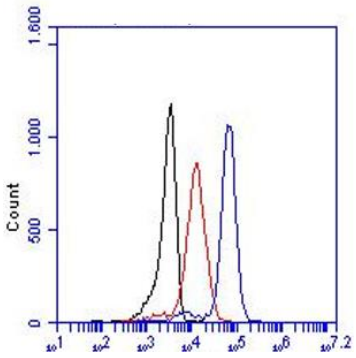
CERTIFICATE OF ANALYSIS – TECHNICAL DATA SHEET

Product name	TLR3, Human, clone TLR3.7		
Catalog number	HM2096-5MG		
Lot number	-	**Expiry date**	-
Volume	-	**Amount**	5 mg
Formulation	0.2 µm filtered in PBS	**Concentration**	>0.5 mg/ml
Host Species	Mouse IgG1	**Conjugate**	None
Endotoxin	<24 EU/mg	**Purification**	Protein G
Storage	4°C		

Application notes

	IHC-F	IHC-P	IF	FC	FS	IA	IP	W
Reference #	5,6	5	3	1,3,4,5	1,6		1,3	
Yes	●	●	●	●	●		●	
No								●
N.D.						●		

N.D.= Not Determined; IHC = Immuno histochemistry; F = Frozen sections; P = Paraffin sections; IF = Immuno Fluorescence; FC = Flow Cytometry; FS = Functional Studies; IA = Immuno Assays; IP = Immuno Precipitation; W = Western blot



FC: Flow cytometric detection of human TLR3 on THP-1 cells (mAb TLR3.7, Cat.# HM2096). Red line represents the isotype control and the black line represents HM2096 with a concentration of 10 µg/ml.

Dilutions to be used depend on detection system applied. It is recommended that users test the reagent and determine their own optimal dilutions. The typical starting working dilution is 1:50.

- F: dried sections, fixed with 4% paraformaldehyde and subsequently washed in PBS and MQ. Sections were quenched with 0.3%H2O2 in methanol and washed in PBS. Sections were permeabilized with 0.4% triton-X100 in PBS. Pretreated slides were blocked with 1% horse serum for 20' and incubated o/n with antibody (Ref.5).
- FC: cells were incubated with 1 µg antibody together with 10 µg human IgG for 30'at 4°C in PBS/0,5% BSA (Ref.3).
- FS: 7.5*104 MRC5 cells were pre-treated with 10-20µg/ml for 1-24h at 37°C. Monoclonal antibody TLR3.7 inhibits dsRNA-induces IFN-beta production (Ref.1).
- IF: Cytospins of monocyte-derived iDCs were fixed for 30' with 3% formaldehyde in PBS, permeabilized with PBS/1%BSA/0.5%saponin. After PBS wash slides were incubated for 1h at RT with 20ug/ml antibody in PBS/1%BSA (Ref.3).
- P: Formalin fixed, paraffin embedded sections were deparaffinized with xylene, followed by washes in 95% and 70% EtOH. Sections were washed with water and permeabilized with 0.4% triton-X100 in PBS. Pretreated slides were blocked with 1% horse serum for 20' and incubated o/n with antibody (Ref.5).
- W: Total cellular protein was loaded on 7.5% SDS-PAGE and blotted on PDVF. Blots were incubated with 2µg/ml antibody o/n at 4°C (Ref.6).
- Positive control: Monocytes, granulocytes, lymphocytes, human fibroblast, MRC-5 & FS-4 cells; Negative control: HEK293 cells.

General Information

Description

The monoclonal antibody TLR3.7 recognizes the 116 kDa human Toll-like receptor 3 (TLR3, CD283). Toll-like receptors (TLRs) are highly conserved from Drosophila to humans and share structural and functional similarities. TLRs constitute of a family of pattern recognition receptors (PRRs) that mediate cellular responses to a large variety of pathogens (viruses, bacteria, and parasites) by specific recognition of so-called 'pathogen-associated molecular patterns'. Activation of TLRs, a family of at least 11 different members that function either as homo- or heterodimers, leads to activation of NFκB-dependent and IFN-regulatory factor-dependent signaling pathways. TLRs have a central role in innate immunity and are also required for the development of an adaptive immune response. TLRs are expressed by various cells of the immune system, such as macrophages and dendritic cells. TLRs are class I receptors, with a single α-helix that spans the cell membrane. They recognize and respond to molecules derived from bacterial, viral and fungal pathogens, such as lipopolysaccharide (LPS) from the outer membrane of Gram negative bacteria, peptidoglycan fragments from bacterial cell walls and single-stranded and double-stranded RNA from viruses. Some forms of RNA and DNA from pathogens exhibit immutable features that distinguish them from nucleic acids of higher organisms. For example, dsRNA, is a common intermediate of viral replication and a potent indicator of infection. Toll-like receptor 3 (TLR3) recognizes viral double-stranded RNA and its synthetic analog polyriboinosinic:polyribocytidylic acid (poly(I:C)). TLR3 is normally located in acidic endosomes where its luminal ectodomain (ECD) encounters dsRNA and induces type I interferon (IFN), inflammatory cytokine/chemokine production and dendritic cell (DC) maturation via the adaptor protein TICAM-1 (also called TRIF). Based on the different subcellular localization of cytosolic RNA receptors and TLR3, these receptors seem to play distinct roles in anti-viral immune responses.

Immunogen

Human Flag-tagged TLR3 stably expressed by Ba/F3 cells

Aliases

CD283, Toll-like receptor 3

Gene

Gene name: TLR3

References

1. Matsumoto, M et al; Establishment of a monoclonal antibody against human Toll-like receptor 3 that blocks double-stranded RNA-mediated signalling. Biochem Biophys Res Commun 2002, 293: 1364
2. Oshiumi, H et al; TICAM-1, an adaptor molecule that participates in Toll-like receptor 3-mediated interferon-beta induction. Nat Immunol 2003, 4: 161
3. Matsumoto, M et al; Subcellular localization of Toll-like receptor 3 in human dendritic cells. J Immunol 2003, 171: 3154
4. Burgener, I et al; Antibodies specific for human or murine Toll-like receptors detect canine leukocytes by flow cytometry. Vet Immunol Immunopathol 2008, 124; 184
5. Jorgenson, R et al; Human endometrial epithelial cells cyclically express Toll-like receptor 3 (TLR3) and exhibit TLR3-dependent responses to dsRNA. Human Immunology 2005; 66:469

Storage&stability

Product should be stored at 4°C. Under recommended storage conditions, product is stable for at least one year.

Precautions

For research use only. Not for use in or on humans or animals or for diagnostics. It is the responsibility of the user to comply with all local/state and federal rules in the use of this product. Hycult Biotech is not responsible for any patent infringements that might result from the use or derivation of this product.

We hereby certify that the above-stated information is correct and that this product has been successfully tested by the Quality Control Department. This product was released for sale according to the existing specifications. This document has been produced electronically and is valid without a signature.

Approved by Manager of QC	Date
Brenda Teunissen	14/10/2019

Do you have any questions or comments regarding this product? Please contact us via support@hycultbiotech.com.