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anti-beta Actin antibody

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Overview

Quantity:	100 μg
Target:	beta Actin (ACTB)
Reactivity:	Human, Mouse, Chicken, Rabbit, Pig, Cow, Hamster, Goat, Fish
Host:	Mouse
Clonality:	Monoclonal
Application:	Western Blotting (WB), ELISA

Product Details

Immunogen:	A synthetic peptide DDDIAALVVDNGSG coupled - KLH
Clone:	2D1D10
Isotype:	IgG
Specificity:	THE TM beta Actin Antibody, mAb, Mouse reacts with mouse, rabbit, chicken, human, hamster, cow, goat, fish, and pig. It has not yet been tested in other species.
Cross-Reactivity (Details):	Cross reactivity in other species has not yet been tested.
Purification:	Protein G affinity column

Target Details

Target:	beta Actin (ACTB)
Alternative Name:	beta Actin (ACTB Products)
Background:	Beta-actin is one of the six different actin isoforms that have been identified. The actin

Target Details

molecules found in cells of various species and tissues tend to be very similar in their immunological and physical properties. As a consequence, it has been difficult to produce potent antisera against this protein. Therefore, the availability of monoclonal antibodies to beta-actin provides a useful and specific tool in the study of the intracellular distribution of beta-actin and the static and dynamic aspects of the cytoskeleton.

Pathways:

Myometrial Relaxation and Contraction, Cell-Cell Junction Organization, Maintenance of Protein Location, Phototransduction

Application Details

Application Notes:

Working concentrations for specific applications should be determined by the investigator. The appropriate concentrations may be affected by secondary antibody affinity, antigen concentration, the sensitivity of the method of detection, temperature, the length of the incubations, and other factors. The suitability of this antibody for applications other than those listed below has not been determined. The following concentration ranges are recommended starting points for this product.

ELISA: 0.1-1.0 µg/mL
Western blot: 1.0 µg/mL

Other applications: user-optimized

Restrictions:

For Research Use only

Lyophilized

Handling

Format:

Reconstitution:	Reconstitute the lyophilized antibody with deionized water to the final concentration of 0.5 mg/mL.
Buffer:	PBS, pH 7.4, containing 0.02 % Sodium azide, without BSA
Preservative:	Sodium azide
Precaution of Use:	WARNING: Reagents contain sodium azide. Sodium azide is very toxic if ingested or inhaled. Avoid contact with skin, eyes, or clothing. Wear eye or face protection when handling. If skin or eye contact occurs, wash with copious amounts of water. If ingested or inhaled, contact a physician immediately. Sodium azide yields toxic hydrazoic acid under acidic conditions. Dilute azide-containing compounds in running water before discarding to avoid accumulation of potentially explosive deposits in lead or copper plumbing.

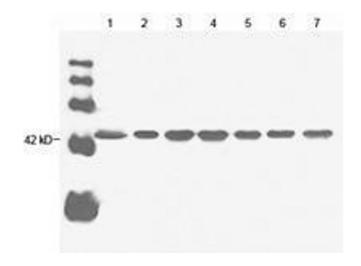
Handling

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Storage:	4 °C/-20 °C
Storage Comment:	The antibody is stable in lyophilized form if stored at -20°C or below. The reconstituted antibody can be stored for 2-3 weeks at 2-8°C. For long term storage, aliquot and store at -20°C or below. Avoid repeated freezing and thawing cycles.
Publications	
Product cited in:	Zhao, Mo, Li, Zou, Cheng, Sun, Xiong, Guan, Lei: "NOTCH-induced aldehyde dehydrogenase 1A1

Zhao, Mo, Li, Zou, Cheng, Sun, Xiong, Guan, Lei: "NOTCH-induced aldehyde dehydrogenase 1A1 deacetylation promotes breast cancer stem cells." in: **The Journal of clinical investigation**, (2014) (PubMed).

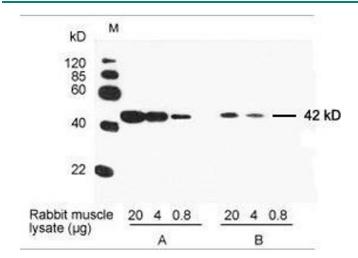
Srivastava, Reinoso, Singh, Fernandez-Bueno, Martino, Garcia-Gutierrez, Pastor, Corell: "Flow cytometry assessment of the purity of human retinal pigment epithelial primary cell cultures." in: **Journal of immunological methods**, Vol. 389, Issue 1-2, pp. 61-8, (2013) (PubMed).

