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antibody and ELISA experts

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Anti-UBE2D1/2/3/4 Antibody Picoband®

Catalog Number: A04728-1

About UBE2D1/2/3/4

The modification of proteins with ubiquitin is an important cellular mechanism for targeting abnormal or short-lived proteins for degradation. Ubiquitination involves at least three classes of enzymes: ubiquitin-activating enzymes, or E1s, ubiquitin-conjugating enzymes, or E2s, and ubiquitin-protein ligases, or E3s. This gene encodes a member of the E2 ubiquitin-conjugating enzyme family. This enzyme is closely related to a stimulator of iron transport (SFT), and is up-regulated in hereditary hemochromatosis. It also functions in the ubiquitination of the tumor-suppressor protein p53 and the hypoxia-inducible transcription factor HIF1alpha by interacting with the E1 ubiquitin-activating enzyme and the E3 ubiquitin-protein ligases. Two transcript variants encoding different isoforms have been found for this gene.

Overview

Product Name	Anti-UBE2D1/2/3/4 Antibody Picoband®
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-UBE2D1/2/3/4 Antibody Picoband® catalog # A04728-1. Tested in ELISA, Flow Cytometry, IF, IHC, ICC, WB 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, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.005mg NaN3.
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	P51668

Technical Details

Immunogen	E.coli-derived human UBE2D1/2/3/4 recombinant protein (Position: D116-M147).
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

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Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	Western blot, 0.25-0.5ug/ml, Human, Rat Immunohistochemistry (Paraffin-embedded Section), 2-5ug/ml, Human, Mouse, Rat Immunocytochemistry/Immunofluorescence, 5ug/ml, Human Flow Cytometry (Fixed), 1-3ug/1x10^6 cells, Human, Mouse, Rat ELISA, 0.1-0.5ug/ml, -

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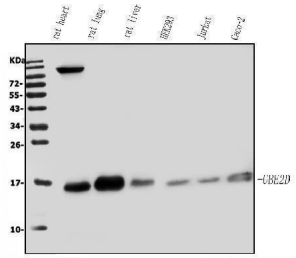
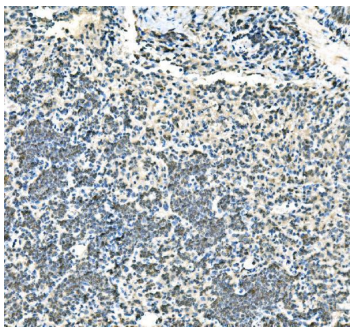
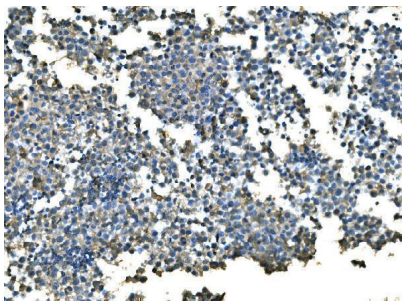
Anti-UBE2D1/2/3/4 Antibody Picoband® (A04728-1) Images



Western blot analysis of UBE2D1/2/3/4 using anti-UBE2D1/2/3/4 antibody (A04728-1). 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: rat heart tissue lysates, Lane 2: rat lung tissue lysates, Lane 3: rat liver tissue lysates, Lane 4: human HEK293 whole cell lysates, Lane 5: human Jurkat whole cell lysates, Lane 6: human CACO-2 whole cell 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-UBE2D1/2/3/4 antigen affinity purified polyclonal antibody (Catalog # A04728-1) 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 1:5000 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 UBE2D1/2/3/4 at approximately 17KD. The expected band size for UBE2D1/2/3/4 is at 17KD.



IHC analysis of UBE2D1/2/3/4 using anti-UBE2D1/2/3/4 antibody (A04728-1). UBE2D1/2/3/4 was detected in paraffin-embedded section of mouse lung lymph node 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 2ug/ml rabbit anti-UBE2D1/2/3/4 Antibody (A04728-1) 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.



IHC analysis of UBE2D1/2/3/4 using anti-UBE2D1/2/3/4 antibody (A04728-1). UBE2D1/2/3/4 was detected in paraffin-embedded section of human testicular 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 2ug/ml rabbit anti-UBE2D1/2/3/4 Antibody (A04728-1) 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.

