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Anti-ALDH1A1 Antibody Picoband™ (monoclonal, 4C3)

Catalog # ABO15104

Product Information

Application	WB, IHC, IF, ICC, FC
Primary Accession	P00352
Host	Mouse
Isotype	Mouse IgG2a
Reactivity	Human
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-ALDH1A1 Antibody Picoband™ (monoclonal, 4C3) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human.
Reconstitution	Adding 0.2 ml of distilled water will yield a concentration of 500 □g/ml.

Additional Information

Gene ID	216
Other Names	Aldehyde dehydrogenase 1A1, 1.2.1.19, ALDH1A1 (HGNC:402)
Calculated MW	54862
Application Details	Western blot, 0.25-0.5 □g/ml, Human Immunohistochemistry(Paraffin-embedded Section), 2-5 □g/ml, Human Immunocytochemistry/Immunofluorescence, 5 □g/ml, Human Flow Cytometry, 1-3 □g/1x10^6 cells, Human
Source	Eukaryota
Contents	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na2HPO4.
Clone Names	Clone: 4C3
Immunogen	E.coli-derived human ALDH1A1 recombinant protein (Position: T6-Q342).
Purification	Immunogen affinity purified.
Storage	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 freezing and thawing.

Protein Information

Name	ALDH1A1 (HGNC:402)

Function	Cytosolic dehydrogenase that catalyzes the NAD(+)-dependent irreversible oxidation of a wide range of aldehydes to their corresponding carboxylic acid (PubMed:25450233, PubMed:26373694, PubMed:22339434, PubMed:23444097). Functions downstream of retinol dehydrogenases and catalyzes the oxidation of retinaldehyde into retinoic acid, the second step in the oxidation of retinol/vitamin A into retinoic acid (PubMed:25413692). This pathway is crucial to control the levels of retinol and retinoic acid, two important molecules which excess can be teratogenic and cytotoxic (PubMed:25413692). Also oxidizes aldehydes resulting from lipid peroxidation like (E)-4-hydroxynon-2-enal/HNE, malonaldehyde and hexanal that form protein adducts and are highly cytotoxic. By participating for instance to the clearance of (E)-4-hydroxynon-2- enal/HNE in the lens epithelium prevents the formation of HNE-protein adducts and lens opacification (PubMed:12941160, PubMed:15623782, PubMed:19296407). Also functions downstream of fructosamine-3-kinase in the fructosamine degradation pathway by catalyzing the oxidation of 3- deoxyglucosone, the carbohydrate product of fructosamine 3-phosphate decomposition, which is itself a potent glycating agent that may react with lysine and arginine side-chains of proteins (PubMed:17175089). Also has an aminobutyraldehyde dehydrogenase activity and is probably part of an alternative pathway for the biosynthesis of GABA/4-aminobutanoate in midbrain, thereby playing a role in GABAergic synaptic transmission (By similarity).
Cellular Location	Cytoplasm, cytosol. Cell projection, axon {ECO:0000250|UniProtKB:P24549}
Tissue Location	Expressed by erythrocytes (at protein level).

Background

Aldehyde dehydrogenase 1 family, member A1, also known as ALDH1A1 or retinaldehyde dehydrogenase 1 (RALDH1), is an enzyme that in humans is encoded by the ALDH1A1 gene. It is mapped to 9q21.13. The protein encoded by this gene belongs to the aldehyde dehydrogenase family. Aldehyde dehydrogenase is the next enzyme after alcohol dehydrogenase in the major pathway of alcohol metabolism. There are two major aldehyde dehydrogenase isozymes in the liver, cytosolic and mitochondrial, which are encoded by distinct genes, and can be distinguished by their electrophoretic mobility, kinetic properties, and subcellular localization. This gene encodes the cytosolic isozyme. Studies in mice show that through its role in retinol metabolism, this gene may also be involved in the regulation of the metabolic responses to high-fat diet.