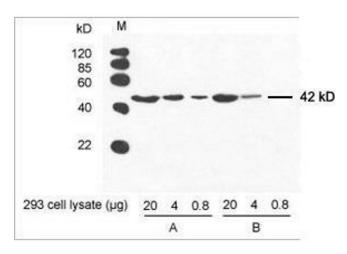
Validation report #102827 for Flow Cytometry (FACS)



Western Blotting

Image 1. Lane 1: 20 μg Hela cell lysate Lane 2: 20 μg sp2/0 cell lysate Lane 3: 20 μg goat muscle lysate Lane 4: 20 μg rabbit muscle lysate Lane 5: 20 μg chicken muscle lysate Lane 6: 20 μg CHO cell lysate Lane 7: 20 μg fish muscle lysate Primary antibody: 1 μg/mL Anti-beta-actin Monoclonal Antibody (Mouse) (ABIN396859) Secondary antibody: Goat Anti-Mouse IgG (H&L) [HRP] Polyclonal Antibody (ABIN398387, 1: 20,000)





Western Blotting

Image 2. Primary antibody: A. 0.1 μg/mL Anti-beta-actin Monoclonal Antibody (Mouse) (ABIN396859) B. 0.1 μg/mL Anti-beta-actin Monoclonal Antibody (Mouse) (Company X) Secondary antibody: Goat Anti-Mouse IgG (H&L) [HRP] Polyclonal Antibody (1: 10,000, ABIN398387) The signal was developed with LumiSensor HRP Substrate Kit (ABIN769939)

Western Blotting

Image 3. Primary antibody: A. 0.1 μg/mL Anti-beta-actin Monoclonal Antibody (Mouse) (ABIN396859) B. 0.1 μg/mL Anti-beta-actin Monoclonal Antibody (Mouse) (Company X) Secondary antibody: Goat Anti-Mouse IgG (H&L) [HRP] Polyclonal Antibody (1: 10,000, ABIN398387) The signal was developed with LumiSensor HRP Substrate Kit (ABIN769939)





Successfully validated (Western Blotting (WB))

by Dr. Randy Brutkiewicz Laboratory, Department of Microbiology and Immunology, Indiana University School of Medicine

Report Number: 102832

Date: Feb 20 2018

Target:	ACTB
Method validated:	Western Blotting (WB)
Positive Control:	lysates from HEK293 cells untransfected or transfected with human MR1 cDNA
Notes:	Passed. ABIN962807 recognizes human beta actin in HEK293 cell lysates.
Primary Antibody:	ABIN962807
Secondary Antibody:	goat anti-rabbit Dye-IR800 conjugated antibody (Advansta, R-05060-250, lot 17083179)
Protocol:	 Grow HEK293 cells in DMEM medium (Lonza, 12-614F, lot 0000618582) supplemented with serum (Hyclone, SH30071.03, lot AAG205460) and antibiotics (Hyclone, SV30010, lot J150013), at 37°C and 5% CO₂ dish to 70-90% confluency. Transfect cells with pCDNA 3.1 neo (-) (Invitrogen) containing human MR1 cDNA (Genecopoeia) using Polyethylenimine (Polysciences, 23966) following the manufacturer's instructions. Lyse cells in cold lysis buffer (10mM Tris pH7.4, 150mM NaCl, 0.5mM EDTA, 2% CHAPS). Determine total protein content of the lysates using Commassie Protein Assay Reagent (Thermo Scientific, 1856209, lot NL179252). Denature 200µg total protein for 5min at 95°C in 20µl Laemmli SDS sample buffer and subsequently separate them on a SDS-PAGE gel using Acrylamide/Bis Premixed (Bio-Rad, 61-0125, lot 260000477) for 2-3h at 100V. Transfer proteins onto PVDF membrane (Millipore, IPVH00010, lot K5AA6843U) with a Western blotting system for ON at 4°C at 150mA. Block the membrane with blocking buffer (2% BSA/PBS/0.05%Tween-20) for 1h at RT. Incubate membrane with: loading control rabbit anti beta-actin (antibodies-online, ABIN962807) diluted 1:500 in blocking buffer ON at 4°C. primary rabbit anti-MR1 antibody (antibodies-online, ABIN1537116, lot SA111213CH) diluted 1:1000 in blocking buffer ON at 4°C. Wash membrane 3x for 10min with PBS/0.05%Tween-20. Incubate membrane with secondary goat anti-rabbit Dye-IR800 conjugated antibody
	(Advansta, R-05060-250, lot 17083179) diluted 1:10000 in PBS/0.05% Tween-20 for 1h at RT.

• Wash membrane 3x for 10min with PBS/0.05% Tween-20.

• Reveal protein bands using an Odyssey imaging system (LI-COR Biosciences).

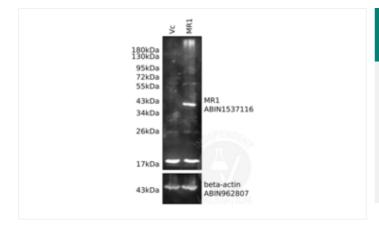


♦ Validation report #102832 for Western Blotting (WB)

Experimental Notes:

The beta Actin antibody ABIN962807 reveals a protein of the expected molecular weight of beta actin in lysates of human HEK293 cells.

Image for Validation report #102832



Validation image no. 1 for anti-Actin, beta (ACTB) antibody (ABIN962807)

Lysates from human MR1-expressing HEK293 cells (MR1) and vector control cells (Vc) were resolved on a 10% SDS-PAGE gel for Western blotting analysis using antibodies specific for MR1 (ABIN1537116) and beta-actin (ABIN962807).