

Toll-like Receptors

Toll-like receptors are a family of type I transmembrane pattern recognition receptors (PRRs) that sense invading pathogens or endogenous damage signals and initiate the innate and adaptive immune response. There are ten functional TLRs in human (TLR1–10) and twelve in mice (TLR1–9, 11–13). Various combinations of TLRs are expressed by different subsets of immune and non-immune cell types such as monocytes, macrophages, dendritic cells, neutrophils, B cells, T cells, fibroblasts, endothelial cells, and epithelial cells. Of the human TLRs, TLR1, 2, 4, 5, 6, and 10 are expressed on the cell surface and primarily recognize microbial membrane and/or cell wall components, while TLR3, 7, 8, and 9 are expressed in the membranes of endolysosomal compartments and recognize nucleic acids. TLRs have a variable number of ligand-sensing, leucine-rich repeats (LRR) at their N-terminal ends and a cytoplasmic Toll/IL-1 R (TIR) domain. The TIR domain mediates interactions between TLRs and adaptor proteins involved in regulating TLR signaling including MyD88, TRIF, TRAM, and TIRAP/MAL. Signaling pathways activated downstream of these adaptor molecules promote the expression of pro-inflammatory cytokines, chemokines, and type I and type III interferons. Although TLRs provide protection against a wide variety of pathogens, inappropriate or unregulated activation of TLR signaling can lead to chronic inflammatory and autoimmune disorders.



TLRsAgonist(s)SourceTLR1/ TLR2Triacyl lipopeptidesBacteriaFlagLipoproteinsMultiple PathogensPeptidoglycan (PGN)BacteriaPorinsBacteriaPorinsBacteriaGPI-mucinFungiEnvelope glycoproteinsVirusesTLR2/ TLR6Diacyl lipopeptidesBacteriaDiacyl lipopeptidesBacteriaTLR3/ TLR3Double-stranded RNAVirusesFungi (LPS)Synthetic analog of double- stranded RNAFUR4Glycoinositol- phospholipidsBacteriaFUR4Glycoinositol- phospholipidsProtozoaFurelope glycoproteinsVirusesTLR3FlagellinBacteriaTLR4Single-stranded RNAVirusesTLR5FlagellinBacteriaTLR5Single-stranded RNAVirusesTLR4Single-stranded RNAVirusesTLR5FlagellinBacteriaTLR6DNAVirusesTLR7Single-stranded RNAVirusesTLR8Single-stranded RNAVirusesTLR9Mitochondrial DNAEndogenousTLR10UnknownUnknown	Toll-like Receptors (TLRs)								
TLR1/ TLR2Triacyl lipopeptidesBacteriaLipoproteinsMultiple PathogensPeptidoglycan (PGN)BacteriaPorinsBacteriaZymosanFungiβ-GlycanFungiGPI-mucinProtozoaEnvelope glycoproteinsVirusesTLR2/ TLR6Diacyl lipopeptidesBacteriaDouble-stranded RNAVirusesTLR3Poly (I:C)Synthetic analog of double- stranded RNATLR4Glycoinositol- phospholipidsProtozoaFuvelope glycoproteinsVirusesTLR3Glycoinositol- phospholipidsProtozoaTLR4FingellinBacteriaTLR5FlagellinBacteriaTLR5Single-stranded RNAVirusesTLR3Single-stranded RNAVirusesTLR4Most-derived HMGB1 and HSPsEndogenousTLR5FlagellinBacteriaTLR6Single-stranded RNAVirusesTLR7Single-stranded RNAVirusesTLR8Single-stranded RNAVirusesTLR9Mitochondrial DNAEndogenousTLR10UnknownUnknown	TLRs	Agonist(s)	Source						
LipoproteinsMultiple PathogensPeptidoglycan (PGN)BacteriaPorinsBacteriaZymosanFungiβ-GlycanFungiβ-GlycanProtozoaEnvelope glycoproteinsBacteriaTLR2/ TLR6Diacyl lipopeptidesBacteriaDouble-stranded RNAVirusesTLR3Poly (I:C)Synthetic analog of double- stranded RNATLR3Glycoinositol- phospholipidsBacteriaGlycoinositol- phospholipidsProtozoaTLR4Glycoinositol- and HSPsVirusesTLR5FlagellinBacteriaTLR4Single-stranded RNAVirusesTLR5Single-stranded RNAVirusesTLR4Most-derived HMGB1 and HSPsEndogenousTLR5Single-stranded RNAVirusesTLR6Single-stranded RNAVirusesTLR7Single-stranded RNAVirusesTLR3Mitochondrial DNAEndogenousTLR40UnknownUnknown	TLR1/ TLR2	Triacyl lipopeptides	Bacteria						
Peptidoglycan (PGN)BacteriaPorinsBacteriaPorinsFungiβ-GlycanFungiβ-GlycanProtozoaEnvelope glycoproteinsWirusesTLR2/ TLR6Diacyl lipopeptidesBacteriaDouble-stranded RNAVirusesTLR3Poly (I:C)Synthetic analog of double- 		Lipoproteins	Multiple Pathogens						
PorinsBacteriaILR2ZymosanFungiβ-GlycanFungiGPI-mucinProtozoaEnvelope glycoproteinsWirusesTLR2/ TLR6Diacyl lipopeptidesBacteriaDiacyl lipopeptidesBacteriaTLR3Double-stranded RNAVirusesTLR3Poly (I:C)Synthetic analog of double- 		Peptidoglycan (PGN)	Bacteria						
TLR2 Zymosan Fungi β-Glycan Fungi GPI-mucin Protozoa Envelope glycoproteins Viruses TLR2/ TLR6 Diacyl lipopeptides Bacteria Diacyl lipopeptides Bacteria TLR3 Double-stranded RNA Viruses TLR3 Poly (I:C) Synthetic analog of double- stranded RNA TLR3 Glycoinositol- phospholipids Bacteria Glycoinositol- phospholipids Protozoa Envelope glycoproteins Viruses TLR4 Envelope glycoproteins Viruses TLR4 Single-stranded RNA Viruses TLR5 Flagellin Bacteria TLR7 Single-stranded RNA Viruses TLR8 Single-stranded RNA Viruses TLR9 Unmethylated CpG DNA Protozoa Viruses Mitochondrial DNA Endogenous Viruses Viruses Viruses		Porins	Bacteria						
β-Glycan Fungi GPI-mucin Protozoa Envelope glycoproteins Viruses TLR2/ TLR6 Diacyl lipopeptides Bacteria Diacyl lipopeptides Bacteria TLR2/ TLR6 Diacyl lipopeptides Bacteria Double-stranded RNA Viruses TLR3 Double-stranded RNA Viruses TLR3 Poly (I:C) Synthetic analog of double- stranded RNA Lipopolysaccharide (LPS) Bacteria Glycoinositol- phospholipids Protozoa Envelope glycoproteins Viruses TLR5 Flagellin Bacteria TLR7 Single-stranded RNA Viruses TLR8 Single-stranded RNA Viruses TLR9 Minethylated CpG DNA Bacteria Protozoa Viruses Protozoa TLR9 Mitochondrial DNA Endogenous TLR10 Unknown Unknown	TLR2	Zymosan	Fungi						
GPI-mucinProtozoaEnvelope glycoproteinsVirusesTLR2/ TLR6Diacyl lipopeptidesBacteriaDiacyl lipopeptidesBacteriaTLR3Double-stranded RNAVirusesTLR3Poly (I:C)Synthetic analog of double- stranded RNALipopolysaccharide (LPS)BacteriaGlycoinositol- phospholipidsProtozoaEnvelope glycoproteinsVirusesTLR3FagellinBacteriaTLR4Single-stranded RNAVirusesTLR5FlagellinBacteriaTLR7Single-stranded RNAVirusesTLR8Single-stranded RNAVirusesTLR9Unmethylated CpG DNAProtozoaTLR10UnknownUnknown		β-Glycan	Fungi						
Envelope glycoproteinsVirusesTLR2/ TLR6Diacyl lipopeptidesBacteriaLipoteichoic acid (LTA)BacteriaTLR3Double-stranded RNAVirusesTLR3Poly (I:C)Synthetic analog of double- stranded RNALipopolysaccharide (LPS)BacteriaGlycoinositol- phospholipidsProtozoaEnvelope glycoproteinsVirusesTLR3FlagellinBacteriaTLR5FlagellinBacteriaTLR7Single-stranded RNAVirusesTLR8Single-stranded RNAVirusesTLR9Unmethylated CpG DNAProtozoaTLR10UnknownUnknown		GPI-mucin	Protozoa						
TLR2/ TLR6Diacyl lipopeptidesBacteriaLipoteichoic acid (LTA)BacteriaTLR3Double-stranded RNAVirusesTLR3Poly (I:C)Synthetic analog of double- stranded RNALipopolysaccharide (LPS)BacteriaGlycoinositol- phospholipidsProtozoaEnvelope glycoproteinsVirusesTLR5FlagellinBacteriaTLR7Single-stranded RNAVirusesTLR8Single-stranded RNAVirusesTLR9Unmethylated CpG 		Envelope glycoproteins	Viruses						
TLR6Lipoteichoic acid (LTA)BacteriaDouble-stranded RNAVirusesTLR3Poly (I:C)Synthetic analog of double-stranded RNAPoly (I:C)Synthetic analog of double-stranded RNALipopolysaccharide (LPS)BacteriaGlycoinositol-phospholipidsProtozoaEnvelope glycoproteinsVirusesHost-derived HMGB1 and HSPsEndogenousTLR7Single-stranded RNAVirusesTLR8Single-stranded RNAVirusesTLR9Unmethylated CpG DNAProtozoaMitochondrial DNAEndogenousTLR10UnknownUnknown	TLR2/	Diacyl lipopeptides	Bacteria						
Double-stranded RNAVirusesTLR3Double-stranded RNASynthetic analog of double- stranded RNALipopolysaccharide (LPS)BacteriaGlycoinositol- 	TLR6	Lipoteichoic acid (LTA)	Bacteria						
TLR3Poly (I:C)Synthetic analog of double- stranded RNALipopolysaccharide (LPS)BacteriaGlycoinositol- phospholipidsProtozoaEnvelope glycoproteinsVirusesHost-derived HMGB1 and HSPsEndogenousTLR5FlagellinBacteriaTLR7Single-stranded RNAVirusesTLR8Single-stranded RNAVirusesTLR9Unmethylated CpG DNAProtozoaMitochondrial DNAEndogenousTLR10UnknownUnknown		Double-stranded RNA	Viruses						
Lipopolysaccharide (LPS)BacteriaGlycoinositol- phospholipidsProtozoaEnvelope glycoproteinsVirusesHost-derived HMGB1 and HSPsEndogenousTLR5FlagellinBacteriaTLR7Single-stranded RNAVirusesTLR8Single-stranded RNAVirusesTLR9Unmethylated CpG DNAProtozoaMitochondrial DNAEndogenousTLR10UnknownUnknown	TLR3	Poly (I:C)	Synthetic analog of double- stranded RNA						
Glycoinositol- phospholipidsProtozoaEnvelope glycoproteinsVirusesHost-derived HMGB1 and HSPsEndogenousTLR5FlagellinBacteriaTLR7Single-stranded RNAVirusesTLR8Single-stranded RNAVirusesTLR9Unmethylated CpG DNAProtozoaMitochondrial DNAEndogenousTLR10UnknownUnknown		Lipopolysaccharide (LPS)	Bacteria						
Envelope glycoproteinsVirusesHost-derived HMGB1 and HSPsEndogenousTLR5FlagellinBacteriaTLR7Single-stranded RNAVirusesTLR8Single-stranded RNAVirusesTLR9Unmethylated CpG DNAProtozoaVirusesNirusesMitochondrial DNAEndogenousTLR10UnknownUnknown	TLR4	Glycoinositol- phospholipids	Protozoa						
Host-derived HMGB1 and HSPsEndogenousTLR5FlagellinBacteriaTLR7Single-stranded RNAVirusesTLR8Single-stranded RNAVirusesTLR9Unmethylated CpG DNAProtozoaVirusesMitochondrial DNAEndogenousTLR10UnknownUnknown		Envelope glycoproteins	Viruses						
TLR5 Flagellin Bacteria TLR7 Single-stranded RNA Viruses TLR8 Single-stranded RNA Viruses Minochondrial DNA DNA Bacteria Mitochondrial DNA Endogenous TLR10 Unknown Unknown		Host-derived HMGB1 and HSPs	Endogenous						
TLR7 Single-stranded RNA Viruses TLR8 Single-stranded RNA Viruses TLR9 Unmethylated CpG DNA Bacteria Protozoa Viruses Mitochondrial DNA Endogenous TLR10 Unknown Unknown	TLR5	Flagellin	Bacteria						
TLR8 Single-stranded RNA Viruses TLR9 Unmethylated CpG Bacteria Protozoa Protozoa Viruses Viruses Mitochondrial DNA Endogenous TLR10 Unknown Unknown	TLR7	Single-stranded RNA	Viruses						
Bacteria Bacteria DNA Protozoa Viruses Viruses Mitochondrial DNA Endogenous TLR10 Unknown Unknown	TLR8	Single-stranded RNA	Viruses						
Unmethylated CpG Protozoa DNA Viruses Mitochondrial DNA Endogenous TLR10 Unknown Unknown			Bacteria						
Mitochondrial DNA Endogenous TLR10 Unknown Unknown		Unmethylated CpG DNA	Protozoa						
Mitochondrial DNA Endogenous TLR10 Unknown Unknown	ILKS		Viruses						
TLR10 Unknown Unknown		Mitochondrial DNA	Endogenous						
	TLR10	Unknown	Unknown						

