BOSTER BIOLOGICAL TECHNOLOGY
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Anti-DBT Antibody Picoband®

Catalog Number: A01145-2

About DBT

The branched-chain alpha-keto acid dehydrogenase complex (BCKD) is an inner-mitochondrial enzyme complex involved in the breakdown of the branched-chain amino acids isoleucine, leucine, and valine. The BCKD complex is thought to be composed of a core of 24 transacylase (E2) subunits, and associated decarboxylase (E1), dehydrogenase (E3), and regulatory subunits. This gene encodes the transacylase (E2) subunit. Mutations in this gene result in maple syrup urine disease, type 2. Alternatively spliced transcript variants have been described, but their biological validity has not been determined.

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Overview

Product Name	Anti-DBT Antibody Picoband®
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-DBT Antibody Picoband® catalog # A01145-2. Tested in WB, IHC, ICC/IF, IP, Flow Cytometry, ELISA applications. This antibody reacts with Human, Mouse, Rat. The brand Picoband indicates this is a premium antibody that guarantees superior quality, high affinity, and strong signals with minimal background in Western blot applications. Only our best-performing antibodies are designated as Picoband, ensuring unmatched performance.
Application	ELISA, Flow Cytometry, IP, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4 mg Trehalose, 0.9 mg NaCl, 0.2 mg Na2HPO4.
Storage Instructions	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.
Host	Rabbit
Uniprot ID	P11182

Technical Details

Immunogen	E.coli-derived human DBT recombinant protein (Position: K46-R462). Human DBT shares 89.7% amino acid (aa) sequence identity with mouse DBT.

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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.
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	Western blot, 0.25-0.5 ug/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 2-5 ug/ml, Human Immunocytochemistry/Immunofluorescence, 5 ug/ml, Human Immunoprecipitation, 2-4 ug/ml, Human Flow Cytometry (Fixed), 1-3 ug/1x10^6 cells, Human ELISA, 0.1-0.5 ug/ml Note: For flow cytometry (FCM), suitable for fixed cells only. Not suitable for FACS or surface flow cytometry using live cells.

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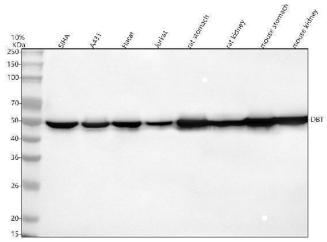
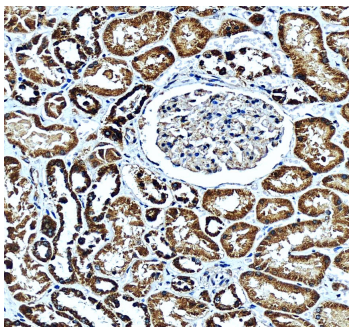
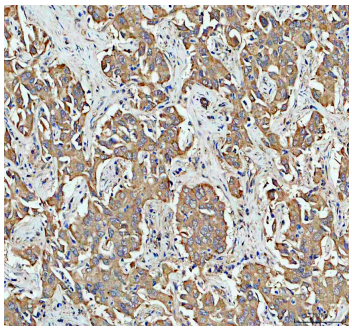
Anti-DBT Antibody Picoband® (A01145-2) Images



Western blot analysis of DBT using anti-DBT antibody (A01145-2). 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 SiHa whole cell lysates, Lane 2: human A431 whole cell lysates, Lane 3: human Hacat whole cell lysates, Lane 4: human Jurkat whole cell lysates, Lane 5: rat stomach tissue lysates, Lane 6: rat kidney tissue lysates, Lane 7: mouse stomach tissue lysates, Lane 8: mouse kidney 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 rabbit anti-DBT antigen affinity purified polyclonal antibody (A01145-2) 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 1:5000 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 DBT at approximately 53 kDa. The expected band size for DBT is at 53 kDa.



IHC analysis of DBT using anti-DBT antibody (A01145-2). DBT was detected in a paraffin-embedded section of human kidney 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 ug/ml rabbit anti-DBT Antibody (A01145-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



IHC analysis of DBT using anti-DBT antibody (A01145-2). DBT 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 ug/ml rabbit anti-DBT Antibody (A01145-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

IF analysis of DBT using anti-DBT antibody (A01145-2) and

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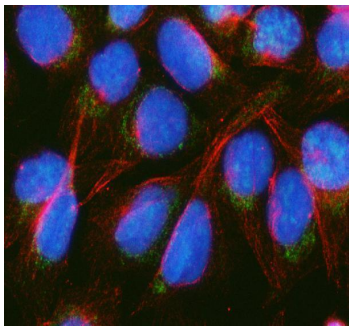
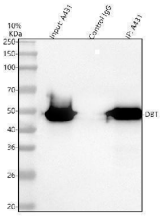
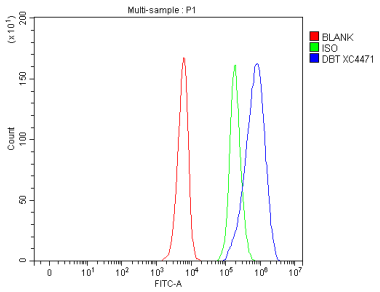


anti-Beta Tubulin antibody (M01857-3). DBT was detected in an immunocytochemical section of U2OS cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 ug/mL rabbit anti-DBT Antibody (A01145-2) and mouse anti-Beta Tubulin antibody (M01857-3) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) and DyLight®550 Conjugated Goat Anti-Mouse IgG (BA1133) were used as secondary antibody at 1:500 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.



Immunoprecipitating DBT in A431 whole cell lysate.Western blot analysis of DBT using anti-DBT antibody (A01145-2).Lane 1: A431 whole cell lysates (30ug),Lane 2: Rabbit control IgG instead of anti-DBT antibody in A431 whole cell lysate,Lane 3: anti-DBT antibody (2ug) + A431 whole cell lysate (500ug).After electrophoresis, proteins were transferred to a membrane. Then the membrane was incubated with rabbit anti-DBT antigen affinity purified polyclonal antibody (A01145-2) at a dilution of 0.5 ug/mL and probed with a mouse anti-rabbit IgG-HRP secondary antibody (Light Chain). The signal is developed using ECL Plus Western Blotting Substrate (Catalog # AR1196-200). A specific band was detected for DBT at approximately 53 kDa. The expected band size for DBT is at 53 kDa.



Flow Cytometry analysis of SiHa cells using anti-DBT antibody (A01145-2). Overlay histogram showing SiHa cells stained with A01145-2 (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 rabbit anti-DBT Antibody (A01145-2, 1 ug/1x106 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10 ug/1x106 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 ug/1x106) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

Anti-DBT Antibody

For Research Use Only. Not for use in diagnostic procedures.