



Rabbit anti-C3d

Cat. No.: RBK034-05 (0.5 ml Concentrate); RBG034 (6 ml Ready-to-use)

Instructions for use

Intended use

This antibody is designed for the specific localisation of complement protein C3d in formalin-fixed, paraffin-embedded tissue sections and in frozen sections. Anti-C3d antibody is intended for in vitro diagnostic use.

Specifications

Specificity:	Human Complement C3d protein
Clone:	polyclonal
Species reactivity:	Human +, others not tested

Summary and Description

The complement protein C3d plays a major role in the activation of complement system. C3d deposition in the peritubular capillaries (PTC) of renal transplants is indicative of acute rejection.

Pfaltz et al. (2009) detected C3d in the epidermal basement membrane in 97% (31/32) cases of bullous pemphigoid (BP). All normal control cases were negative. In the same study 27% (3/11) cases of pemphigus vulgaris (PV) showed intercellular deposition of C3d. The authors consider C3d staining helpful for diagnosis of BP, especially in the cases in which only formalin-fixed (FFPE) tissue is available for analysis.

Reagent provided

Purified rabbit polyclonal antibody in PBS pH 7.4 with carrier protein and preservative for stabilisation in the following formats:

Concentrate:	0.5 ml	(Cat. No. RBK034-05)
Ready-to-use:	6 ml	(Cat. No. RBG034)

Dilution of primary antibody

Dilution of Zytomed Systems' concentrated antibody depends on the detection system used. The final working dilution must always be determined by the user. The elaboration of staining protocol should be done by an experienced specialist. For Zytomed Systems' recommendations see chapter 'Staining procedure'.

Storage and handling

The antibody should be stored at 2-8°C without further dilution.

Dilutions of the concentrated antibody should be done in a suitable antibody dilution buffer (e.g. ZUC025 from Zytomed Systems). The diluted antibody should be stored at 2-8°C after use. The stability of this working solution depends on various parameters and has to be confirmed by appropriate controls.

The antibody provided is suitable for use until the expiry date indicated on the label, if stored at 2-8°C. Do not use product after the expiry date. Positive and negative controls should be run simultaneously with all specimens. If unexpected staining is observed which cannot be explained by variations in laboratory procedures and a problem with the antibody is suspected, contact Zytomed Systems' technical support or your local distributor.

Precautions

Use through qualified personnel only.

Wear protective clothing to avoid contact of reagents and specimens with eye, skin and mucous membranes. If reagents or specimens come in contact with sensitive area, wash with large amounts of water.

Microbial contamination of the reagent must be avoided, since otherwise non-specific staining may occur.

Sodium azide (NaN_3), used for stabilisation, is not considered hazardous material in the concentration used.

Reaction of sodium azide with lead or copper in drainage pipes can result in the formation of highly explosive metallic azides. Sodium azide should be discarded in a large volume of running water to avoid formation of deposits. A material safety data sheet (MSDS) for the pure substance is available upon request.

Staining procedure for formalin-fixed paraffin sections

Refer to the following table for conditions specifically recommended for this antibody. Also refer to detection system data sheets for guidance on specific staining protocols or other requirements.

Parameters	Zytomed Systems recommendations
*Pre-treatment	Heat Induced Epitope Retrieval (for example in Citrate Buffer pH 6.0 (ZUC028))
*Control tissue	acute rejected kidney transplant
*Working dilution	1:100-1:500 (for concentrates)
*Incubation time	30 - 60 minutes

Quality control

The recommended positive control tissue for this antibody is tissue from an acutely rejected kidney transplant. We recommend carrying out a positive and a negative control with every staining run. Please refer to the instructions of the detection system for guidance on general quality control procedures.

Troubleshooting

If you observe unusual staining or other deviations from the expected results please read these instructions carefully, refer to the instructions of the detection system for relevant information or contact your local distributor.

Expected results

This antibody stains positive in the cytoplasmic membrane or in the cytoplasm in formalin-fixed, paraffin-embedded tissue sections. Further details about the expression pattern of C3d can be found in the chapter 'Summary and Description'. Interpretation of the staining results is solely the responsibility of the user. Any experimental result should be confirmed by a medically established diagnostic procedure.

Limitations of the Procedure

Immunohistochemistry is a complex technique involving both histological and immunological detection methods. Tissue processing and handling prior to immunostaining, for example variations in fixation and embedding or the inherent nature of the tissue can cause inconsistent results (Nadji and Morales, 1983). Endogenous peroxidase, pseudoperoxidase activity in erythrocytes or biotin may cause non-specific staining depending on the detection system used. Tissues containing Hepatitis B Surface Antigen (HBsAg) may give false positive results with HRP (horse radish peroxidase) detection systems (Omata *et al*, 1980). Inadequate counterstaining and mounting can influence the interpretation of the results.

Zytomed Systems warrants that the product will meet all requirements described from its shipping date until the expiry date is reached, if the product is stored and utilised as recommended. No additional guarantees can be given. Under no circumstances shall Zytomed System be liable for any damages arising out of the use of the reagent provided.

Performance characteristics

Zytomed Systems has conducted studies to evaluate the performance of the antibody for use with a standard detection system. The product has been found to be sensitive and specific to the antigen of interest with minimal or no cross-reactivity.

Bibliography

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Bickerstaff A *et al* Am J Pathol 2008, 173:347-357
Kuypers DR *et al* Transplantation 2003, 76:102-108
Eggertsen G *et al* APMIS 2001, 109:825-834
Nadji M and Morales AR 1983, Ann NY Acad Sci 420:134-139
Omata M *et al* 1980, Am J Clin Pathol 73: 626-632



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Explanations of the symbols on the product

label

Symbols are used in accordance with ISO 15223-1. Further symbols on the product label might be:



RUO

GSH07: Warning / Attention

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