Recombinant TLR Proteins from R&D Systems

R&D Systems offers the widest selection of recombinant human and mouse TLR proteins for investigating the ligand-binding and biochemical properties of different toll-like receptors. Stringent production and purification standards ensure that R&D Systems* proteins will provide researchers with industry-leading bioactivity and lot-to-lot consistency.



Recombinant Human TLR4/MD-2 Complex Blocks LPS-Induced TNF-α Secretion by PMA-Differentiated U937 Cells. The U937 human histiocytic lymphoma cell line was differentiated with PMA and treated with 10 ng/mL lipopolysaccharide (LPS) that had been preincubated for 1 hour with the indicated concentrations of Recombinant Human TLR4/MD-2 Complex (R&D Systems, Catalog # 3146-TM). Following 20 hours of incubation with the LPS-TLR4/MD-2 complex, TNF- α secretion was measured using the Human TNF-a Quantikine® ELISA Kit (R&D Systems, Catalog # DTAOOC).



Recombinant Human TLR3 Inhibits Poly (I:C)-Induced IL-8 Secretion by TLR3-transfected HEK293 Cells. The HEK293 human embryonic kidney cell line was transfected with TLR3 and treated with increasing concentrations of Poly (I:C) (Tocris, Catalog # 4287). CXCL8/IL-8 secretion was measured using the Human CXCL8/IL-8 Quantikine* ELISA Kit (R&D Systems, Catalog # D8000C; orange line). The stimulatory effect induced by 10 µg/mL of poly (I:C) was inhibited by treating the cells with increasing concentrations of Recombinant Human TLR3 (R&D Systems, Catalog # 1487-TR; blue line). The ED_{so} for this effect is typically 5-10 µg/mL.





