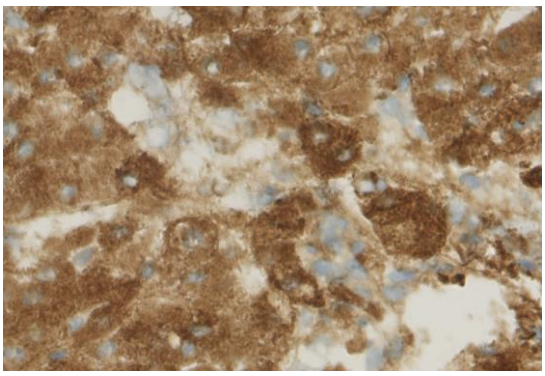
CERTIFICATE OF ANALYSIS – TECHNICAL DATA SHEET

Product name	C3b/iC3b/C3d, Human, clone 1H8		
Catalog number	HM2287-20UG		
Lot number	xxxxxXxxxx	**Expiry date**	MMM YYYY
Volume	200 µl	**Amount**	20 µg
Formulation	0.2 µm filtered in PBS+0.1%BSA+0.02%NaN3	**Concentration**	100 µg/ml
Host Species	Mouse IgG2a	**Conjugate**	None
Endotoxin	N.A.	**Purification**	Protein G
Storage	4°C		

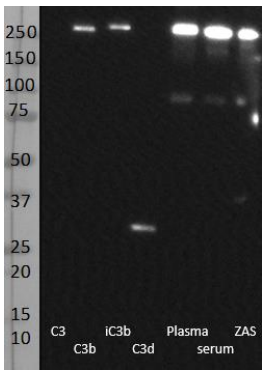
Application notes

	IHC-F	IHC-P	IF	FC	FS	IA	IP	W
Reference #				1			2	2
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



IHC-F: Frozen section of human liver tissue. HM2287 was diluted 1:1000.



W: Western blot analysis performed with human C3, C3b, iC3b, and C3d proteins; Human EDTA Plasma, serum and Zymosan activated serum(ZAS) samples with antibody HM2287 at 2 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.

- FC: Antibody 1H8 stains the extracellular domain of C3b/iC3b/C3d. As positive control Balb/c thymus (activated with anti-Mouse T-Cells and Human Serum) was used and as negative control secondary antibody. (Ref.1)
- IHC-F: Incubation with primary antibody (1:1000) for 30 minutes at 37 degrees
- W: A reduced sample treatment and SDS-Page was used. The band sizes are: ~180 kDa for C3b;~75 kDa for ß-chain C3b (which is visible in the samples); ~178 kDa for iC3b; ~33 kDa for C3d; and ~36kDa for C3dg (which can be seen in the ZAS sample). C3 is not recognized.

General Information

Description

The monoclonal antibody 1H8 recognizes Complement Factor C3b/iC3b/C3d, it specifically binds human C3 as well as the breakdown products C3b, iC3b and C3dg. C3 plays a central role in the activation of complement system. Its activation is required for both classical and alternative complement activation pathways. People with C3 deficiency are susceptible to bacterial infection. One form of C3-convertase, also known as C4b2a, is formed by a heterodimer of activated forms of C4 and C2. It catalyzes the proteolytic cleavage of C3 into C3a and C3b, generated during activation through the classical pathway as well as the lectin pathway. C3a is an anaphylotoxin and the precursor of some cytokines such as ASP, and C3b serves as an opsonizing agent. Factor I can cleave C3b into C3c and C3d, the latter of which plays a role in enhancing B cell responses. In the alternative complement pathway, C3 is cleaved by C3bBb, another form of C3-convertase composed of activated forms of C3 (C3b) and factor B (Bb). Once C3 is activated to

C3b, it exposes a reactive thioester that allows the peptide to covalently attach to any surface that can provide a nucleophile such as a primary amine or a hydroxyl group. Activated C3 can then interact with factor B. Factor B is then activated by factor D, to form Bb. The resultant complex, C3bBb, is called the alternative pathway (AP) C3 convertase. C3bBb is deactivated in steps. First, the proteolytic component of the convertase, Bb, is removed by complement regulatory proteins having decay-accelerating factor (DAF) activity. Next, C3b is broken down progressively to first iC3b, then C3c + C3dg, and then finally C3d. Factor I is the protease that performs these cuts with CR1 as cofactor. Clone 1H8 recognize separate, non-overlapping epitopes on C3 fragments. Levels of C3 in the blood may be measured to support or refute a particular medical diagnosis. For example, low C3 levels are associated with some types of kidney disease such as post-infectious glomerulonephritis and shunt nephritis.

Aliases Complement component C3.

References

1. Lindorfer, M et al. A novel approach to preventing the hemolysis of paroxysmal nocturnal hemoglobinuria: both complement-mediated cytolysis and C3 deposition are blocked by a monoclonal antibody specific for the alternative pathway of complement. Blood 2010, *115*:2283
2. Pawluczkowycz, A et al. Hematin Promotes Complement Alternative Pathway-Mediated Deposition of C3 Activation Fragments on Human Erythrocytes: Potential Implications for the Pathogenesis of Anemia in Malaria. J of Immunol, 2007, *179*: 5543

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.

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Date

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