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Anti-Glutaminase/GLS Antibody Picoband™ (monoclonal, 3G13)

Catalog # ABO15106

Product Information

Application	WB, IHC, IF, ICC, FC
Primary Accession	O94925
Host	Mouse
Isotype	Mouse IgG2a
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-Glutaminase/GLS Antibody Picoband™ (monoclonal, 3G13) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Reconstitution	Adding 0.2 ml of distilled water will yield a concentration of 500 □g/ml.

Additional Information

Gene ID	2744
Other Names	Glutaminase kidney isoform, mitochondrial, GLS, 3.5.1.2, K-glutaminase, L-glutamine amidohydrolase, Glutaminase kidney isoform, mitochondrial 68 kDa chain, GLS, GLS1, KIAA0838
Calculated MW	73461
Application Details	Western blot, 0.25-0.5 □g/ml, Human, Mouse, Rat 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: 3G13
Immunogen	E.coli-derived human Glutaminase/GLS recombinant protein (Position: K396-N654).
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	GLS
Synonyms	GLS1, KIAA0838
Function	Catalyzes the first reaction in the primary pathway for the renal catabolism of glutamine. Plays a role in maintaining acid-base homeostasis. Regulates the levels of the neurotransmitter glutamate, the main excitatory neurotransmitter in the brain (PubMed:30239721, PubMed:30575854, PubMed:30970188).
Cellular Location	[Isoform 1]: Mitochondrion {ECO:0000250|UniProtKB:P13264}. Cytoplasm, cytosol. Note=The 74-kDa cytosolic precursor is translocated into the mitochondria and processed via a 72-kDa intermediate to yield the mature 68- and 65-kDa subunits {ECO:0000250|UniProtKB:P13264} [Glutaminase kidney isoform, mitochondrial 68 kDa chain]: Mitochondrion matrix {ECO:0000250|UniProtKB:P13264} Note=Produced by the proteolytic processing of the 74-kDa cytosolic precursor. {ECO:0000250|UniProtKB:P13264}
Tissue Location	Isoform 1 and isoform 3 are detected in brain cortex. Isoform 3 is highly expressed in astrocytoma, ganglioglioma and ependymoma. Isoform 1 is highly expressed in brain and kidney, but not detected in liver. Isoform 3 is highly expressed in heart and pancreas, detected at lower levels in placenta, lung, pancreas and kidney, but is not detected in liver. Isoform 2 is expressed in cardiac and skeletal muscle.

Background

This gene encodes the K-type mitochondrial glutaminase. The encoded protein is an phosphate-activated amidohydrolase that catalyzes the hydrolysis of glutamine to glutamate and ammonia. This protein is primarily expressed in the brain and kidney plays an essential role in generating energy for metabolism, synthesizing the brain neurotransmitter glutamate and maintaining acid-base balance in the kidney. Alternate splicing results in multiple transcript variants.

Images

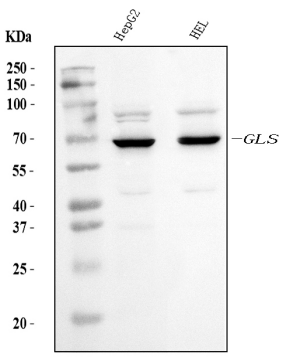

Figure 1. Western blot analysis of Glutaminase/GLS using anti-Glutaminase/GLS antibody (M01272-3). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.
Lane 1: human HepG2 whole cell lysates,
Lane 2: human HEL 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-Glutaminase/GLS antigen affinity purified monoclonal antibody (Catalog # M01272-3) 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 Glutaminase/GLS at approximately 56-73 kDa. The expected band size for Glutaminase/GLS is at 73 kDa.

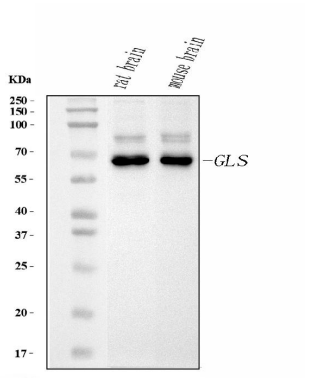

Figure 2. Western blot analysis of Glutaminase/GLS using anti-Glutaminase/GLS antibody (M01272-3). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.
Lane 1: rat brain tissue lysates,
Lane 2: mouse brain tissue 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-Glutaminase/GLS antigen affinity purified monoclonal antibody (Catalog # M01272-3) 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 Glutaminase/GLS at approximately 56-73 kDa. The expected band size for Glutaminase/GLS is at 73 kDa.

