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Anti-SPHK1 Antibody Picoband™

Catalog Number: A01390-2

About SPHK1

SPHK1(Sphingosine Kinase 1), is an enzyme that in humans is encoded by the SPHK1 gene. Melendez et al.(2000) mapped the SPHK1 gene to chromosome 17q25.2 based on sequence identity with ESTs mapped to this region. Kohama et al.(1998) demonstrated that recombinant mouse Sphk1 can specifically phosphorylate D-erythro-sphingosine and that D, L-threo-dihydrosphingosine and N, N-dimethylsphingosine can act as competitive inhibitors of recombinant Sphk1. Pitson et al.(2000) found that recombinant SPHK1 and endogenous SPHK1 purified from placenta had identical enzymatic characteristics, suggesting posttranslational modification does not effect functional properties.

Overview

Product Name	Anti-SPHK1 Antibody Picoband™
Reactive Species	Human
Description	Boster Bio Anti-SPHK1 Antibody Picoband™ catalog # A01390-2. Tested in ELISA, Flow Cytometry, WB applications. This antibody reacts with Human.
Application	ELISA, Flow Cytometry, 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	Q9NYA1

Technical Details

Immunogen	E.coli-derived human SPHK1 recombinant protein (Position: E55-N357).
Predicted Reactive Species	Human
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 µg/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, 0.25-0.5 μg/ml, Human Flow Cytometry, 1-3 μg/1x10^6 cells, Human Direct ELISA, 0.1-0.5 μg/ml, Human

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Anti-SPHK1 Antibody Picoband™ (A01390-2) Images

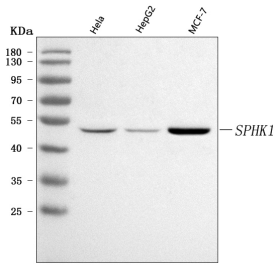

Figure 1. Western blot analysis of SPHK1 using anti-SPHK1 antibody (A01390-2).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.
Lane 1: human Hela whole cell lysates,
Lane 2: human HepG2 whole cell lysates,
Lane 3: human MCF-7 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-SPHK1 antigen affinity purified polyclonal antibody (Catalog # A01390-2) 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 SPHK1 at approximately 52 kDa. The expected band size for SPHK1 is at 42 kDa.

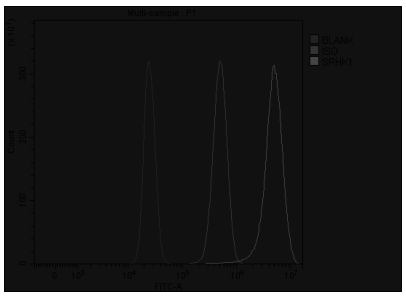

Figure 2. Flow Cytometry analysis of HepG2 cells using anti-SPHK1 antibody (A01390-2).
Overlay histogram showing HepG2 cells stained with A01390-2 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-SPHK1 Antibody (A01390-2, 1 ug/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10 ug/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 ug/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.