

CD61

Concentrated and Prediluted Monoclonal Antibody

Control Number: 901-3139-090917

Catalog Number:	ACI 3139 A, C	API 3139 AA
Description:	0.1, 1.0 ml, concentrated	6.0 ml, prediluted
Dilution:	1:50-1:100	Ready-to-use
Diluent:	Renoir Red	N/A

Intended Use:

For In Vitro Diagnostic Use

CD61 [2f2] is a mouse monoclonal antibody that is intended for laboratory use in the qualitative identification of CD61 protein by immunohistochemistry (IHC) in formalin-fixed paraffin-embedded (FFPE) human tissues. The clinical interpretation of any staining or its absence should be complemented by morphological studies using proper controls and should be evaluated within the context of the patient's clinical history and other diagnostic tests by a qualified pathologist.

Summary and Explanation:

The CD61 antigen, also known as GPIIIa, is a glycoprotein of 105 kD and has been shown to be expressed in myeloid cells, monocytes, endothelial cells, smooth muscle cells, macrophages and platelets (1-6). CD61 has been shown to be useful in evaluating megakaryocytopoiesis as it relates to myelodysplastic disorders, acute myeloid leukemias and acute megakaryoblastic leukemias (2,3). Immunohistochemistry with CD61 has also been useful in identifying platelet adhesion in advanced atherosclerosis and was helpful in identifying fat embolism in pulmonary tissue (4,5). The identification of CD61 expression in patients with insudative platelet arteriopathy helped facilitate recognition of vascular calcineurin inhibitor toxicity in renal allograft biopsies (6).

Principle of Procedure:

Antigen detection in tissues and cells is a multi-step immunohistochemical process. The initial step binds the primary antibody to its specific epitope. After labeling the antigen with a primary antibody, a secondary antibody is added to bind to the primary antibody. An enzyme label is then added to bind to the secondary antibody; this detection of the bound antibody is evidenced by a colorimetric reaction.

Source: Mouse monoclonal

Species Reactivity: Human; others not tested

Clone: 2f2

Isotype: IgG1

Total Protein Concentration: ~10 mg/ml. Call for lot specific Ig concentration.

Epitope/Antigen: Recombinant protein encoding part of the external domain of human CD61

Cellular Localization: Cell membrane / cytoplasm

Positive Tissue Control: Bone marrow

Known Applications:

Immunohistochemistry (formalin-fixed paraffin-embedded tissues)

Supplied As: Buffer with protein carrier and preservative

Storage and Stability:

Store at 2°C to 8°C. Do not use after expiration date printed on vial. If reagents are stored under conditions other than those specified in the package insert, they must be verified by the user. Diluted reagents should be used promptly; any remaining reagent should be stored at 2°C to 8°C.

Protocol Recommendations:

Peroxide Block:

Block for 5 minutes with Biocare's Peroxidized 1.

Pretreatment: Perform heat retrieval using Biocare's Diva Decloaker. Refer to the Diva Decloaker data sheet for specific instructions.

Protein Block (Optional): Incubate for 5-10 minutes at RT with Biocare's Background Punisher.

Primary Antibody: Incubate for 30 minutes at RT.

Protocol Recommendations Cont'd:

Probe: Incubate for 10 minutes at RT with a secondary probe.

Polymer: Incubate for 10 minutes at RT with a tertiary polymer.

Chromogen:

Incubate for 5 minutes at RT with Biocare's DAB – OR – Incubate for 5-7 minutes at RT with Biocare's Warp Red.

Counterstain:

Counterstain with hematoxylin. Rinse with deionized water. Apply Tacha's Bluing Solution for 1 minute. Rinse with deionized water.

Technical Note:

This antibody has been standardized with Biocare's MACH 4 detection system. It can also be used on an automated staining system and with other Biocare polymer detection kits. Use TBS buffer for washing steps.

