



Monoclonal Mouse Anti-Human Beta-Catenin Clone β-Catenin-1

Code M3539

Intended use

For in vitro diagnostic use

Monoclonal Mouse Anti-Human Beta-Catenin, Clone β -Catenin-1 is intended for use in immunohistochemistry (IHC). Results aid in the classification of of desmoid tumors (1) Differential classification is aided by the results from a panel of antibodies. 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. This antibody is intended to be used after the primary diagnosis of tumor has been made by conventional histopathology using nonimmunological histochemical stains.

Summary and explanation

The catenins are structurally related cytoplasmic proteins which have been classified as alpha (α), beta (β), and gamma (γ) according to their electrophoretic mobility (3, 4). The β -catenin gene is located on chromosome 3p21 and encodes a 88 kD protein (3, 4). This cytoplasmic protein is multi-functional, playing an essential role in the cadherin-mediated anchoring and organization of the cytoskeleton (3). Beta-catenin is also involved in regulation of gene expression as a mediator of the Wnt signaling pathway. Cellular β -catenin levels are tightly regulated by a multi-protein complex comprised of serine/threonine kinase GSK3 β , the APC tumor suppressor gene product and axin, which facilitates phosphorylation and subsequent degradation of the β -catenin protein. Dysregulation of β -catenin degradation leads to cytoplasmic accumulation of the protein, followed by translocation to the nucleus. Nuclear β -catenin forms complexes with DNA binding proteins such as TCF and LEF, activating gene transcription (5).

Antibodies to beta-catenin have also been shown to aid in the classification of colorectal cancer (2) and colon adenoma (3).

Refer to *Dako General Instructions for Immunohistochemical Staining* or the detection system instructions of IHC procedures for: Principle of Procedure, Materials Required, Not Supplied, Storage, Specimen Preparation, Staining Procedure, Quality Control, Troubleshooting, Interpretation of Staining, General Limitations.

Reagent provided

Monoclonal mouse antibody provided in liquid form as tissue culture supernatant in 0.05 mol/L Tris-HCl, pH 7.2 and 0.015 mol/L sodium azide. This product contains stabilizing protein.

Clone: β-Catenin-1. Isotype: IgG1, kappa

Mouse IgG concentration: See label on vial.

The protein concentration between lots may vary without influencing the optimal dilution. The titer of each individual lot is compared and adjusted to a reference lot to ensure a consistent immunohistochemical staining performance from lot-to-lot.

Immunogen

Recombinant C-terminal β-catenin fusion protein (6)

Specificity

Anti-beta-catenin, clone β -catenin-1 recognized human β -catenin protein in Western blots of human epithelial A431 cells and mouse β -catenin in blots of mouse fibroblast NIH/3T3 cells. No cross-reactivity with α and γ -catenin was observed (6).

Precautions

- 1. For in vitro diagnostic use.
- 2. For professional users.
- 3. This product contains sodium azide (NaN₃), a chemical highly toxic in pure form. At product concentrations, though not classified as hazardous, NaN₃ may react with lead and copper plumbing to form highly explosive build-ups of metal azides. Upon disposal, flush with large volumes of water to prevent metal azide build-up in plumbing.
- 4. As with any product derived from biological sources, proper handling procedures should be used.
- 5. Wear appropriate Personal Protective Equipment to avoid contact with eyes and skin.
- 6. Unused reagents should be disposed of according to local, State, and Federal regulations.

Storage

Store at 2–8 °C. Do not use after expiration date stamped on vial. If reagents are stored under any conditions other than those specified, the conditions must be verified by the user. There are no obvious signs to indicate instability of this product. Therefore, positive and negative controls should be run simultaneously with patient specimens. If unexpected staining is observed which cannot be explained by variations in laboratory procedures and a problem with the antibody is suspected, contact Dako Technical Support.

Specimen preparation

Paraffin sections: Anti-beta-catenin, clone β-catenin-1 can be used on formalin-fixed, paraffin-embedded tissue sections.

The deparaffinized tissue sections must be treated with heat prior to the IHC staining procedure. Target retrieval involves immersion of tissue sections in a pre-heated buffer solution and maintaining heat, either in a water bath (95–99 °C) or in a steamer (95–99 °C). For greater adherence of tissue sections to glass slides, the use of silanized slides (Code S3003) is recommended. Target Retrieval Solution (Code S1700) or 10x Concentrate (Code S1699) is recommended using a 20-minute heating protocol.

Staining procedure

These are guidelines only. Optimal conditions may vary depending on specimen type and preparation method, and should be validated individually by each laboratory. The performance of this antibody should be established by the user when utilized with other manual staining systems or automated platforms.

<u>Dilution:</u> M3539 may be used at a dilution of 1:200. Dilute the antibody in Dako Antibody Diluent (Code S0809). The recommended negative control reagent is Dako Negative Control, Mouse IgG1 (Code X0931).

<u>Visualization:</u> The recommended visualization system is Dako EnVision+ kits, e.g. Code K4005. Follow the recommended procedure for the detection system selected.

Quality control: Positive and negative control tissues as well as negative control reagent should be run simultaneously using the same protocol as the patient specimens.

Staining interpretation

The cellular staining pattern is membranous. Neoplastic cells can display nuclear and diffuse cytoplasmatic staining.

Performance characteristics

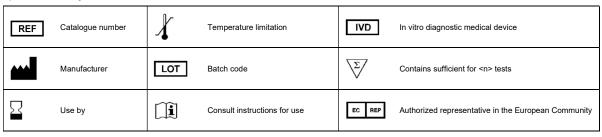
<u>Normal tissues:</u> In colon, the epithelial cells show a moderate to strong staining reaction. In liver, hepatocytes show a weak to moderate staining reaction (7).

<u>Abnormal tissues:</u> The antibody labeled 24/30 sporadic desmoid tumors and 8/12 desmoid tumors in patients with familial adenomatous polyposis (1). The antibody labeled 2/2 colon adenomas (7).

References

- 1. Carlson JW, Fletcher CDM. Immunohistochemistry for β-catenin in the differential diagnosis of spindle cell lesions: analysis of a series and review of the literature. Histopathol 2007;51:509-14.
- 2. Iwamoto M, Ahnen DJ, Franklin WA, Maltzman TH. Expression of β-catenin and full-length APC protein in normal and neoplastic colonic tissue. Carcinogenesis 2000;21:1935-40.
- 3. Bracke ME, Van Roy FM, Mareel MM. The E-cadherin/catenin complex in invasion and metastasis. Cur Top Microbiol Immunol 1996; 213 (Pt 1):123-61.
- 4. Ozawa M, Baribault H, Kemler R. The cytoplasmic domain of the cell adhesion molecule uvomorulin associates with three independent proteins structurally related in different species. EMBO J 1989;8:1711-7.
- 5. Willert K, Nusse R. Curr Opin Genet Dev 1998;8:95-102.
- 6. Antibody Certification. Report on file, Dako.
- 7. Verification Test Results for: GA702 FLEX Monoclonal Mouse Anti-Human Beta-Catenin Clone β-Catenin-1, Ready-to-Use (Dako Omnis). 2013. Report on file, Dako. D18293.

Explanation of symbols





Agilent Technologies, Inc. 5301 Stevens Creek Blvd. Santa Clara, CA 95051 United States

Tel. +44 161 492 7050 www.agilent.com

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