

Anti-ASS1 Antibody Picoband™ (monoclonal, 5I5)

Catalog # ABO14984

Product Information

Application	WB, IHC, IF, ICC, FC
Primary Accession	P00966
Host	Mouse
Isotype	Mouse IgG2a
Reactivity	Rat, Human, Mouse, Monkey
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-ASS1 Antibody Picoband™ (monoclonal, 5I5) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Monkey, Mouse, Rat.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500ug/ml.

Additional Information

Gene ID	445
Other Names	Argininosuccinate synthase, 6.3.4.5, Citrulline--aspartate ligase, ASS1 (HGNC:758)
Calculated MW	46530
Application Details	Western blot, 0.25-0.5 µg/ml, Human, Mouse, Rat, Monkey Immunohistochemistry (Paraffin-embedded Section), 2-5 µg/ml, Human, Mouse, 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: 5I5
Immunogen	E.coli-derived human ASS1 recombinant protein (Position: S3-S365).
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	ASS1 (HGNC:758)
Function	One of the enzymes of the urea cycle, the metabolic pathway transforming neurotoxic ammonia produced by protein catabolism into innocuous urea in the liver of ureotelic animals. Catalyzes the formation of arginosuccinate from aspartate, citrulline and ATP and together with ASL it is responsible for the biosynthesis of arginine in most body tissues.
Cellular Location	Cytoplasm, cytosol
Tissue Location	Expressed in adult liver.

Background

Argininosuccinate synthetase is an enzyme that in humans is encoded by the ASS1 gene. It is mapped to 9q34.11. The protein encoded by this gene catalyzes the penultimate step of the arginine biosynthetic pathway. There are approximately 10 to 14 copies of this gene including the pseudogenes scattered across the human genome, among which the one located on chromosome 9 appears to be the only functional gene for argininosuccinate synthetase. Mutations in the chromosome 9 copy of this gene cause citrullinemia. Two transcript variants encoding the same protein have been found for this gene.

Images

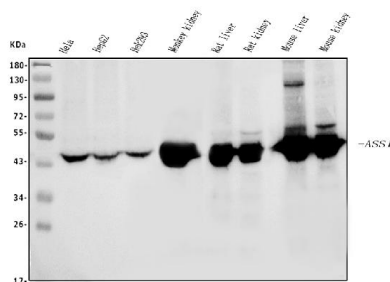


Figure 1. Western blot analysis of ASS1 using anti-ASS1 antibody (M02212-1).

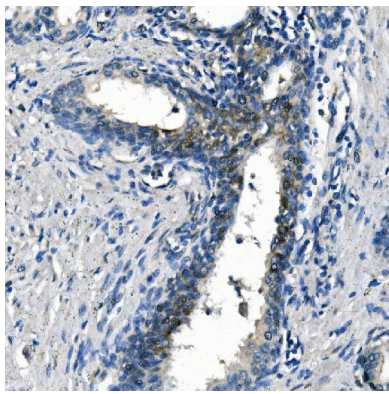
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.

Lane 1: human Hela whole cell lysates,
Lane 2: human HEPG2 whole cell lysates,
Lane 3: human Hek293 whole cell lysates,
Lane 4: monkey kidney tissue lysates,
Lane 5: rat liver tissue lysates,
Lane 6: rat kidney tissue lysates,
Lane 7: mouse liver tissue lysates,
Lane 8: mouse kidney tissue 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-ASS1 antigen affinity purified monoclonal antibody (Catalog # M02212-1) 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 ASS1 at approximately 47KD. The expected band size for ASS1 is at 47KD.

Figure 2. IHC analysis of ASS1 using anti-ASS1 antibody (M02212-1).

ASS1 was detected in paraffin-embedded section of human prostate 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-ASS1 Antibody (M02212-1) 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.