Limitations:

The optimum antibody dilution and protocols for a specific application can vary. These include, but are not limited to fixation, heat-retrieval method, incubation times, tissue section thickness and detection kit used. Due to the superior sensitivity of these unique reagents, the recommended incubation times and titers listed are not applicable to other detection systems, as results may vary. The data sheet recommendations and protocols are based on exclusive use of Biocare products. Ultimately, it is the responsibility of the investigator to determine optimal conditions. The clinical interpretation of any positive or negative staining should be evaluated within the context of clinical presentation, morphology and other histopathological criteria by a qualified pathologist. The clinical interpretation of any positive or negative staining should be complemented by morphological studies using proper positive and negative internal and external controls as well as other diagnostic tests.

Quality Control:

Refer to CLSI Quality Standards for Design and Implementation of Immunohistochemistry Assays; Approved Guideline-Second edition (I/LA28-A2) CLSI Wayne, PA USA (www.clsi.org). 2011

Precautions:

1. This antibody contains less than 0.1% sodium azide. Concentrations less than 0.1% are not reportable hazardous materials according to U.S. 29 CFR 1910.1200, OSHA Hazard communication and EC Directive 91/155/EC. Sodium azide (NaN₃) used as a preservative is toxic if ingested. Sodium azide may react with lead and copper plumbing to form highly explosive metal azides. Upon disposal, flush with large volumes of water to prevent azide build-up in plumbing. (Center for Disease Control, 1976, National Institute of Occupational Safety and Health, 1976) (7)
2. Specimens, before and after fixation, and all materials exposed to them should be handled as if capable of transmitting infection and disposed of with proper precautions. Never pipette reagents by mouth and avoid contacting the skin and mucous membranes with reagents and specimens. If reagents or specimens come into contact with sensitive areas, wash with copious amounts of water. (8)
3. Microbial contamination of reagents may result in an increase in nonspecific staining.
4. Incubation times or temperatures other than those specified may give erroneous results. The user must validate any such change.
5. Do not use reagent after the expiration date printed on the vial.
6. The SDS is available upon request and is located at <http://biocare.net>.

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References:

1. Jiménez-Marín A, *et al.* Molecular characterization and expression analysis of the gene coding for the porcine beta(3) integrin subunit (CD61). *Gene*. 2008 Jan 31; 408(1-2):9-17.
2. Fox SB, *et al.* Megakaryocytes in myelodysplasia: an immunohistochemical study on bone marrow trephines. *Histopathology*. 1990 Jul; 17(1):69-74.
3. Thiele J, *et al.* Atypical micromegakaryocytes, promegakaryoblasts and megakaryoblasts: a critical evaluation by immunohistochemistry, cytochemistry and morphometry of bone marrow trephines in chronic myeloid leukemia and myelodysplastic syndromes. *Virchows Arch B Cell Pathol Incl Mol Pathol*. 1992; 62(5):275-82.
4. Gonzalez J, *et al.* High fat diet induces adhesion of platelets to endothelium in two models of dyslipidemia. *J Obes*. 2014; 2014:591270.
5. Neri M, *et al.* CD61 and fibrinogen immunohistochemical study to improve the post-mortem diagnosis in a fat embolism syndrome clinically demonstrated by transesophageal echocardiography. *Forensic Sci Int*. 2010 Oct 10; 202(1-3):e13-7.
6. Meehan SM, *et al.* Platelet CD61 expression in vascular calcineurin inhibitor toxicity of renal allografts. *Hum Pathol*. 2008 Apr; 39(4):550-6.
7. Center for Disease Control Manual. Guide: Safety Management, NO. CDC-22, Atlanta, GA. April 30, 1976 "Decontamination of Laboratory Sink Drains to Remove Azide Salts."
8. Clinical and Laboratory Standards Institute (CLSI). Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline-Fourth Edition CLSI document M29-A4 Wayne, PA 2014.

Troubleshooting:

Follow the antibody specific protocol recommendations according to data sheet provided. If atypical results occur, contact Biocare's Technical Support at 1-800-542-2002.

