

Anti-ROC1/RBX1 Antibody Picoband™

Catalog Number: PB9798

About RBX1

RING-box protein 1, also known as ROC1, is a protein that in humans is encoded by the RBX1 gene. This gene is mapped to chromosome 22q13.2 based on an alignment of the RBX1 sequence with the genomic sequence. ROC1 is recruited by cullin-1 to form a quaternary SCF (HOS)-ROC1 holoenzyme (with SKP1 and the BTRCP homolog HOS). SCF (HOS)-ROC1 binds IKK-beta-phosphorylated I-kappa-B-alpha and catalyzes its ubiquitination in the presence of ubiquitin, E1, and CDC34. Conclusively, ROC1 plays a unique role in the ubiquitination reaction by heterodimerizing with cullin-1 to catalyze ubiquitin polymerization.

Overview

Product Name	Anti-ROC1/RBX1 Antibody Picoband™
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-ROC1/RBX1 Antibody Picoband™ catalog # PB9798. Tested in Flow Cytometry, IF, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	Flow Cytometry, IF, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 5mg BSA, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg 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	P62877

Technical Details

Immunogen	A synthetic peptide corresponding to a sequence at the C-terminus of human ROC1, identical to the related mouse sequence.
Predicted Reactive Species	Hamster
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot.
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, 0.1-0.5ug/ml, Mouse, Rat</p> <p>Immunocytochemistry/Immunofluorescence, 2ug/ml, Human</p> <p>Flow Cytometry, 1-3ug/1x10⁶ cells, Human</p>

Anti-ROC1/RBX1 Antibody Picoband™ (PB9798) Images



Figure 1. Western blot analysis of ROC1 using anti-ROC1 antibody (PB9798).

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 50 ug of sample under reducing conditions.

Lane 1: Rat Testis Tissue Lysate,

Lane 2: Rat Brain Tissue Lysate,

Lane 3: Mouse Brain Tissue Lysate,

Lane 4: Mouse Spleen Tissue Lysate.

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-ROC1 antigen affinity purified polyclonal antibody (Catalog # PB9798) 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 ROC1 at approximately 15 kDa. The expected band size for ROC1 is at 15 kDa.

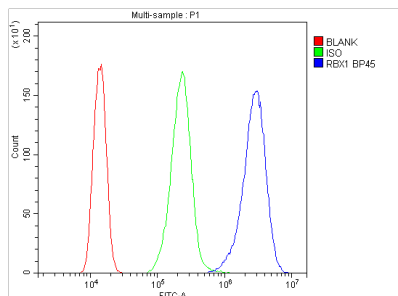


Figure 2. Flow Cytometry analysis of A431 cells using anti-ROC1 antibody (PB9798).

Overlay histogram showing A431 cells stained with PB9798 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-ROC1 Antibody (PB9798, 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 (Red line) was also used as a control.

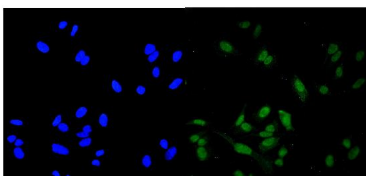


Figure 3. IF analysis of ROC1 using anti-ROC1 antibody (PB9798).

ROC1 was detected in immunocytochemical section of HeLa 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 2ug/mL rabbit anti-ROC1 Antibody (PB9798) 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.

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