

# Anti-IRAK/IRAK1 Antibody Picoband™

Catalog Number: A01021

#### **About IRAK1**

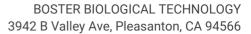
Interleukin-1 receptor-associated kinase 1, also called IRAK1, is an enzyme that in humans is encoded by the IRAK1 gene. By radiation hybrid analysis, this gene is mapped to chromosome Xq28. Serine/threonine-protein kinase plays a critical role in initiating innate immune response against foreign pathogens. This gene involved in Toll-like receptor (TLR) and IL-1R signaling pathways. This gene encodes the interleukin-1 receptor-associated kinase 1, one of two putative serine/threonine kinases that become associated with the interleukin-1 receptor (IL1R) upon stimulation. This gene is partially responsible for IL1-induced upregulation of the transcription factor NF-kappa B.

#### Overview

Product Name	Anti-IRAK/IRAK1 Antibody Picoband™
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-IRAK/IRAK1 Antibody Picoband™ catalog # A01021. Tested in ELISA, Flow Cytometry, IF, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	ELISA, Flow Cytometry, IF, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na <sub>2</sub> HPO <sub>4</sub> , 0.05mg NaN <sub>3</sub> .
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	P51617

### **Technical Details**

Immunogen	E. coli-derived human IRAK recombinant protein (Position: M377-D479).
Predicted Reactive Species	Human
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
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.



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Purification	Immunogen affinity purified.
Suggested Dilutions	Dilute the sample so that the expected range of concentrations fall within the detection range of this kit.  If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples.  Some PubMed article(s) citing the expression level of this target are as follows:  Boster Bio's internal QC testing used:  Western blot, 0.1-0.5ug/ml  Immunocytochemistry/Immunofluorescence, 2ug/ml  Flow Cytometry, 1-3ug/1x10 <sup>6</sup> cells  Direct ELISA, 0.1-0.5ug/ml



## Anti-IRAK/IRAK1 Antibody Picoband™ (A01021) Images



Figure 1. Western blot analysis of IRAK using anti-IRAK antibody (A01021).

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: human HepG2 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-IRAK antigen affinity purified polyclonal antibody (Catalog # A01021) 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 1:10000 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 IRAK at approximately 76KD. The expected band size for IRAK is at 76KD.

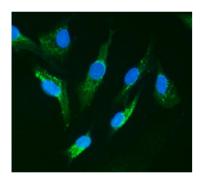


Figure 2. IF analysis of IRAK using anti-IRAK antibody (A01021).

IRAK was detected in immunocytochemical section of A549 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-IRAK Antibody (A01021) 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.

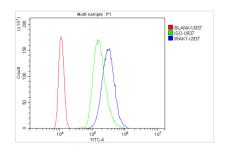


Figure 3. Flow Cytometry analysis of U937 cells using anti-IRAK antibody (A01021).

Overlay histogram showing U937 cells stained with A01021 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-IRAK Antibody (A01021,  $1ug/1x10^6$  cells) for 30 min at  $20^\circ$ C. DyLight § 488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x10<sup>6</sup> cells) was used as secondary antibody for 30 minutes at  $20^\circ$ C. Isotype control antibody (Green line) was rabbit IgG ( $1ug/1x10^6$ ) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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