Recombinant TLR Proteins								
Molecule	Species	Source	Catalog #					
TLD1	Human	NSO	1484-TR					
	Mouse	NSO	1476-TR					
TI P2	Human	NSO	2616-TR					
	Mouse	Sf21 (baculovirus)	1530-TR					
TLR3	Human	NSO	1487-TR					
	Mouse	NSO	3005-TR					
TLR4	Human	NSO	1478-TR					
TLR4/MD-2	Human	NSO	3146-TM					
TLR5	Mouse	СНО	7915-TR					
TIRE	Human	СНО	7755-TR					
ILKO	Mouse	Sf21 (stably transfected)	1533-TR					
TLR9	Mouse	СНО	7960-TR					
TLR10	TLR10 Human CHO		6619-TR					
TLR11	Mouse	СНО	7640-TR					
TLR12	Mouse	СНО	8086-TR					

Tocris[®] Small Molecules for TLR Research

Tocris is the leading supplier of novel and exclusive tools for life science research. Their portfolio includes a collection of TLR agonists and inhibitors that can be used to characterize the signaling pathways downstream of specific toll-like receptors and determine the effects of TLR signaling on the immune response.

Tocris Small Molecules for TLR Research

Small Molecule	Description	Catalog #
CU-T12-9	Potent TLR1/2 agonist	5414
Pam ₃ CSK ₄	TLR1/2 agonist	4633
Pam ₃ CSK ₄ Biotin	Biotinylated Pam ₃ CSK ₄	4636
CU CPT 22	Selective TLR1/2 inhibitor	4884
Pam ₂ CSK ₄	TLR2/6 agonist	4637
Pam ₂ CSK ₄ Biotin	Biotinylated Pam ₂ CSK ₄	4638
Poly(I:C)	TLR3 agonist	4287
CU CPT 4a	Selective TLR3 inhibitor	4883
C34	TLR4 inhibitor	5373
DSR 6434	Potent TLR7 agonist	4809
Imiquimod	TLR7 agonist	3700
Resiquimod	TLR7 agonist	4536
RWJ 21757	TLR7 agonist	2719
Hydroxychloroquine sulfate	TLR9 inhibitor	5648

For more information, please visit | tocris.com

Immunology Literature from Tocris:

Immunology Product Listing

A collection of over 190 products for immunology research, including research tools for studying chemokine and cytokine signaling, chemotaxis, the complement system, immune cell signaling and inflammation.



Multiple Sclerosis Poster

 $\label{eq:main} \begin{array}{l} \mbox{Multiple sclerosis} \ (\mbox{MS}) \ \mbox{is an autoimmune disease characterized} \\ \mbox{by focal demyelination and axon degeneration}. \ \mbox{Created by Alastair} \end{array}$

Wilkins and Richard Ibitoye of University of Bristol, this poster summarizes the neurobiology and current therapies of MS.



To download or request a copy please visit | tocris.com/requestliterature

Unconjugated & Fluorochrome-conjugated TLR Antibodies

R&D Systems and Novus Biologicals together offer the most comprehensive selection of unconjugated and fluorochrome-conjugated antibodies for detecting toll-like receptors and related molecules using a variety of different applications. Our catalogs include antibodies that are validated for flow cytometry, immunocytochemistry/immunofluorescence, immunohistochemistry, Western blot, and blocking/ neutralization.

Blocking/Neutralization Antibodies for TLRs



Ligand-Induced IL-8 TLR2 Secretion and Neutralization using an Anti-Human TLR2 Antibody. The HEK293 human embryonic kidney cell line transfected with human TLR2 was treated with increasing concentrations of the synthetic tripalmitoylated lipopeptide Pam_CSK, (Tocris, Catalog # 4633). IL-8 secretion was measured using the Human CXCL8/IL-8 Quantikine® ELISA Kit (R&D Systems, Catalog # D8000C; orange line). The stimulatory effect induced by 0.5 µg/mL Pam₂CSK₄ was neutralized by treating the cells with increasing concentrations of a Mouse Anti-Human TLR2 Monoclonal Antibody (R&D Systems, Catalog # MAB2616; blue line). The ND₅₀ is typically 0.03-0.15 µg/mL.



TLR4/MD2 Ligand-Induced IL-8 Secretion and Neutralization using an Anti-Human TLR4 Antibody. The HEK293 human embryonic kidney cell line cotransfected with human TLR4 and MD-2 was treated with increasing concentrations of lipopolysaccharide (LPS). IL-8 secretion was measured using the Human CXCL8/IL-8 Quantikine® ELISA Kit (R&D Systems, Catalog # D8000C; orange line). The stimulatory effect induced by 75 ng/mL LPS was neutralized by treating the cells with increasing concentrations of a Goat Anti-Human TLR4 Antigen Affinity-purified Polyclonal Antibody (R&D Systems, Catalog # AF1478; blue line). The ND_{50} is typically 1.5–7.5 µg/mL.

For additional information on TLRs and other pattern recognition receptor families, explore R&D Systems pattern recognition receptorrelated signaling pathways at rndsystems.com/Pathways/Immunology

- Nod-like Receptor Signaling
 Pathways
- Toll-like Receptor Signaling
 Pathways
- Inflammasome Activation Pathways
- Pathogen or Damage-activated C-Type Lectin Receptor Signaling Pathways
- RIG-I-like Receptor Signaling
 Pathways

Printed copies of R&D Systems[®] pathways or our pattern recognition receptor-related miniposters listed below can be requested at rndsystems.com

- Pattern Recognition Receptors & the Innate Immune Response
- Inflammasomes: Intracellular Regulators of Pathogen Recognition, Host Defense, and Inflammation





TLR Antibodies for Flow Cytometry, Immunocytochemistry, Immunohistochemistry, or Western Blot

Detection of Plasma Membrane TLRs



Detection of TLR1 on Human Monocytes by Flow Cytometry. Human peripheral blood monocytes were stained with an APC-conjugated Goat Anti-Human TLR1 Antigen Affinity-purified Polyclonal Antibody (R&D Systems, Catalog # FAB1484A; filled histogram) or an APC-conjugated Isotype Control (R&D Systems, Catalog # IC108A; open histogram).



Detection of TLR2 in Human Monocytes by Flow Cytometry. Human peripheral blood monocytes were stained with a PE-conjugated Mouse Anti-Human TLR2 Monoclonal Antibody (R&D Systems, Catalog # FAB2616P; filled histogram) or a PEconjugated Isotype Control (R&D Systems, Catalog # ICO041P; open histogram).



Detection of TLR4 in RAW264.7 Cells by Immunocytochemistry. TLR4 was detected in the immersion-fixed RAW264.7 mouse monocyte/ macrophage cell line using a Rabbit Anti-Mouse TLR4 Monoclonal Antibody (R&D Systems, Catalog # MAB27591) at 1 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Rabbit IgG Secondary Antibody (R&D Systems, Catalog # NLO04; red) and counterstained with DAPI (blue). Specific staining was localized to cell surfaces and cytoplasm.



Detection of TLR5 on Human Monocytes by Flow Cytometry. Human peripheral blood monocytes were stained with an Alexa Fluor[®] 488-conjugated Mouse Anti-Human TLR5 Monoclonal Antibody (R&D Systems, Catalog # FAB6704G; filled histogram) or an Alexa Fluor 488-conjugated Mouse IgG₁ Isotype Control (R&D Systems, Catalog # ICO02G; open histogram).