Images

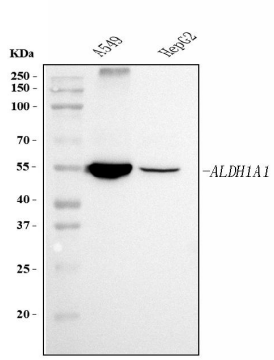


Figure 1. Western blot analysis of ALDH1A1 using anti-ALDH1A1 antibody (M01392-2).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.
Lane 1: human A549 whole cell lysates,
Lane 2: human HepG2 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-ALDH1A1 antigen affinity purified monoclonal antibody (Catalog # M01392-2) at 0.5 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 ALDH1A1 at approximately 55 kDa. The expected band size for ALDH1A1 is at 55 kDa.

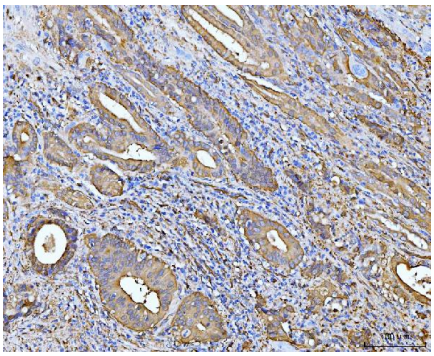


Figure 2. IHC analysis of ALDH1A1 using anti-ALDH1A1 antibody (M01392-2).
ALDH1A1 was detected in a paraffin-embedded section of human gall bladder adenosquamous carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-ALDH1A1 Antibody (M01392-2) 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) (Catalog # SA1021) with DAB as the chromogen.

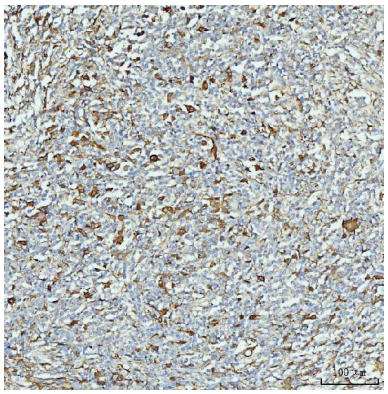


Figure 3. IHC analysis of ALDH1A1 using anti-ALDH1A1 antibody (M01392-2).
ALDH1A1 was detected in a paraffin-embedded section of human lymphadenoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-ALDH1A1 Antibody (M01392-2) 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) (Catalog # SA1021) with DAB as the chromogen.

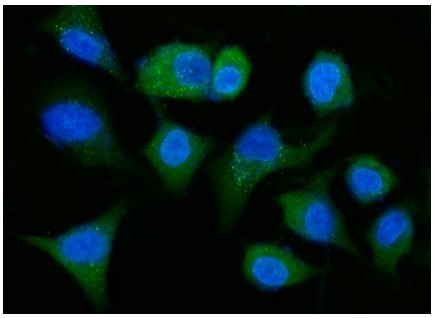


Figure 4. IF analysis of ALDH1A1 using anti-ALDH1A1 antibody (M01392-2).
ALDH1A1 was detected in an immunocytochemical section of A549 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 µg/mL mouse anti-ALDH1A1 Antibody (M01392-2) 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.

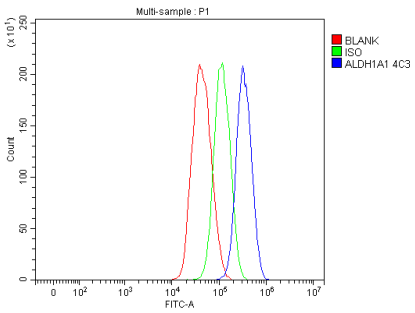


Figure 5. Flow Cytometry analysis of HepG2 cells using anti-ALDH1A1 antibody (M01392-2).
Overlay histogram showing HepG2 cells stained with M01392-2 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-ALDH1A1 Antibody (M01392-2, 1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10^6) used under the

same conditions. Unlabelled sample (Red line) was also used as a control.

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