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antibody and ELISA experts

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Anti-HGD Antibody Picoband™

Catalog Number: A01909-1

About HGD

The HGD gene encodes homogentisate 1,2-dioxygenase (HGD), an enzyme involved in the catabolism of phenylalanine and tyrosine. This enzyme is involved in the catabolism of the amino acids tyrosine and phenylalanine. Mutations in this gene are the cause of the autosomal recessive metabolism disorder alkaptonuria. This gene is mapped to chromosome 3q21-q23 by a preliminary PCR screen of hamster/human somatic cell hybrid genomic DNA samples and by fluorescence in situ hybridization.

Overview

Product Name	Anti-HGD Antibody Picoband™
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-HGD Antibody Picoband™ catalog # A01909-1. Tested in ELISA, Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	ELISA, Flow Cytometry, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na_2HPO_4, 0.05mg NaN_3.
Storage Instructions	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.
Host	Rabbit
Uniprot ID	Q93099

Technical Details

Immunogen	E. coli-derived human HGD recombinant protein (Position: D374-N445).
Predicted Reactive Species	Human
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P) and ICC.
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Rabbit IgG
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.

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Suggested Dilutions	Dilute the sample so that the expected range of concentrations fall within the detection range of this kit. If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples. Some PubMed article(s) citing the expression level of this target are as follows: Boster Bio's internal QC testing used: Western blot,0.1-0.5ug/ml Immunohistochemistry (Paraffin-embedded Section),0.5-1ug/ml Immunocytochemistry/Immunofluorescence, 5ug/ml Flow Cytometry, 1-3ug/1x10^6 cells Direct ELISA,0.1-0.5ug/ml

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Anti-HGD Antibody Picoband™ (A01909-1) Images

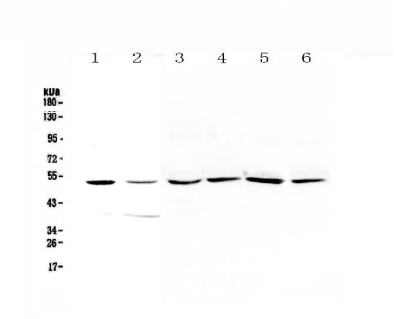

Figure 1. Western blot analysis of HGD using anti-HGD antibody (A01909-1).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: human HepG2 whole cell lysates,
Lane 2: human A549 whole cell lysates,
Lane 3: rat liver tissue lysates,
Lane 4: rat kidney tissue lysates,
Lane 5: mouse liver tissue lysates,
Lane 6: 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 rabbit anti-HGD antigen affinity purified polyclonal antibody (Catalog # A01909-1) at 0.5 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit 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 # EK1002) with Tanon 5200 system. A specific band was detected for HGD at approximately 50KD. The expected band size for HGD is at 50KD.

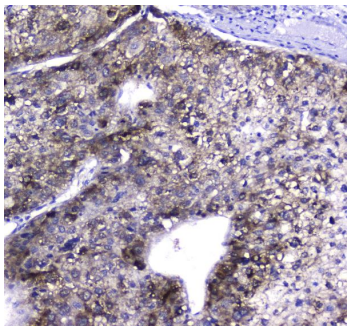

Figure 2. IHC analysis of HGD using anti-HGD antibody (A01909-1).
HGD was detected in paraffin-embedded section of human liver cancer tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-HGD Antibody (A01909-1) overnight at 4°C. Biotinylated goat anti-rabbit 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 # SA1022) with DAB as the chromogen.

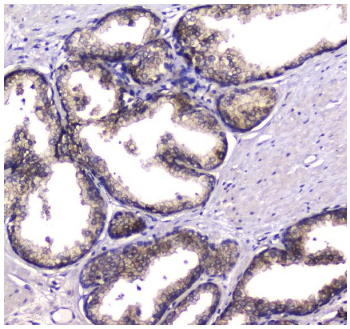

Figure 3. IHC analysis of HGD using anti-HGD antibody (A01909-1).
HGD was detected in paraffin-embedded section of human prostatic cancer tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-HGD Antibody (A01909-1) overnight at 4°C. Biotinylated goat anti-rabbit 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 # SA1022) with DAB as the chromogen.

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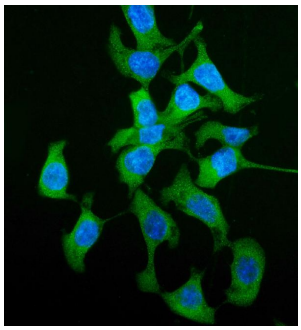

Figure 4. IF analysis of HGD using anti-HGD antibody (A01909-1).
HGD was detected in immunocytochemical section of CACO-2 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5ug/mL rabbit anti-HGD Antibody (A01909-1) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) 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.

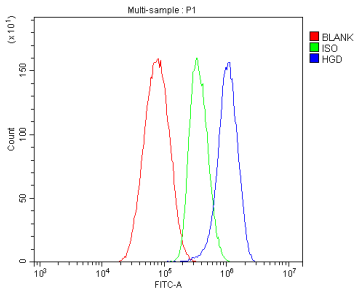

Figure 5. Flow Cytometry analysis of HEPG2 cells using anti-HGD antibody (A01909-1).
Overlay histogram showing HEPG2 cells stained with A01909-1 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-HGD Antibody (A01909-1, 1ug/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1ug/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.