Detection of TLR5 in Human Intestine by Immunohistochemistry. TLR5 was detected in immersion-fixed, paraffin-embedded sections of human intestine using a Mouse Anti-Human TLR5 Monoclonal Antibody (R&D Systems, Catalog # MAB6704) at 15 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Mouse HRP-DAB Cell & Tissue Staining Kit (R&D Systems, Catalog # CTS002; brown) and counterstained with hematoxylin (blue). Lower panels show a lack of labeling when primary antibodies are omitted and the tissue is stained with only secondary antibody followed by incubation with detection reagents.

Detection of Endosomal TLRs



Detection of TLR3 in A549 Cells by Flow Cytometry. The A549 human lung carcinoma cell line was stained with an APC-conjugated Goat Anti-Human TLR3 Antigen Affinity-purified Polyclonal Antibody (R&D Systems, Catalog # IC1487A; filled histogram) or an APC-conjugated lsotype Control (R&D Systems, Catalog # IC108A; open histogram). To facilitate intracellular staining, cells were fixed with Flow Cytometry Fixation Buffer (R&D Systems, Catalog # FC004) and permeabilized with Flow Cytometry Permeabilization/Wash Buffer I (R&D Systems, Catalog # FC005).



Detection of TLR7 in Ramos Human Burkitt's Lymphoma Cells by Flow Cytometry. The Ramos human Burkitt's lymphoma cell line was stained with a PE-conjugated Mouse Anti-Human TLR7 Monoclonal Antibody (R&D Systems, Catalog # IC5875P; filled histogram) or a PE-conjugated Mouse IgG₂₄ Isotype Control (R&D Systems, Catalog # ICO03P; open histogram). To facilitate intracellular staining, cells were fixed with Flow Cytometry Fixation Buffer (R&D Systems, Catalog # FC004) permeabilized with Flow Cytometry and Permeabilization/Wash Buffer I (R&D Systems, Catalog # FC005).



Detection of Mouse and Rat TLR7 by Western Blot. Lysates from mouse thymus tissue or the NR8383 rat alveolar macrophage cell line were separated by SDS-PAGE and immunoblotted using 1 µg/mL of the Rat Anti-Mouse/Rat TLR7 Monoclonal Antibody (R&D Systems, Catalog # MAB7156) followed by an HRP-conjugated Anti-Rat IgG Secondary Antibody. A specific band for TLR7 was detected at approximately 130 to 140 kDa under reducing conditions (as indicated).



Detection of TLR9 in Mouse Splenocytes by Flow Cytometry. Mouse splenocytes were stained with an APC-conjugated Rat Anti-Mouse B220/CD45R Monoclonal Antibody (R&D Systems, Catalog # FAB1217A) and either a (A) Rabbit Anti-Mouse TLR9 Monoclonal Antibody (R&D Systems, Catalog # MAB7960) or (B) Normal Rabbit IgG Control (R&D Systems, Catalog # AB-105-C), followed by a PE-conjugated Anti-Rabbit IgG Secondary Antibody (R&D Systems, Catalog # F0110).

Toll-like Receptor Antibodies from R&D Systems

R&D Systems antibodies undergo rigorous lot-specific quality control testing for all of the applications listed on our datasheets to ensure superior performance, reliability, and consistency. For more information, please visit rndsystems.com/Antibodies

		Unconjugated A	Antibodies	Fluorochrome-conjugated Antibodies for Flow Cytometry					
								Alex	a Fluor®
Molecule	Species	Clone	Catalog # (Applications)	APC	Fluorescein	PE	PerCP	488	700
	н	Polyclonal	AF1484 (FC, WB)	1		1			
TLR1	М	Polyclonal	AF1475 (FC, WB)			1			
	М	285923	MAB1475 (WB)						
	Н	383936	MAB2616 (B/N, FC)	1	1	1	1		1
	н	Polyclonal	AF2616 (E, FC, WB)						
ILR2	М	203325	MAB1530 (FC)	1	1				
	М	Polyclonal	AF1530 (WB)						
	н	512505	MAB1487 (WB)						
71 00	н	Polyclonal	AF1487 (WB)	1					
M	М	313129	MAB3005 (WB)	1		1			
	М	Polyclonal	AF3005 (WB)						
	Н	285227	MAB1478 (WB)						
TI R4	Н	610029	MAB14782 (WB)						
	н	610017	MAB14783 (IHC)						
	н	610015	MAB6248 (FC)	1	1	1	1		1
	н	Polyclonal	AF1478 (B/N, FC, ICC/IF, IHC, WB)						
	М	267518	MAB2759 (FC, ICC/IF)	1		1			
	М	1203B	MAB27591 (FC, ICC/IF)						
TLR5	н	624915	MAB6704 (FC, IHC)					1	
TIDO	М	418601	MAB1533 (FC)	1		1			
ILRO	М	Polyclonal	AF1533 (WB)						
71.07	н	533707	MAB5875 (FC)			1	1	1	
ILR/	M/R	726606	MAB7156 (WB)						
TLR8	н	935166	MAB8999 (FC)						
	н	229106	MAB3658 (FC)						
TLR9	н	Polyclonal	AF3658 (FC, IHC)					1	
	М	1138D	MAB7960 (FC, ICC/IF)						
TLR10	Н	670719	MAB6619 (WB)						
TLR11	М	786404	MAB7640 (FC)	1		1			
TLR12	М	1229C	MAB8086 (FC, WB)			1			
Application Key:	B/N Block/Neutra	lize EELISA FC	Flow Cytometry ICC/IF Immu	unocytochemistr	y/Immunofluores	scence IHC Im	munohistochem	nistry WB Wes	tern Blot

Additional Select Toll-like Receptor Antibodies from Novus Biologicals

Novus Biologicals offers a wide selection of TLR antibodies including some of the most highly referenced clones on the market. Most of these antibodies are conjugated to multiple different fluorochromes including a series of Alexa Fluor[®] and DyLight[®] dyes to provide a full range of options for multicolor experiments. For more information, please visit novusbio.com

	Unconjugated Antibodies Fluorochrome-conjugated Antibodies for Flow Cytometry											
									Alexa Fluor®			
Molecule	Species	Clone	Catalog # (Applications)	APC	Fluorescein	PE	PerCP	405	488	647	700	
TLR1	H/M/R	Polyclonal	NB100-56563 (FC, IHC, WB)									
	н	TL2.1	NB100-56722 (B/N, FA, FC, ICC/IF, IHC, IP)		1	1	1	1	1	~	1	
TLR2	H/M	T2.5	NBP1-42362 (FC, IHC, IP)		1							
	М	11G5	NBP2-27165 (FC, WB)*	1		1	1	1	1	1	1	
TLR3	Н/М	40C1285.6	NBP2-24875 (FC, ICC/IF, IHC, IP, WB)*	1	1	1	1	1	1	1	1	
	H/M	TLR3.7			1	1						
	н	HTA125	NB100-56723 (B/N, FA, FC, ICC/IF, IP, IV)*	1	1	1	1	1	1	~	1	
TLR4	Н/М	76B357.1	NB100-56566 (FC, ICC/IF, IHC, ChIP, WB)*	1	1	1	1			1		
	М	MTS510	NB100-56560 (FC, IP)*	1	1	1	1	1	1	1	1	
	H/M	85B152.5	NBP1-97728 (FC, WB)*	1	1	1	1	1	1	1	1	
TLR5 H/M	Н/М	19D759.2	NBP2-24787 (FC, IHC, WB)*	1	1	1	1	1	1	1	1	
	н	86B1153.2	NB100-56536 (FC, IHC)*	1	1	1	1	1	1	1	1	
TLR6	н	hPer6	NBP1-43142 (FC, WB)*	1		1	1	1	1	1	1	
	н	TLR6.127	NBP1-51493 (FC, ICC/IF, IHC, IP)									
	Н/М	4G6	NBP2-27332 (FC, ICC/ IF, WB)*	1		1	1	1	1	1	1	
ILR7	Н/М	Polyclonal	NBP2-24906 (FC, ICC/IF, IHC, WB)									
71.00	Н/М	44C143	NBP2-24917 (FC, IHC, SW, WB)*	1	1	1	1	1	1	1	1	
ILR8	Н/М	Polyclonal	NBP1-77203 (E, ICC/ IF, WB)									
	н	eB72-1665	NBP1-43140 (FC, IHC, IP, WB)*	1		1	1	1	1	1	1	
TLR9	TLR9 H/M/R	26C593.2	NBP2-24729 (E, FA, FC, ICC/IF, IHC, IP, IV, SW, WB)*	1	5	✓	1	1	1	1	1	
	М	M9.D6	NBP1-43141 (FC, WB)*	1	1	1	1	1	1	1	1	
TLR10	Н	3C10C5	NBP1-70343 (FC)*	\checkmark		1	1	1	1	1	1	
TLR13	M/R	Polyclonal	NBP2-24539 (FC, WB)									
Application	n Key: B/N	Block/Neutra	lize ChIP Chromatin Immuno	precipitation	E ELISA FA F	unctional Ass	ay FC Flow Cy	tometry	. –			

