

# **CERTIFICATE OF ANALYSIS – TECHNICAL DATA SHEET**

Product name C3b/iC3b, Human, clone 5G9

Catalog number HM2285-20UG

Lot number xxxxxXxxxx-X Expiry date MMM YYYY

**Volume** 200 μl **Amount** 20 μg

Formulation 0.2 μm filtered in PBS+0.1%BSA Concentration 100 μg/ml

Host Species Mouse IgG2a Conjugate None

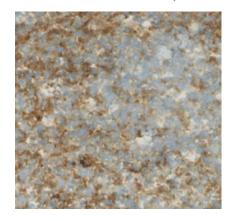
Endotoxin <24 EU/mg Purification Protein G

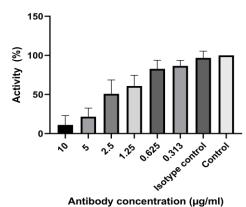
Storage 4°C

# Application notes

	IHC-F	IHC-P	IF	FC	FS	IA	IP	W
Reference #					1			
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 tonsil. Antibody HM2285 was used in a 1:200 dilution.

FS: Classical pathway inhibition by anti C3/iC3b antibody (HM2285)

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.

- IHC-F: Incubation with primary antibody (1:200) for 30 minutes at 37 degrees.
- FS: The inhibition of the classical pathway by HM2285, targeting C3/iC3b, was evaluated at varying concentrations using pooled human serum within the HK3010 human Classical Complement Pathway assay. An isotype control (MOPC-173, BioLegend) was also included to discern isotype-specific interactions. The pooled human serum served as a control.

### **General Information**

#### Description

The monoclonal antibody 5G9 recognizes Complement Factor C3b/iC3b, it specifically binds human C3 as well as the breakdown products C3b and iC3b. C3 plays a central role in the activation of the 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.

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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 5G9 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.

### References

 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, 115:11

#### 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

Do you have any questions or comments regarding this product? Please contact us via <a href="mailto:support@hycultbiotech.com">support@hycultbiotech.com</a>.

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