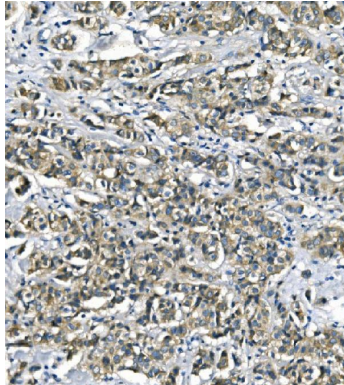


Figure 3. IHC analysis of ASS1 using anti-ASS1 antibody (M02212-1).

ASS1 was detected in paraffin-embedded section of human breast 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-ASS1 Antibody (M02212-1) 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.

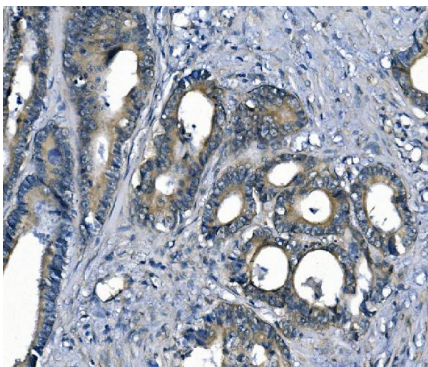


Figure 4. IHC analysis of ASS1 using anti-ASS1 antibody (M02212-1).

ASS1 was detected in paraffin-embedded section of human colon 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-ASS1 Antibody (M02212-1) 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.

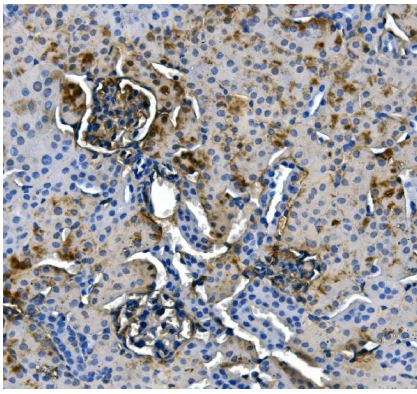


Figure 5. IHC analysis of ASS1 using anti-ASS1 antibody (M02212-1).

ASS1 was detected in paraffin-embedded section of rat liver 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-ASS1 Antibody (M02212-1) 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 ASS1 using anti-ASS1 antibody (M02212-1).

ASS1 was detected in paraffin-embedded section of mouse kidney 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 $\mu\text{g/ml}$ mouse anti-ASS1 Antibody (M02212-1) 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.

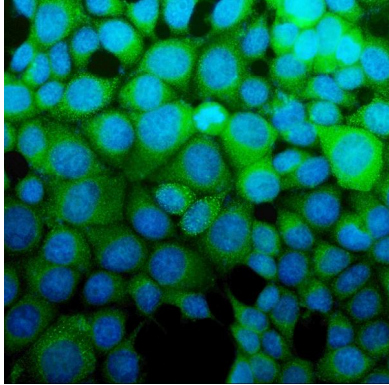


Figure 7. IF analysis of ASS1 using anti-ASS1 antibody (M02212-1). ASS1 was detected in immunocytochemical section of MCF-7 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 $\mu\text{g/mL}$ mouse anti-ASS1 Antibody (M02212-1) 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.

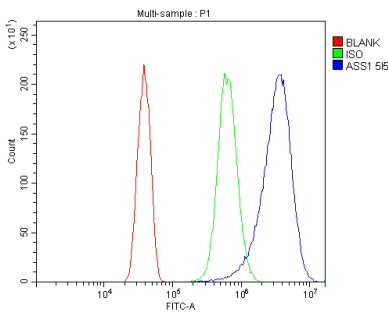


Figure 8. Flow Cytometry analysis of SiHa cells using anti-ASS1 antibody (M02212-1). Overlay histogram showing SiHa cells stained with M02212-1 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-ASS1 Antibody (M02212-1, 1 $\mu\text{g}/1 \times 10^6$ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 $\mu\text{g}/1 \times 10^6$ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 $\mu\text{g}/1 \times 10^6$) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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