IHC analysis of UBE2D1/2/3/4 using anti-UBE2D1/2/3/4 antibody (A04728-1). UBE2D1/2/3/4 was detected in paraffin-embedded section of human bladder cancer tissue. Heat

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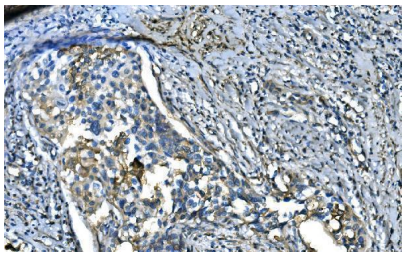
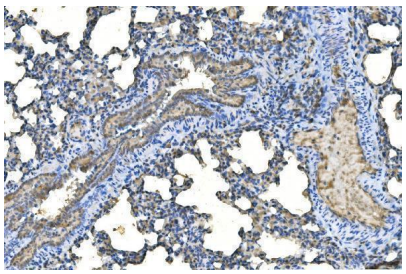
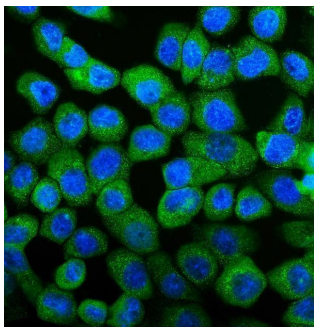


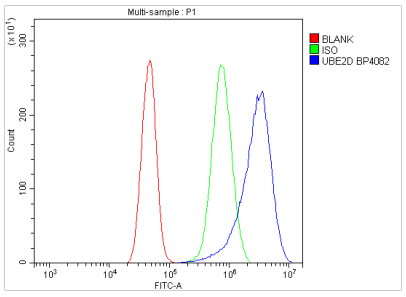
mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-UBE2D1/2/3/4 Antibody (A04728-1) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used 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.



IHC analysis of UBE2D1/2/3/4 using anti-UBE2D1/2/3/4 antibody (A04728-1). UBE2D1/2/3/4 was detected in paraffin-embedded section of rat lung 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 2ug/ml rabbit anti-UBE2D1/2/3/4 Antibody (A04728-1) 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.



IF analysis of UBE2D1/2/3/4 using anti-UBE2D1/2/3/4 antibody (A04728-1). UBE2D1/2/3/4 was detected in immunocytochemical section of A431 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-UBE2D1/2/3/4 Antibody (A04728-1) 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.



Flow Cytometry analysis of A431 cells using anti-UBE2D1/2/3/4 antibody (A04728-1). Overlay histogram showing A431 cells stained with A04728-1 (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-UBE2D1/2/3/4 Antibody (A04728-1, 1ug/1x106 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x106 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1ug/1x106) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

Flow Cytometry analysis of RAW264.7 cells using anti-UBE2D1/2/3/4 antibody (A04728-1). Overlay histogram showing RAW264.7 cells stained with A04728-1 (Blue line). To facilitate intracellular staining, cells were fixed with 4%

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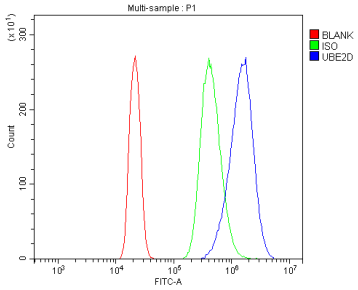
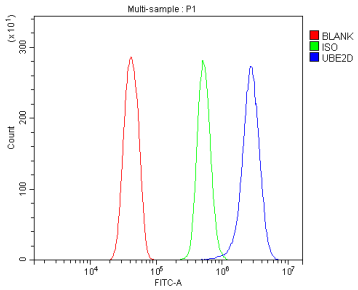


paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-UBE2D1/2/3/4 Antibody (A04728-1, 1 ug/1x106 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10 ug/1x106 cells) was 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.



Flow Cytometry analysis of RH35 cells using anti-UBE2D1/2/3/4 antibody (A04728-1). Overlay histogram showing RH35 cells stained with A04728-1 (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-UBE2D1/2/3/4 Antibody (A04728-1, 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-UBE2D1/2/3/4 Antibody

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