ICC/IF Immunocytochemistry/Immunofluorescence IHC Immunohistochemistry IP Immunoprecipitation IV In vitro WB Western Blot

* In addition to the fluorochrome-conjugated forms listed, these antibodies are also available in several DyLight®-conjugated forms. DyLight conjugates include DyLight 350, 405, 405LS, 488, 550, 650, 680, and 755. Please visit novusbio.com for more information.

Proteins & Antibodies for TLR Co-Receptors, Signaling Regulators, and Adaptor Proteins

TLR co-receptors such as CD14, CD36, and MD-2 have essential roles in ligand recognition, while intracellular, TIR domain-containing signaling adaptors such as MyD88, TRIF, TRAM, and TIRAP are required for activating signaling pathways downstream of TLRs. Additionally, molecules such as RP105/CD180, SARM1, PRAT4A, and PRAT4B are involved in regulating TLR signaling or TLR localization. R&D Systems and Novus Biologicals provide high quality antibodies to detect these proteins by multiple different methods.



Recombinant Human CD14 Enhances LPS-Stimulated IL-8 Secretion by THP-1 Cells. The THP-1 human acute monocytic leukemia cell line was treated with 15 ng/mL lipopolysaccharide (LPS) and the indicated concentrations of Recombinant Human CD14 (R&D Systems, Catalog # 383-CD). CXCL8/IL-8 secretion was measured using the Human CXCL8/IL-8 Quantikine* ELISA Kit (R&D Systems, Catalog # D8000C). The ED₅₀ for this effect is typically 0.75-4.5 ng/mL.



Detection of CD14 in Human Peripheral Blood Mononuclear Cells by Flow Cytometry. Human peripheral blood mononuclear cells were stained with an APC-conjugated Mouse Anti-Human CD14 Monoclonal Antibody (R&D Systems, Catalog # FAB3832A).



Recombinant Mouse LBP-Induced IL-8 Secretion and Neutralization using an Anti-Mouse LBP Antibody. The THP-1 human acute monocytic leukemia cell line was treated with 5 ng/mL lipopolysaccharide and the indicated concentrations of Recombinant Mouse Lipopolysaccharide-Binding Protein (LBP; R&D Systems, Catalog # 6635-LP). CXCL8/IL-8 secretion was measured using the Human CXCL8/IL-8 Quantikine® ELISA Kit (R&D Systems, Catalog # D8000C; orange line). The stimulatory effect induced by 50 ng/mL Recombinant Mouse LBP was neutralized by treating the cells with increasing concentrations of a Rat Anti-Mouse LBP Monoclonal Antibody (R&D Systems, Catalog # MAB6635; blue line). The ND₅₀ is typically $1-5 \mu g/mL$.





Detection of MyD88 by Immunocytochemistry and Flow Cytometry. (A) MyD88 was detected in immersionfixed RAW264.7 mouse monocyte/macrophage cell line using a Goat Anti-Mouse/Rat MyD88 Antigen Affinity-purified Polyclonal Antibody (R&D Systems, Catalog # AF3109) at 1.7 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights[™] 557-conjugated Anti-Goat IgG Secondary Antibody (R&D Systems, Catalog # NLOO1; red) and counterstained with DAPI (blue). Specific staining was localized to the cytoplasm. (B) MyD88 was detected in mouse splenocytes by staining with the Goat Anti-Mouse/Rat MyD88 Antigen Affinity-purified Polyclonal Antibody (R&D Systems, Catalog # AF3109; filled histogram) followed by staining with an APC-conjugated Anti-Goat IgG Secondary Antibody (R&D Systems, Catalog # F0108). To facilitate intracellular staining, cells were fixed with paraformaldehyde and permeabilized with saponin.

Recombinant Proteins & Antibodies for TLR Co-Receptors, Signaling Regulators, & Adaptor Proteins from R&D Systems

	Pro	oteins		Unconjugated A	ntibodies		Fluorochror	ne-conjugated A	Antibodies for F	low Cytometry
Molecule	Species	Catalog #	Species	Clone	Catalog # (Applications)	APC	Fluorescein	PE	PerCP	*Alexa Fluor® 405/594/647/700/750
CD14	Human	383-CD	Human	134620	MAB3832 (B/N, FC, WB)	1	1	J	5	1
	Mouse	982-CD	Mouse	159010	MAB982 (FC, WB)	1			1	✓
	Human	1055 CD	Human	255606	MAB19551 (FC)	1	1	1		
	Human	1900-00	Human	255619	MAB1955 (WB)					
CD36	Mouse	2519-CD	Mouse	324205	MAB25191 (FC, IHC)	1				
			Mouse	324216	MAB2519 (WB)					
	Human	870-LP	Human	Polyclonal	AF870 (WB)					
LBP	Human	6445-LP	Mouse	749405	MAB6635 (B/N)					
	Mouse	6635-LP	Mouse	Polyclonal	AF6635 (WB)		1.15 1.0			27 / COL
	Human	925-MD	Human	Polyclonal	AF925 (FC, IHC, WB)		San Sal		X	
MD-1	MD-1		Human	153014	MAB925 (WB)			1 6 B 🖊	- All	
	Maura	120 MD	Mouse	142004	MAB130 (WB)		C.A.			
	wouse	130-1010	Mouse	Polyclonal	AF130 (WB)		1 8 30		Contra a	
MD 2	Humon	1797 MD	Human	288307	MAB1787 (WB)		123			No. Contraction
WID-2	numan	1/0/-IVID	Human	Polyclonal	AF1787 (WB)					· ·
			Human	316628	MAB29281 (FC)			CERTAIN OF		
			Human	316603	MAB2928 (ICC/IF)					
MyD88			Human	Polyclonal	AF2928 (FC, ICC/ IF, SW, WB)		a faith		Acc	
			Mouse	316902	MAB3109 (ICC/IF)		1 4 14	18 7 B.	- date	
			Mouse	Polyclonal	AF3109 (FC, ICC/ IF, SW, WB)		Detection Immunoh	of TRAM/TIO	CAM2 in C2 TRAM/TI	CAM2 was
	Human	7484-PR					detected	in the imme cell line using	rsion-fixed C	Vouse TRAM/
PRAI4A	Mouse	4429-PR	Mouse	Polyclonal	AF4429 (WB)		TICAM2	Monoclonal	Antibody (R	&D Systems,
PRAT4B			Mouse	Polyclonal	AF5015 (FC, WB)		Catalog #	MAB43481) a perature Cell	at 10 ug/mL s were stair	for 3 hours at
RP105/ CD180	Mouse	1378-RP					NorthernL Secondary	ights™ 557- ⁄ Antibody (F	conjugated R&D System	Anti-Rat IgG s, Catalog #
			Human	754711	MAB7037 (ICC/IF)		NL013; re	d) and counter	erstained wit	h DAPI (blue).
SARM1			Human	Polyclonal	AF7037 (ICC/IF, IP, WB)		Specific st	aming was loo	alized to the	cytopiasm.
TRAM/TI-			Human/ Mouse/ Rat	Polyclonal	AF4348 (FC, ICC/ IF, WB)					
CAM2			Mouse	757706	MAB4348 (WB)					
			Mouse	757712	MAB43481 (ICC/ IF)					
			Human	567212	MAB6216 (WB)					
TRIF			Human	Polyclonal	AF6216 (ICC/ IF, WB)					
Application Ke	v: B/N Block	Neutralize F	C Flow Cytom	etry ICC/IF Imm	nunocvtochemistrv/					

