BOSTER
antibody and ELISA experts

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3942 B Valley Ave, Pleasanton, CA 94566
888-466-3604 | support@bosterbio.com | www.bosterbio.com

Anti-IDH1 Antibody Picoband™ (monoclonal, 16H7)

Catalog Number: M00129-1

About IDH1

Isocitrate dehydrogenase 1 (NADP+), soluble is an enzyme that in humans is encoded by the IDH1 gene. Isocitrate dehydrogenases catalyze the oxidative decarboxylation of isocitrate to 2-oxoglutarate. These enzymes belong to two distinct subclasses, one of which utilizes NAD (+) as the electron acceptor and the other NADP (+). Five isocitrate dehydrogenases have been reported: three NAD (+)-dependent isocitrate dehydrogenases, which localize to the mitochondrial matrix, and two NADP (+)-dependent isocitrate dehydrogenases, one of which is mitochondrial and the other predominantly cytosolic. Each NADP (+)-dependent isozyme is a homodimer. The protein encoded by this gene is the NADP (+)-dependent isocitrate dehydrogenase found in the cytoplasm and peroxisomes. It contains the PTS-1 peroxisomal targeting signal sequence. The presence of this enzyme in peroxisomes suggests roles in the regeneration of NADPH for intraperoxisomal reductions, such as the conversion of 2, 4-dienoyl-CoAs to 3-enoyl-CoAs, as well as in peroxisomal reactions that consume 2-oxoglutarate, namely the alpha-hydroxylation of phytanic acid. The cytoplasmic enzyme serves a significant role in cytoplasmic NADPH production. Alternatively spliced transcript variants encoding the same protein have been found for this gene.

Overview

Product Name	Anti-IDH1 Antibody Picoband™ (monoclonal, 16H7)
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-IDH1 Antibody Picoband™ (monoclonal, 16H7) catalog # M00129-1. Tested in Flow Cytometry, IF, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	Flow Cytometry, IF, ICC, WB
Clonality	Monoclonal 16H7
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.
Storage Instructions	Store at -20˚C for one year from date of receipt. After reconstitution, at 4˚C for one month. It can also be aliquotted and stored frozen at -20˚C for six months. Avoid repeated freeze-thaw cycles.
Host	Mouse
Uniprot ID	O75874

Technical Details

Immunogen	A synthetic peptide corresponding to a sequence at the C-terminus of human IDH1, different from the related mouse and rat sequences by one amino acid.
Predicted Reactive Species	Hepatitis Virus
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Mouse IgG (EK1001) for Western blot, and HRP Conjugated anti- Mouse IgG Super Vision Assay Kit (SV0001-1) for ICC.

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Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Mouse IgG1
Form	Liquid
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	Dilute the sample so that the expected range of concentrations fall within the detection range of this kit. If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples. Some PubMed article(s) citing the expression level of this target are as follows: Boster Bio's internal QC testing used: Western blot, 0.1-0.5ug/ml, Human, Mouse, Rat Immunocytochemistry/Immunofluorescence, 2ug/ml, Human Flow Cytometry, 1-3ug/1x10^6 cells, Human

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Anti-IDH1 Antibody Picoband™ (monoclonal, 16H7) (M00129-1) Images

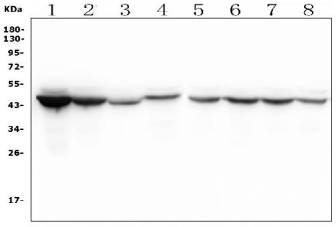

Figure 1. Western blot analysis of IDH1 using anti-IDH1 antibody (M00129-1).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: human HepG2 tissue lysates,
Lane 2: human Caco-2 whole cell lysates,
Lane 3: human U-87MG whole cell lysates,
Lane 4: human THP-1 whole cell lysates,
Lane 5: human Hela whole cell lysates.
Lane 6: human K562 whole cell lysates.
Lane 7: human PC-3 whole cell lysates.
Lane 8: human HEK293 whole cell lysates.
After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-IDH1antigen affinity purified polyclonal antibody (Catalog # M00129-1) at 0.5 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for IDH1 at approximately 47KD. The expected band size for IDH1 is at 47KD.

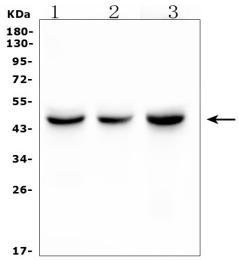

Figure 2. Western blot analysis of IDH1 using anti-IDH1 antibody (M00129-1).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: rat liver tissue lysates,
Lane 2: rat RH35 whole cell lysates,
Lane 3: mouse liver whole cell lysates,
After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-IDH1antigen affinity purified polyclonal antibody (Catalog # M00129-1) at 0.5 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for IDH1 at approximately 47KD. The expected band size for IDH1 is at 47KD.

Figure 3. IF analysis of IDH1 using anti-IDH1 antibody (M00129-1).

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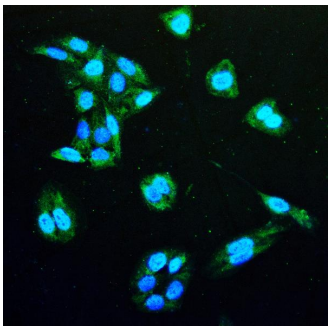

IDH1 was detected in immunocytochemical section of U2OS cell. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2ug/mL mouse anti-IDH1 Antibody (M00129-1) overnight at 4°C. DyLight488 Conjugated Goat Anti-Mouse IgG (BA1126) was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

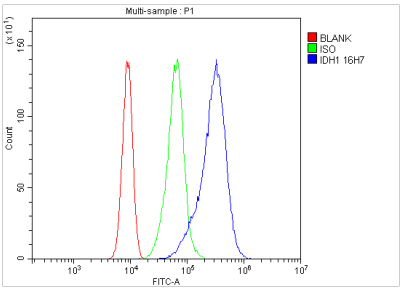

Figure 4. Flow Cytometry analysis of CACO-2 cells using anti-IDH1 antibody (M00129-1).
Overlay histogram showing CACO-2 cells stained with M00129-1 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-IDH1 Antibody (M00129-1, 1ug/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10ug/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1ug/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.