

Anti-IDH2 Antibody Picoband™ (monoclonal, 6B13)

Catalog # ABO14977

Product Information

Application	WB, IHC, IF, ICC, FC
Primary Accession	P48735
Host	Mouse
Isotype	Mouse IgG2a
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-IDH2 Antibody Picoband™ (monoclonal, 6B13) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500ug/ml.

Additional Information

Gene ID	3418
Other Names	Isocitrate dehydrogenase [NADP], mitochondrial, IDH, 1.1.1.42, ICD-M, IDP, NADP(+)-specific ICDH, Oxalosuccinate decarboxylase, IDH2
Calculated MW	50909
Application Details	Western blot, 0.1-0.25 µg/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 2-5 µg/ml, Human, Rat Immunocytochemistry/Immunofluorescence, 5 µg/ml, Human Flow Cytometry, 1-3 µg/1x10 ⁶ cells, Human
Source	Eukaryota
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl and 0.2mg Na2HPO4.
Clone Names	Clone: 6B13
Immunogen	A synthetic peptide corresponding to a sequence at the C-terminus of human IDH2, identical to the related mouse and rat sequences.
Purification	Immunogen affinity purified.
Storage	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.

Protein Information

Name	IDH2
Function	Plays a role in intermediary metabolism and energy production (PubMed: 19228619 , PubMed: 22416140). It may tightly associate or interact with the pyruvate dehydrogenase complex (PubMed: 19228619 , PubMed: 22416140).
Cellular Location	Mitochondrion {ECO:0000250 UniProtKB:P33198}.

Background

Isocitrate dehydrogenase [NADP], mitochondrial is an enzyme that in humans is encoded by the IDH2 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 mitochondria. It plays a role in intermediary metabolism and energy production. This protein may tightly associate or interact with the pyruvate dehydrogenase complex. Alternative splicing results in multiple transcript variants.

Images

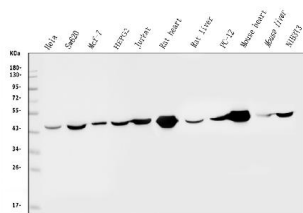


Figure 1. Western blot analysis of IDH2 using anti-IDH2 antibody (M00510-4).

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 HeLa whole cell lysates,
Lane 2: human Sw620 whole cell lysates,
Lane 3: human MCF-7 whole cell lysates,
Lane 4: human HEPG2 whole cell lysates,
Lane 5: human Jurkat whole cell lysates,
Lane 6: rat heart tissue lysates,
Lane 7: rat liver tissue lysates,
Lane 8: rat PC-12 whole cell lysates,
Lane 9: mouse heart tissue lysates,
Lane 10: mouse liver tissue lysates,
Lane 11: mouse NIH/3T3 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-IDH2 antigen affinity purified monoclonal antibody (Catalog # M00510-4) at 0.25 µg/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 IDH2 at approximately 45KD. The expected band size for IDH2 is at 45KD.

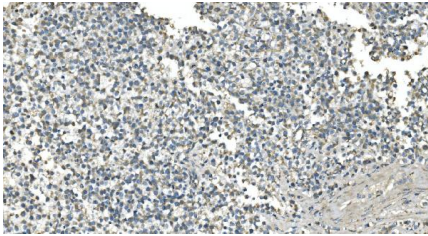


Figure 2. IHC analysis of IDH2 using anti-IDH2 antibody (M00510-4).

IDH2 was detected in paraffin-embedded section of human testicular cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-IDH2 Antibody (M00510-4) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

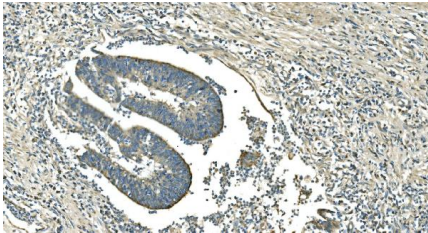


Figure 3. IHC analysis of IDH2 using anti-IDH2 antibody (M00510-4).

IDH2 was detected in paraffin-embedded section of human rectal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-IDH2 Antibody (M00510-4) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

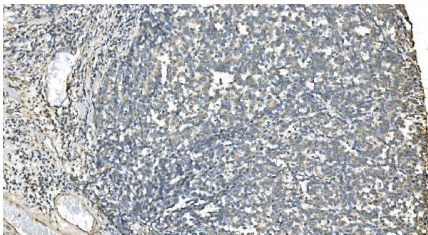


Figure 4. IHC analysis of IDH2 using anti-IDH2 antibody (M00510-4).

IDH2 was detected in paraffin-embedded section of human melanoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-IDH2 Antibody (M00510-4) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

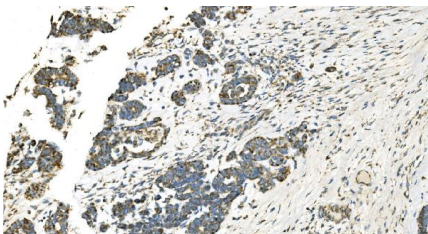
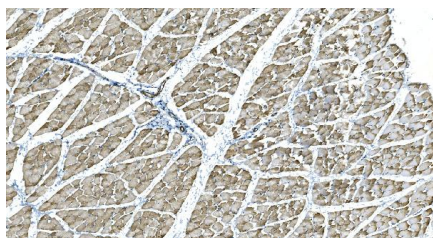


Figure 5. IHC analysis of IDH2 using anti-IDH2 antibody (M00510-4).

IDH2 was detected in paraffin-embedded section of human ovarian cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-IDH2 Antibody (M00510-4) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

Figure 6. IHC analysis of IDH2 using anti-IDH2 antibody (M00510-4).

IDH2 was detected in paraffin-embedded section of rat



skeletal muscle tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-IDH2 Antibody (M00510-4) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

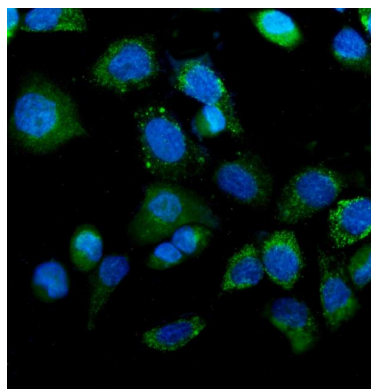


Figure 7. IF analysis of IDH2 using anti-IDH2 antibody (M00510-4). IDH2 was detected in immunocytochemical section of A431 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 µg/mL mouse anti-IDH2 Antibody (M00510-4) overnight at 4°C. DyLight®488 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.

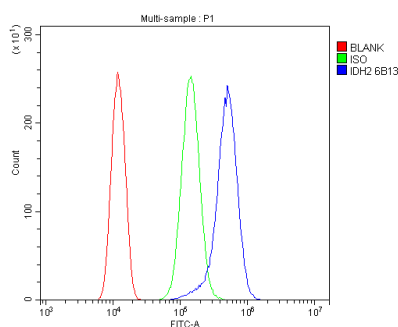


Figure 8. Flow Cytometry analysis of SiHa cells using anti-IDH2 antibody (M00510-4). Overlay histogram showing SiHa cells stained with M00510-4 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-IDH2 Antibody (M00510-4, 1 µg/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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