Immunofluorescence IHC Immunohistochemistry IP Immunoprecipitation SW Simple Western WB Western Blot

*Indicates one or more of the following conjugates is available.

Additional Select Antibodies for TLR Co-Receptors, Signaling Regulators, & Adaptor Proteins from Novus Biologicals

	Unc	onjugated Anti	bodies	Fluorochrome-conjugated Antibodies					ies for Flow Cytometry			
									Alexa	a Fluor®		
Molecule	Species	Clone	Catalog # (Applications)	APC	Fluorescein	PE	PerCP	405	488	647	700	
0014	Human	HCD14	NB100-78083 (FC)*	1	1	1	1	1	1	1	1	
0014	Human	M5E2	NB100-77758 (FC, ICC/IF, IHC)*	1	1	1	1	1	1	1	1	
0026	Human	SM0	NB100-65522 (FC, IHC)*	1		1	1	1	1	1	1	
0030	Mouse/Rat	D-2712	NB110-59724 (IHC, IP, WB)*		1			1	1	1	1	
LBP	Human	Polyclonal	NBP1-88371 (IHC)									
MD-1	Human/ Mouse	Polyclonal	NB100-56700 (IHC, WB)									
	Mouse	MD-14	NB100-77635 (FC)									
	Human/ Mouse/Rat	Polyclonal	NB100-56655 (IHC, WB)									
MD-2	Human/ Mouse/Rat	Polyclonal	NBP1-77201 (E, ICC/IF, IHC, WB)									
	Human/ Mouse	9F1B1	NBP1-75512 (E, ICC/IF, IHC, WB)									
MyD88	Human/ Mouse	4D6	NBP2-27369 (FC, ICC/IF, WB)*	1		1	1	1	1	1	1	
	Human/ Mouse/Rat	Polyclonal	NBP1-91153 (ICC/IF, IHC, WB)									
PRAI4A	Human/ Mouse/Rat	Polyclonal	NBP2-23847 (IHC)									
PRAT4B	Human	Polyclonal	NBP1-81085 (ICC/IF, IHC, WB)									
RP105/CD180	Human	MHR73-11	NBP1-49025 (FC, IHC, IP)*	1		1	1	1	1	~	1	
SARM1	Human/ Mouse	Polyclonal	NBP1-77200 (E, ICC/IF, IHC, WB)									
	Human	20D1055.1	NB100-56730 (WB)									
TIRAP/MAL	Human/ Mouse	Polyclonal	NBP2-47605 (ICC/IF, IHC)									
	Human	Polyclonal	NBP1-89566 (IHC)									
TRIF	Human/ Mouse/Rat	Polyclonal	NB120-13810 (ICC/IF, SW, WB)									
UNC93B	Human/ Mouse/Rat	Polyclonal	NBP2-24743 (WB)									
Application Key: WB Western Blot	E ELISA FC FIG	ow Cytometry	ICC/IF Immunocytochemistry/Immun	ofluorescen	ce IHC Immun	ohistochen	nistry IP Imr	nunoprecij	oitation S	W Simple	Western	

*In addition to the fluorochrome-conjugated forms listed, these antibodies are also available in several DyLight®-conjugated forms. DyLight conjugates include DyLight 350, 405, 405LS, 488, 550, 650, 680, and 755. Please visit novusbio.com for more information.

Antibodies for Detecting TLR Signaling Molecules

A

Following ligand recognition by TLRs, downstream signal transduction cascades result in the activation of several kinases including IKK, MKK, p38 MAPK, JNK, and TBK1. These kinases activate transcription factors such as AP-1, NF_KB, IRF3, and IRF7, which promote the expression of pro-inflammatory cytokines, chemokines, and type I and type III interferons. R&D Systems and Novus Biologicals offer antibodies to detect the kinases and transcription factors involved in TLR signaling, while Tocris offers inhibitors and activators of these molecules. Together, these reagents can be used to fully characterize TLR signaling pathways.



Intracellular Staining of ReIA/NF_KB p65 in HeLa Cells by Flow Cytometry. HeLa human cervical epithelial carcinoma cells were stained with an Alexa Fluor[®] 488-conjugated Mouse Anti-Human/ Mouse ReIA/NF_KB p65 Monoclonal Antibody (R&D Systems, Catalog # IC5078G; filled histogram) or an Alexa Fluor 488-conjugated Mouse IgG_{2B} Isotype Control (R&D Systems, Catalog # IC0041G; open histogram).



Detection of IRAK4 by Western Blot and Immunocytochemistry. (A) Recombinant Human IRAK1, IRAK2, IRAK4 (2 ng/lane), and Iysates from the A431 human epithelial carcinoma cell line or the K562 human chronic myelogenous leukemia cell line were separated by SDS-PAGE and immunoblotted using 1 µg/mL of the Goat Anti-Human IRAK4 Antigen Affinity-purified Polyclonal Antibody (R&D Systems, Catalog # AF3919) followed by an HRP-conjugated Anti-Goat IgG Secondary Antibody (R&D Systems, Catalog # HAF109). A specific band for IRAK4 was detected at approximately 55 kDa under reducing conditions (as indicated). (B) IRAK4 was detected in the immersion-fixed THP-1 human acute monocytic leukemia cell line using the Goat 15 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (R&D Systems, Catalog # AF3919) at 15 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (R&D Systems, Catalog # AF3919) at 0.5 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (R&D Systems, Catalog # NL001; red) and counterstained with DAPI (blue). Specific staining was localized to the cytoplasm.

