RayBiotech, Inc.

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Certificate of Analysis and Data Sheet

Mouse Anti Human C4d

With HRP-Conjugated Second Antibody

Catalog No.	Target Species	Isotype
DS-MB-00163	Human	IgG1

Preparation

Immunogen: Native, from human plasma
Purification: Purified IgG prepared by affinity chromatography on Protein A

Formulation

Product Type: Monoclonal Antibody
Product Form: Purified IgG - liquid
Buffer Solution: Borate buffered saline pH 8.4
Preservative Stabilizers: 0.09% Sodium Azide (NaN_3)
Approx. Protein Concentration: IgG concentration 1.0mg/ml

Specificity

DS-MB-00163 recognizes the secreted protein complement component 4d (C4d). The complement 1 complex cleaves complement 4 (C4) to form C4b and C4a. C4b levels are strictly regulated. Single site cleavage of the C4b's alpha chain by Factor I forms iC4b and blocks C3 convertase, inhibiting opsonisation and activation of the classical pathway. This requires C4 binding protein or CR1 as a cofactor. iC4b is further degraded into C4d and C4c. C4d's short half life means that C4d is present in serum at high enough concentrations to make it a useful marker for classical complement activation.

C4 has been linked to susceptibility to systemic lupus erythematosous.

Immunohistology: It has been reported that this antibody works very well on acetone-fixed, frozen renal biopsies. Strong staining is observed in the glomeruli and in some cases the peritubular capillaries.

*DS-MB-00163 has given variable results on formalin-fixed, paraffin-embedded sections. It has been observed that pre-treatment with 88% formic acid for 20 minutes at room temperature is beneficial (6).

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Applications

Table Summary of antibody applications and working conditions

Options / Functions	YES	NO	Not determined	Recommended Work dilution or concentration
ELISA	.			1/5000 - 1/20000
Western Blot	.			
IHC - Frozen	.			1/100 - 1/750
IHC – Paraffin*	.			
Immunofluorescence	.			1/250 - 1/600

Note: Other applications are not tested yet. Optimal dilutions should be determined by each laboratory for each application.

Secondary Antibody Applications

Options / Functions	YES	NO	Not determined	Recommended Work dilution or concentration
Immunoassay (ELISA, Western blot)	•			1:5000-1:10000

Storage

Store at +4°C or at -20°C if preferred. Storage in frost-free freezers is not recommended.
This product should be stored undiluted. Avoid repeated freezing and thawing as this may denature the antibody. Should this product contain a precipitate we recommend microcentrifugation before use.
Shelf Life: 18 months from date of dispatch

References

1. Mauiyyedi, S. *et al*. (2002) Acute humoral rejection in kidney transplantation: II. Morphology, immunopathology, and pathologic classification. J Am Soc Nephrol. 13: 779-787.

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3. Knechtle, S.J. *et al* (2003) Campath-1H induction plus rapamycin monotherapy for renal transplantation: results of a pilot study. Am J Transplant. 3: 722-730.

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5. Rogers, J. *et al*. (1992) Complement Activation by beta-amyloid in Alzheimer disease. PNAS 89:10016-10020.

6. Stoltzner, S. E. *et al.* (2000) Temporal accural of complement proteins in amyloid plaques in Down's syndrome with Alzheimer's disease. Am J. Pathol. 156: 489-499.