BOSTER
antibody and ELISA experts

BOSTER BIOLOGICAL TECHNOLOGY
3942 B Valley Ave, Pleasanton, CA 94566
888-466-3604 | support@bosterbio.com | www.bosterbio.com

Anti-KEAP1 Antibody Picoband®

Catalog Number: A00514-3

About KEAP1

KEAP1 (KELCH-LIKE ECH-ASSOCIATED PROTEIN 1), is a protein that in humans is encoded by the Keap1 gene. The KIAA0132 gene is mapped on 19p13.2. Keap1 contains a central BTB/POZ domain and a C-terminal double glycine repeat (DGR), or Kelch, module. Keap1 has been shown to interact with Nrf2, a master regulator of the antioxidant response, which is important for the amelioration of oxidative stress. In the presence of the electrophilic agent diethylmalate, Nrf2 activity is released from Keap1 and Nrf2 translocate to the nucleus. Under quiescent conditions, Nrf2 is anchored in the cytoplasm through binding to Keap1, which, in turn, facilitates the ubiquitination and subsequent proteolysis of Nrf2. Because Nrf2 activation leads to a coordinated antioxidant and anti-inflammatory response, and Keap1 represses Nrf2 activation, Keap1 has become a very attractive drug target.

Overview

Product Name	Anti-KEAP1 Antibody Picoband®
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-KEAP1 Antibody Picoband® catalog # A00514-3. Tested in ELISA, Flow Cytometry, IF, 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, 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	Q14145

Technical Details

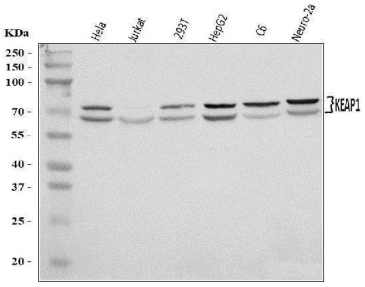
Immunogen	E.coli-derived human KEAP1 recombinant protein (Position: K84-K312).
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for ICC.
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Rabbit IgG

BOSTER BIOLOGICAL TECHNOLOGY
3942 B Valley Ave, Pleasanton, CA 94566
888-466-3604 | support@bosterbio.com | www.bosterbio.com

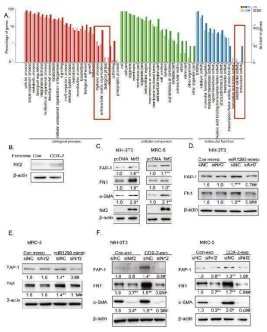
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	Western blot, 0.25-0.5 ug/ml, Human, Mouse, Rat Immunocytochemistry/Immunofluorescence, 5 ug/ml, Human Flow Cytometry (Fixed), 1-3 ug/1x10^6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

BOSTER BIOLOGICAL TECHNOLOGY
3942 B Valley Ave, Pleasanton, CA 94566
888-466-3604 | support@bosterbio.com | www.bosterbio.com

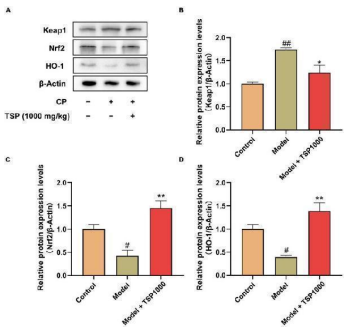
Anti-KEAP1 Antibody Picoband® (A00514-3) Images



Western blot analysis of KEAP1 using anti-KEAP1 antibody (A00514-3). 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 Hela whole cell lysates, Lane 2: human Jurkat whole cell lysates, Lane 3: human 293T whole cell lysates, Lane 4: human HepG2 whole cell lysates, Lane 5: rat C6 whole cell lysates, Lane 6: mouse Neuro-2a 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-KEAP1 antigen affinity purified polyclonal antibody (Catalog # A00514-3) 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 KEAP1 at approximately 66-72 kDa. The expected band size for KEAP1 is at 70 kDa.

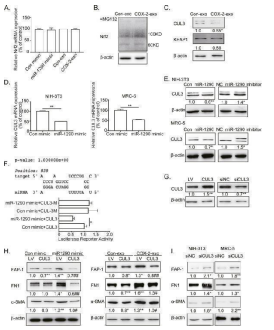


Exo-miR-1290 promoted CAFs activation by upregulating Nrf2 expression in fibroblasts. A GO enrichment analysis showed that the antioxidant activities were involved in the COX-2 exosomes-mediated CAFs activation. B Western blotting assays of NIH-3T3 cells detected with anti-Nrf2 antibody after treatments with exosomes from A549-Con or A549-COX-2 cells. C Western blotting assays of fibroblasts detected with anti-alpha-SMA, FN1, and FAP-1 antibodies after treatments with Nrf2 overexpression. D , E Western blotting assays of fibroblasts detected with anti-FN1, and FAP-1 antibodies after treatments with siNrf2 and/or miR-1290 mimic transfection. (D. NIH-3T3, E. MRC-5). F Western blotting assays of fibroblasts detected with anti-FN1, alpha-SMA, and FAP-1 antibodies after treatments with siNrf2 and exosomes. beta-actin was used as an internal reference. Data were presented as the mean ± SEM from three independent experiments. * P

