

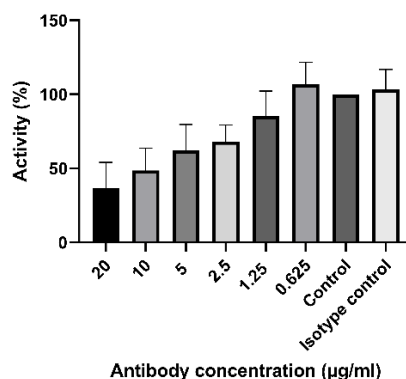
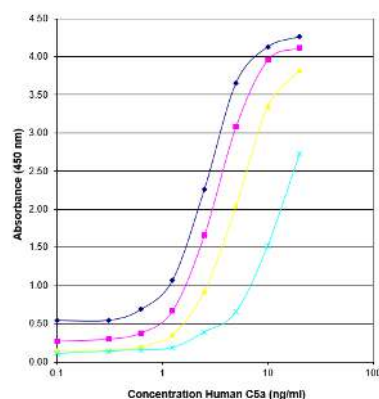
CERTIFICATE OF ANALYSIS – TECHNICAL DATA SHEET

Product name	C5/C5a, Human, clone 557		
Catalog number	HM2077-20UG		
Lot number	xxxxxXxxxx	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 #						1,2,3		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



IA: Immunoassay experiment:
HM2077 was used as a detection
antibody in different concentrations.

FS: Classical pathway inhibition by anti
C5/C5a antibody (HM2077)

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.

- IA: antibody was used as detection antibody in ELISA. (Ref.1)
- W: used as ascites at a 1/1000 dilution. Incubation 2h on nitrocellulose blotted samples. (Ref.1) Positive control: Recombinant C5; Negative control: C3.
- FS: The inhibition of the classical pathway by HM2077, targeting C5/C5a, 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 557 recognizes an epitope of complement factor 5 (C5) and C5a. The complement system is composed of over 30 proteins, activated in response to tissue injury, invading pathogens or other foreign surfaces. The complement pathways can be divided in the activation pathways and lytic pathway. The activation pathways lead via C3 to the cleavage of the fifth complement component C5. C5a was first described as a cleavage product of C5 with chemotactic and anaphylatoxic properties. Further characterization revealed that C5a is an essential part of the innate immune response and evidence now suggests that it may also play a role in adaptive immunity. Complement fragment C5a is a 74 residue pro-inflammatory polypeptide. C5a induces smooth muscle contraction, increases vascular permeability, causes degranulation of mast cells and basophils, and release of lysosomal enzymes. In addition C5a stimulates the directed migration of neutrophils, eosinophils, basophils and monocytes. C5a binds to at least two

seven-transmembrane domain receptors, C5aR (C5R1, CD88) and C5L2 (gpr77), expressed ubiquitously on a wide variety of cells but particularly on the surface of immune cells like macrophages, neutrophils and T cells. The former is a well-established receptor that initiates G-protein-coupled signaling via mitogen-activated protein kinase pathways, thereby by inducing synthesis of cytokines such as TNF-alpha, IL-1beta, IL-6 and IL-8. Its in vivo blockade greatly reduces inflammatory injury. Much less is known about C5L2, occupancy of which by C5a does not initiate increased intracellular Ca(2+). The widespread expression of C5a receptors throughout the body allows C5a to elicit a broad range of effects. Thus, C5a has been found to be a significant pathogenic driver in a number of immuno-inflammatory diseases. Nowadays C5a is also implicated in non-immunological functions associated with developmental biology, CNS development and neurodegeneration, tissue regeneration, and haematopoiesis. The antibody 557 is capable to inhibit the binding of C5a to the C5a receptor through a competitive mechanism, it does not block the cleavage of C5 into C5a and C5b.

Immunogen	BALB/c mice were immunized with human C5
Aliases	Complement C5, C3 and PZP-like alpha-2-macroglobulin domain-containing protein 4
Gene	Gene name: C5
References	<ol style="list-style-type: none"> 1. Klos, A et al, Detection of native human complement components C3 and C5 and their primary activation peptides C3a and C5a (anaphylatoxic peptides) by ELISAs with monoclonal antibodies. J Imm Meth 1988, 111: 241 2. Kola, A et al. Epitope mapping of a C5a neutralizing mAb using a combined approach of phage display, synthetic peptides and site-directed mutagenesis. Immunotechnology 1996, 2: 115 3. Kola, A. et al, Analysis of the C5a anaphylatoxin core domain using a C5a phage library selected on differentiated U937 cells. Mol Immunol 1999, 36: 145
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

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