Sample-Size Antibodies Now Available Choose From Over 10,000 Antibodies

R&D Systems® Antibodies for TLR Signaling Molecules

	Unconjuga	ted Antibodies	Tocris Small Molecule Activators/Inhibitors Available
Molecule	Species	Catalog # (Applications)	
	Human	MAB4299 (SW, WB)	X
ΙκΒ-α	Human/Mouse	AF4299 (WB)	
Phospho-IκB-α (S32/S36)	Human	AF4809 (WB)	
	Human/Rat	MAB3425 (WB)	
ТКВ-р	Human/Mouse	AF5225 (WB)	
	Human	MAB4300 (WB)	
ΙκΒ-ε	Human	AF4300 (IHC)	
	Mouse	AF4637 (WB)	
ΙΚΚ-α	Human/Mouse/Rat	AF3768 (ICC/IF, WB)	X
Phospho-IKK-a (S176/S180)	Human	MAB3768 (WB)	
IKK-B	Human	AF4535 (WB)	X
	Mouse	MAB7155 (WB)	
IK K-w	Human/Mouse/Rat	AF2684 (ICC/IF, SW, WB)	X
	Human/Mouse/Rat	AF4365 (WB)	
	Human	AF3199 (ICC/IF, WB)	
1717-2	Human/Mouse/Rat	MAB3199 (ICC/IF, WB)	
IRAK1	Human	AF4048 (WB)	Х
IRAK2	Human	MAB6690 (WB)	
IRAK3	Human	AF6264 (WB)	
IRAK4	Human	AF3919 (ICC/IF, WB)	X
INI/ Don Crossifie	Human/Mouse/Rat	AF1387 (IHC, WB)	X
JNK Pari Specific	Human/Mouse/Rat	MAB1387 (WB)	
Phaanha INII (T492 ()(495)	Human/Mouse/Rat	AF1205 (IHC, SW, WB)	
Phospho-JNK (1183/1185)	Human/Mouse/Rat	MAB1205 (ICC/IF, SW, WB)	
15112.4	Human/Mouse/Rat	MAB17761 (ICC/IF, WB)	X
	Human/Mouse/Rat	MAB1776 (WB)	
JNK1/2	Human/Mouse/Rat	MAB2076 (ICC/IF, WB)	
INIZO .	Human/Mouse/Rat	MAB1846 (ICC/IF, WB)	Х
	Human/Mouse/Rat	AF1846 (WB)	
МККЗ	Human/Mouse/Rat	MAB2515 (ICC/IF, WB)	
МККЗ/МКК6	Human/Mouse/Rat	MAB2514 (WB)	
MKK4	Human	MAB3390 (ICC/IF)	
Phospho-MKK4 (S257/T261)	Human/Mouse/Rat	AF2990 (ICC/IF, WB)	
	Human/Mouse/Rat	MAB1604 (ICC/IF, WB)	
МКК6	Human/Mouse/Rat	AF16041 (WB)	
	Human/Mouse/Rat	AF1604 (WB)	
MKKZ	Human	AF3579 (IHC, WB)	
	Human	MAB3579 (IHC)	
n28~	Human/Mouse/Rat	AF8691 (IHC, SW, WB)	X
poor	Human/Mouse/Rat	MAB869 (WB)	
Pheenho n 28 m (T190 ()(192))	Human	MAB8691 (WB)	
Filospilo-p384 (1180/1182)	Human	MAB8692 (ICC/IF)	
	Human/Mouse/Rat	AF1347 (IHC, SW, WB)	Х
ρ38γ	Human/Mouse/Rat	MAB1347 (SW, WB)	
	Human/Mouse/Rat	AF1644 (WB)	
295	Human	AF1519 (IHC, WB)	Х
1200	Human	MAB1519 (WB)	
RIP1/RIPK1	Human/Mouse/Rat	MAB3585 (SW, WB)	X
TAB1	Human/Mouse	AF3578 (ICC/IF, WB)	
TAK1	Human	MAB5307 (WB)	
TANK	Human/Mouse	AF4755 (WB)	
TRAE 2 looform 2	Human	AF3278 (WB)	
IRAF-3 ISOTORM 2	Human/Mouse/Rat	MAB3278 (WB)	
TRAF-6	Human	AF3284 (WB)	
Application Key: ICC/IF Immunocytochemi	stry/Immunofluorescence IHC Immuno	histochemistry SW Simple Western WB	Western Blot

R&D Systems® Antibodies for TLR-related Transcription Factors

	Uncor	njugated Antibodies	Fluoro	ochrome-conjugated Antik	Tocris Small Molecule Activators/ Inhibitors Available	
Molecule	Species	Catalog # (Applications)	APC	Alexa Fluor [®] 488	PE	
c-Fos	Human	AF7254 (WB)				
FeeD (0000	Human	AF2214 (IHC, WB)				
FOSB/GUS3	Human/Mouse	MAB2214 (WB)				
EDA 1	Human	AF4935 (IHC, WB)				
TRA-1	Human	MAB4935 (WB)				
	Human	AF4019 (FC, ICC/IF, WB)				
IRF3	Human	MAB4019 (FC, WB)	1	1	1	
	Mouse	AF4454 (WB)				
a lun	Human	MAB2670 (ICC/IF, WB)				X
c-Jun	Human	AF2670 (WB)				
Phospho-c-Jun (S63)	Human	MAB8930 (ICC/IF, SW, WB)				
JunB	Human	MAB4456 (WB)				
	Human	AF4456 (WB)				
JunD	Human/Mouse	MAB5526 (WB)				
	Human/Mouse	AF5526 (WB)				
NE-P1	Human	MAB2697 (WB)				Х
INFKDI	Human/Mouse	AF2697 (ChIP, WB)				
ΝϜκΒ2	Human	MAB28881 (ChIP, ICC/ IF, WB)				х
	Human	MAB4606 (ICC/IF, WB)				
c-Rel	Human/Mouse	AF2699 (ChIP, ICC/IF, SW, WB)				
	Mouse	MAB2699 (WB)				
	Human	MAB50781 (FC)				
RelA/NF _K B p65	Human/Mouse	MAB5078 (FC, ICC/IF, WB)	1	1	1	
	Human/Mouse	AF5078 (ChIP, SW, WB)				
Phospho-ReIA/NFκB p65 (S529)	Human	MAB7624 (WB)				
Phospho-ReIA/NFκB p65	Human	MAB7226 (ICC/IF, WB)				
(\$536)	Human	MAB72261 (ICC/IF, SW, WB)				
RelB Human MAB2698 (ICC/IF, IHC, WB)						
Application Key: FC Flow Cyton SW Simple Western WB West	netry ICC/IF Imm ern Blot	nunocytochemistry/Immunofluor	escence IHC Imm	nunohistochemistry Chll	Chromatin Immu	noprecipitation

Additional Select Antibodies for TLR Signaling Molecules from Novus Biologicals

	Unconjuga	ated Antibodies			Fluorochr	ome-conjugated /	Antibodies		
Molecule	Species	Catalog # (Applications)	APC	PE	PerCP	Alexa Fluor®			
						405	488	647	700
ΙκΒ-α	Human/Mouse/Rat	NB100-56507 (FC, ICC/ IF, IHC, IP, SW, WB)*	1	1	1	1	1	1	1
ΙΚΚ-α	Human/Mouse	NB100-56704 (FC, ICC/ IF, IHC, IP, SW, WB)*	1	1	1	1	1	1	1
ικκ-β	Human/Mouse	NB100-56509 (FC, ICC/ IF, IHC, IP, SW, WB)*	1	1	1	1	1	1	1
IRAK4	Human/Mouse/Rat	NB500-597 (FC, IP, WB)							
RIP1	Human/Mouse/Rat	NBP1-77077 (E, ICC/IF, IHC, WB)							
TRAF-3	Human/Mouse/Rat	NB100-56176 (IHC, IP, WB)							
	Human/Mouse	NB100-56436 (FC, WB)							
TRAF-6	Human/Mouse/Rat	NB100-56179 (IHC, IP, WB)							
Application WB Wester	Key: E ELISA FC Flo n Blot	w Cytometry ICC/IF Immu	nocytochemistry/	Immunofluoresce	nce IHC Immun	ohistochemistry	IP Immunopreci	pitation SW Sim	ple Western

*In addition to the fluorochrome-conjugated forms listed, these antibodies are also available in several DyLight®-conjugated forms. DyLight conjugates include DyLight 350, 405, 405LS, 488, 550, 650, 680, and 755. Please visit novusbio.com for more information.

