

Anti-CBL Antibody Picoband™

Catalog Number: A00152-2

About CBL

CBL(Cbl proto-oncogene) is also known as C-CBL, RNF55, CBL2 and E3 ubiquitin protein ligase. CBL is mapped to chromosome 11q23.3-qter by molecular characterization of the breakpoints in 2 somatic cell hybrids. The encoded protein is one of the enzymes required for targeting substrates for degradation by the proteasome. This protein mediates the transfer of ubiquitin from ubiquitin conjugating enzymes(E2) to specific substrates. This protein also contains an N-terminal phosphotyrosine binding domain that allows it to interact with numerous tyrosine-phosphorylated substrates and target them for proteasome degradation. As such it functions as a negative regulator of many signal transduction pathways. This gene has been found to be mutated or translocated in many cancers including acute myeloid leukaemia. Mutations in this gene are also the cause of Noonan syndrome-like disorder.

Overview

Product Name	Anti-CBL Antibody Picoband™
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-CBL Antibody Picoband™ catalog # A00152-2. Tested in ELISA, Flow Cytometry, IHC, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	ELISA, Flow Cytometry, IHC, 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	P22681

Technical Details

Immunogen	E.coli-derived human CBL recombinant protein (Position: M470-E766).
Predicted Reactive Species	Chicken
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).
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.
Suggested Dilutions	<p>Dilute the sample so that the expected range of concentrations fall within the detection range of this kit.</p> <p>If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples.</p> <p>Some PubMed article(s) citing the expression level of this target are as follows:</p> <p>Boster Bio's internal QC testing used:</p> <p>Western blot, 0.25-0.5 ug/ml, Human, Mouse, Rat</p> <p>Immunohistochemistry(Paraffin-embedded Section), 2-5 ug/ml, Human</p> <p>Flow Cytometry, 1-3 ug/1x10⁶ cells, Human</p> <p>Direct ELISA, 0.1-0.5 ug/ml, Human</p>

Anti-CBL Antibody Picoband™ (A00152-2) Images

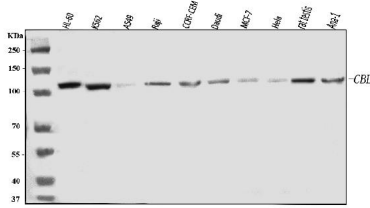


Figure 1. Western blot analysis of CBL using anti-CBL antibody (A00152-2).

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 HL-60 whole cell lysates,
Lane 2: human K562 whole cell lysates,
Lane 3: human A549 whole cell lysates,
Lane 4: human Raji whole cell lysates,
Lane 5: human CCRF-CEM whole cell lysates,
Lane 6: human Daudi whole cell lysates,
Lane 7: human MCF-7 whole cell lysates,
Lane 8: human Hela whole cell lysates,
Lane 9: rat testis tissue lysates,
Lane 10: mouse Ana-1 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 rabbit anti-CBL antigen affinity purified polyclonal antibody (Catalog # A00152-2) 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 CBL at approximately 120 kDa. The expected band size for CBL is at 120 kDa.

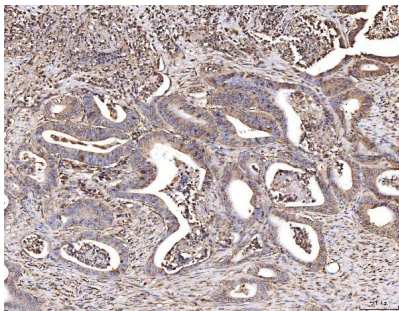
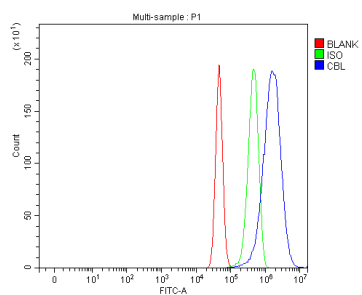


Figure 2. IHC analysis of CBL using anti-CBL antibody (A00152-2).

CBL was detected in a paraffin-embedded section of human colorectal 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-CBL Antibody (A00152-2) 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.

Figure 3. Flow Cytometry analysis of U87 cells using anti-CBL antibody (A00152-2).

Overlay histogram showing U87 cells stained with A00152-2 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-CBL Antibody (A00152-2, 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 ($1 \mu\text{g}/1 \times 10^6$) used under the same conditions.
Unlabelled sample (Red line) was also used as a control.

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