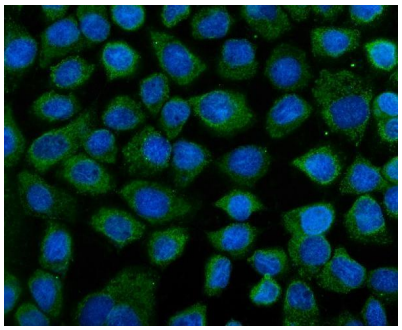


The effects of TSP on the Nrf2/HO-1 signaling pathway. (A) The representative images of western blotting results. (B–D) Quantification of the protein expression of Keap1, Nrf2, and HO-1. All data were expressed as mean ± SEM (n = 3). # P < 0.05, ## P < 0.01 vs. Control group; * P < 0.05, ** P < 0.01 vs. Model group.Index in PubMed under a CC BY license. PMID: 35719151

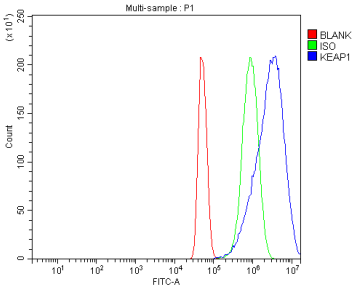
BOSTER BIOLOGICAL TECHNOLOGY
3942 B Valley Ave, Pleasanton, CA 94566
888-466-3604 | support@bosterbio.com | www.bosterbio.com



CUL3 was involved in exo-miR-1290-mediated Nrf2 upregulation and CAFs activation. A qRT-PCR assays of Nrf2 mRNA expression in NIH-3T3 cells treated with miR-1290 mimic or A549-COX-2 exosomes. B Western blotting assays of NIH-3T3 cells were detected with anti-Nrf2 antibody after treatments with MG132 and exosomes from A549-Con or A549-COX-2 cells. C Western blotting assays of NIH-3T3 cells detected with anti-CUL3 and KEAP1 antibodies after treatments with A549-con or A549-COX-2 exosomes. E Western blotting assays of fibroblasts detected with anti-CUL3 antibody after treatments with miR-1290 mimic or inhibitor. F The miR-1290 binding site of 3'UTR of CUL3 mRNA predicted by RNAhybrid 2.2 and miRWalk. Firefly luciferase reporter was used to analyze the activity of the miR-1290 binding site of the reporter with the wild-type CUL3 3'UTR (CUL3) or with the mutational CUL3 3'UTR (CUL3-M). G Western blotting assays of fibroblasts detected with anti-CUL3 antibody after CUL3 overexpression or siCUL3 treatments. H Western blotting assays of fibroblasts detected with anti- FN1, alpha-SMA, and FAP-1 antibodies after treatments with CUL3 overexpression and miR-1290 mimic transfection, or exosome. I Western blotting assays of fibroblasts were detected with anti- FN1, alpha-SMA, and FAP-1 antibodies after treatments with siCUL3. beta-actin was used as an internal reference. Data were presented as the means ± SEM from three independent experiments. * P



IF analysis of KEAP1 using anti-KEAP1 antibody (A00514-3). KEAP1 was detected in an 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 5 ug/mL rabbit anti-KEAP1 Antibody (A00514-3) 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 U251 cells using anti-KEAP1 antibody (A00514-3). Overlay histogram showing U251 cells stained with A00514-3 (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-KEAP1 Antibody (A00514-3, 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.

3 Publications Citing This Product

1. PubMed ID: 10.1039/C6RA14831J, Individual and combined antioxidant effects of ginsenoside F2 and cyanidin-3-O-glucoside in human

BOSTER BIOLOGICAL TECHNOLOGY
3942 B Valley Ave, Pleasanton, CA 94566
888-466-3604 | support@bosterbio.com | www.bosterbio.com

embryonic kidney 293 cells

2. PubMed ID: 10.1016/j.carbpol.2018.01.075, Polysaccharide from Ostrea rivularis attenuates reproductive oxidative stress damage via activating Keap1-Nrf2/ARE pathway

3. PubMed ID: 10.1002/jcp.26769, Andrographolide protects chondrocytes from oxidative stress injury by activation of the Keap1–Nrf2–Are signaling pathway

Visit bosterbio.com/anti-keap1-picoband-trade-antibody-a00514-3-boster.html to see all 3 publications.

Anti-KEAP1 Antibody

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