interferon- β ; 8-Br, 8-bromoadenosine-3',5'-cyclic monophosphate; NA, not available.