

## **Product Datasheet**

# Anti-IDH2 Antibody (monoclonal, 6B13) (orb692221)

Description	Anti-IDH2 Antibody (monoclonal, 6B13). Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Species/Host	Mouse
Reactivity	Human, Mouse, Rat
Conjugation	Unconjugated
Tested Applications	FC, ICC, IF, IHC, WB
Immunogen	A synthetic peptide corresponding to a sequence at the C-terminus of human IDH2, identical to the related mouse and rat sequences.
Form/Appearance	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 $\mu$ g/ml.
Storage	Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at - 20°C in small aliquots to prevent freeze-thaw cycles.
Note	For research use only
Application notes	Western blot, 0.1-0.25µg/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 2-5µg/ml, Human, Rat Immunocytochemistry/Immunofluorescence, 5µg/ml, Human Flow Cytometry (Fixed), 1-3µg/1x106 cells, Human. Add 0.2ml of distilled water will yield a concentration of 500ug/ml
lsotype	Mouse IgG2a
Clonality	Monoclonal
Clone Number	6B13

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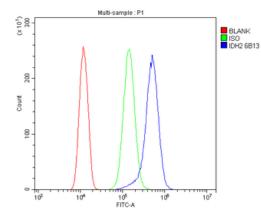
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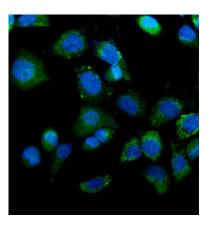
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Antibody Type	Primary Antibody
MW	45 kDa
Uniprot ID	P48735
Expiration Date	12 months from da

12 months from date of receipt.



Flow Cytometry analysis of SiHa cells using anti-IDH2 antibody. Overlay histogram showing SiHa cells (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-IDH2 Antibody (1  $\mu$ g/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (5-10  $\mu$ g/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1  $\mu$ g/1x10^6) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.



IF analysis of IDH2 using anti-IDH2 antibody. IDH2 was detected in immunocytochemical section of A431 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5  $\mu$ g/mL mouse anti-IDH2 Antibody overnight at 4°C. DyLight®488 Conjugated Goat Anti-Mouse IgG 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.

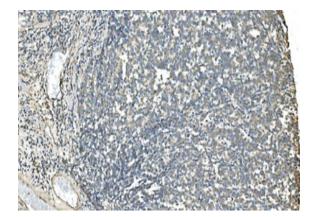
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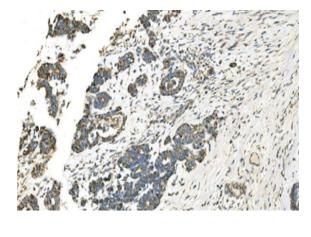
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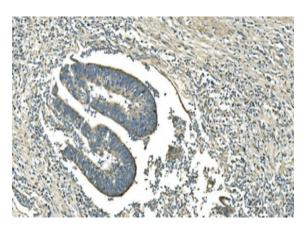
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IHC analysis of IDH2 using anti-IDH2 antibody. IDH2 was detected in paraffin-embedded section of human melanoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2  $\mu$ g/ml mouse anti-IDH2 Antibody 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 Strepavidin-Biotin-Complex (SABC) with DAB as the chromogen.



IHC analysis of IDH2 using anti-IDH2 antibody. 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 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 Strepavidin-Biotin-Complex (SABC) with DAB as the chromogen.



IHC analysis of IDH2 using anti-IDH2 antibody. 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 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 Strepavidin-Biotin-Complex (SABC) with DAB as the chromogen.

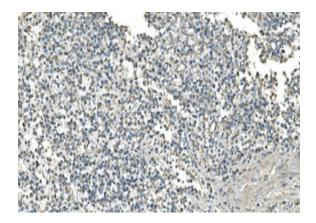
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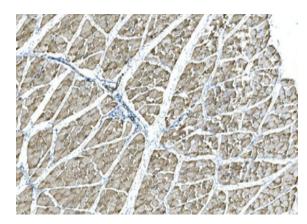
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IHC analysis of IDH2 using anti-IDH2 antibody. IDH2 was detected in paraffin-embedded section of human testicular cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2  $\mu$ g/ml mouse anti-IDH2 Antibody 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 Strepavidin-Biotin-Complex (SABC) with DAB as the chromogen.



IHC analysis of IDH2 using anti-IDH2 antibody. 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 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 Strepavidin-Biotin-Complex (SABC) with DAB as the chromogen.

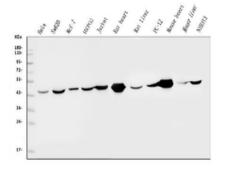
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Western blot analysis of IDH2 using anti-IDH2 antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50 ug of sample under reducing conditions. 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 150 mA 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 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 with Tanon 5200 system. A specific band was detected for IDH2 at approximately 45 KD. The expected band size for IDH2 is at 45 KD.

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