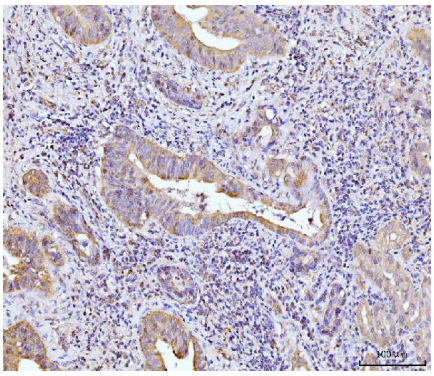

Figure 3. IHC analysis of Glutaminase/GLS using anti-Glutaminase/GLS antibody (M01272-3). Glutaminase/GLS was detected in a paraffin-embedded section of human gall bladder adenosquamous carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-Glutaminase/GLS Antibody (M01272-3) 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.

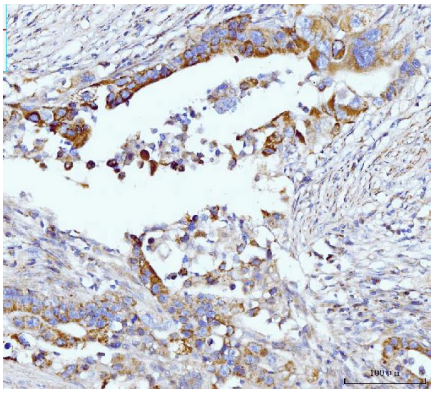

Figure 4. IHC analysis of Glutaminase/GLS using anti-Glutaminase/GLS antibody (M01272-3). Glutaminase/GLS was detected in a paraffin-embedded section of human appendiceal adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-Glutaminase/GLS Antibody (M01272-3) 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.

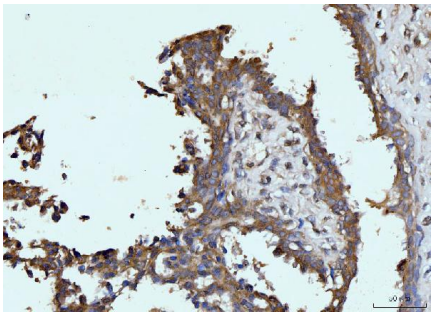

Figure 5. IHC analysis of Glutaminase/GLS using anti-Glutaminase/GLS antibody (M01272-3). Glutaminase/GLS was detected in a paraffin-embedded section of human breast cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-Glutaminase/GLS Antibody (M01272-3) 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.

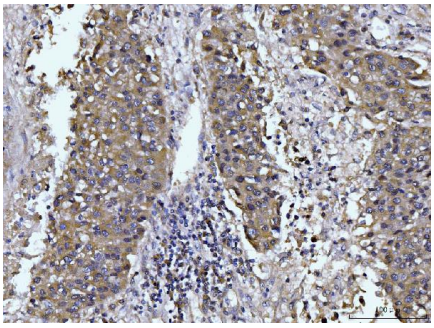

Figure 6. IHC analysis of Glutaminase/GLS using anti-Glutaminase/GLS antibody (M01272-3). Glutaminase/GLS was detected in a paraffin-embedded section of human liver cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-Glutaminase/GLS Antibody (M01272-3) 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.

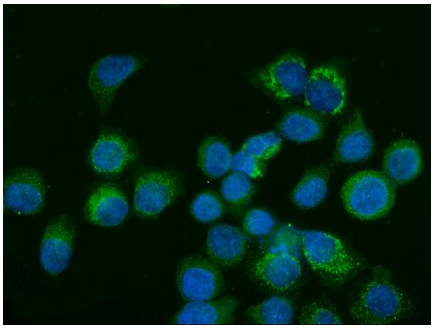

Figure 7. IF analysis of Glutaminase/GLS using anti-Glutaminase/GLS antibody (M01272-3). Glutaminase/GLS was detected in an immunocytochemical section of SiHa cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 µg/mL mouse anti-Glutaminase/GLS Antibody (M01272-3) 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.

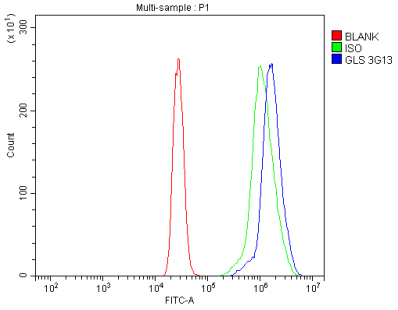

Figure 8. Flow Cytometry analysis of U937 cells using anti-Glutaminase/GLS antibody (M01272-3). Overlay histogram showing U937 cells stained with M01272-3 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-Glutaminase/GLS Antibody (M01272-3, 1 µg/1x10^6 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.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.