Additional Select Antibodies for TLR-related Transcription Factors from Novus Biologicals

	Une	conjugated Antibodies		Fluorochrome-con	jugated Antibodies			
Molecule	Species	Catalog # (Applications)	Alexa Fluor [®]					
			405	488	647	700		
c-Fos	Human/Mouse/Rat	NBP1-89065 (ICC/IF, IHC, WB)						
IRF7	Human/Mouse/Rat	NBP1-77263 (E, ICC/IF, IHC, WB)						
c-Jun	Human/Mouse/Rat	NB110-55569 (FC, ICC/IF, IHC, IP, WB)						
NF _K B1	Human/Mouse	NBP1-77395 (ICC/IF, IHC, SW, WB)	1	1	1	1		
ΝFκB2	Human/Mouse/Rat	NBP1-87760 (ICC/IF, IHC, SW, WB)						
Application Key: WB Western Blo	Application Key: E ELISA FC Flow Cytometry ICC/IF Immunocytochemistry/Immunofluorescence IHC Immunohistochemistry IP Immunoprecipitation SW Simple Western WB Western Blot							

ELISA Kits for Select Cytokines Induced by TLRs

For detecting cytokines and chemokines that are induced by TLR signaling, R&D Systems offers complete, ready-to-run Quantikine[®] ELISA Kits and the more flexible DuoSet[®] ELISA Development Systems.

· Detailed protocol booklets

Colorimetric detection

Quantikine® ELISA Kit Features

- · Complete, ready-to-use kits
- Exhaustively tested for superior quality and reproducibility

Assay Principle



A microplate pre-coated with capture antibody is provided. Samples or standards are added and any analyte present is bound by the immobilized antibody. Unbound materials are washed away (Step 1). A second HRP-labeled detection antibody is added and binds to the captured analyte. Unbound detection antibody is washed away (Step 2). Tetramethylbenzidine (TMB) substrate solution is added to the wells and a blue color develops in proportion to the amount of analyte present in the sample. Color development is stopped turning the color in the wells to yellow. The absorbance of the color at 450 nm is measured (Step 3).

DuoSet[®] ELISA Development System Features

When complete kits are not an option, DuoSet[®] ELISA Development Systems offer an economical alternative. DuoSet[®] Kits contain the essential components required to develop an immunoassay, but unlike Quantikine[®] ELISA Kits, they require the user to set up the assay by coating a microplate with the provided capture antibody. DuoSet[®] Kits also provide a biotinylated detection antibody and streptavidin-HRP, enabling chemiluminescent or colorimetric detection, a mass-calibrated standard, and detailed protocol.

- · Contains all of the essential components required to develop an immunoassay for a specific target
- · Contains carefully selected and validated antibodies, reducing development time
- · Provides sufficient reagents for five or fifteen 96-well plates
- · Includes mass-calibrated recombinant standard, reducing assay variability
- · Can be adapted for use across multiple platforms

Molecule	Species	Quantikine [®] ELISA Kits Catalog #	Quantikine [®] High Sensitivity ELISA Kits Catalog #	DuoSet [®] ELISA Development Systems Catalog #
	Human	DIF50		DY285
ΙΓΙΝ-γ	Mouse	MIF00		DY485
11 10/11 150	Human	DLB50	HSLB00C	DY201
IL-10/IL-1F2	Mouse	MLBOOC		DY401
	Human	D6050	HS600B	DY206
IL-0	Mouse	M6000B		DY406
IL-8/CXCL8	Human	D8000C	HS800	DY208
TNF-α	Human	DTAOOC	HSTAOOD	DY210
	Mouse	MTAOO		DY410

Multiplex Assays from R&D Systems

R&D Systems also offers Proteome Profiler[™] Antibody Arrays and Luminex[®] Bead-Based Assays for simultaneously profiling the levels of multiple analytes in a single sample. Multiplexing allows researchers to maximize data collection from small sample volumes, minimize experimental variability, and optimize productivity.

Proteome Profiler[™] Antibody Arrays

Proteome Profiler Antibody Arrays are available for determining the relative levels of phosphorylated MAPKs, NFκB-related analytes, chemokines, or cytokines in a single sample.



Simultaneous Detection of Multiple Analytes in IFN-γ-, LPS-treated THP-1 Cell Lysates using the Proteome **Profiler™** Human Cytokine Array. The THP-1 human acute monocytic leukemia cell line was untreated or treated with 1 µg/mL Recombinant Human IFN-γ (R&D Systems, Catalog # 285-IF) for 8 hours and 1 µg/mL lipopolysaccharide (LPS) for 16 hours. Cytokine expression in 200 µg of cell lysate was analyzed using the Proteome Profiler Human Cytokine Array (R&D Systems, Catalog # ARY005B).

Select Proteome Profiler[™] Antibody Arrays from R&D Systems

Kit	Catalog #
Proteome Profiler Human Phospho-MAPK Array Kit	ARY002B
Proteome Profiler Human NF κ B Pathway Array Kit	ARY029
Proteome Profiler Human Cytokine Array Kit	ARY005B
Proteome Profiler Human XL Cytokine Array Kit	ARY022
Proteome Profiler Human Chemokine Array Kit	ARY017
Proteome Profiler Mouse Cytokine Array Kit, Panel A	ARY006
Proteome Profiler Mouse XL Cytokine Array Kit	ARY028
Proteome Profiler Mouse Chemokine Array Kit	ARY020

Please visit rndsystems.com/ProteomeProfiler for a list of analytes detected by each array and the qualified sample types.

Luminex[®] Assays

R&D Systems offers two versions of our Luminex[®] bead-based assays. Our standard Luminex Assays offer the largest, customizable menu of analytes for bead-based multianalyte profiling using cell culture supernatnants, serum, or plasma samples. These assays allow up to 100 analytes to be simultaneously profiled using polystyrene microparticles or 50 analytes using magnetic particles. Please visit rndsystems.com/LuminexAssay to see our industry-leading selection of analyte combinations.

Luminex High Performance Assays offer defined analyte panels for bead-based multianalyte profiling. These assays rely on panel-optimized diluents that provide maximum performance for a smaller group of analytes than our standard Luminex Assays. Each assay is fully validated for all sample types indicated for a given panel. In-house testing demonstrates that analyte concentrations determined using our Luminex High Performance Assays correlate closely with those obtained using our single analyte Quantikine[®] ELISA Kits. Please visit rndsystems.com/LuminexPerformance to view a list of available kits.



Custom Services from Bio-techne

When your work demands unique reagents or scientific support, turn to the decades of product development legacy behind Bio-techne's trusted brands. Together with a dedicated project manager, our expert scientists, quality assurance team, and world-class technical support, we will deliver custom solutions tailored to bring you success faster and more economically.

Benefits of Custom Services from Bio-Techne

- Scientific expertise
- Consistency
- Supply
- Large-scale production
- Regulatory support
- Quality results
- Timeliness
- ISO-certified Quality Management System and FDA registered
- · Long-term cost savings
- Confidentiality
- Dedicated project managers

Custom Services

- Protein Services
- Assay Services
- Antibody Services
- Recombinant Antibody Conversion
- Sister Clone Availability
- Luminex[®] Custom Services
- Biomarker Testing Service
- Chemistry Services
- Custom Compound Libraries from Tocris
- Bioactivity Testing Services
- Ubiquitin/Proteasome Custom Services

What You Can Expect

- · Identify the need
- Consult with our experts
- Refine the project specifics, milestones, and deliverables
- Review a statement of work
- Receive regular project updates
- Accept delivery of custom product or service













Globalinfo@bio-techne.combio-techne.com/find-us/distributorsTEL +1 612 379 2956North AmericaTEL 800 343 7475Europe | Middle East | AfricaTEL +44 (0)1235 529449Chinainfo.cn@bio-techne.comTEL +86 (21) 52380373



For research use or manufacturing purposes only. Trademarks and registered trademarks are the property of their respective owners.

BR_TLR AAI2016_2139