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antibody and ELISA experts

BOSTER BIOLOGICAL TECHNOLOGY
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Anti-SIX4 Antibody Picoband™

Catalog Number: A05695-1

About SIX4

Homeobox protein SIX4 is a protein that in humans is encoded by the SIX4 gene. This gene encodes a member of the homeobox family, subfamily SIX. The drosophila homolog is a nuclear homeoprotein required for eye development. Studies in mouse show that this gene product functions as a transcription factor, and may have a role in the differentiation or maturation of neuronal cells.

Overview

Product Name	Anti-SIX4 Antibody Picoband™
Reactive Species	Human
Description	Boster Bio Anti-SIX4 Antibody Picoband™ catalog # A05695-1. Tested in ELISA, IF, ICC, WB, Flow Cytometry applications. This antibody reacts with Human.
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	Q9UIU6

Technical Details

Immunogen	E.coli-derived human SIX4 recombinant protein (Position: Q387-L781).
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 µg/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	Dilute the sample so that the expected range of concentrations fall within the detection range of this kit.

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	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 Immunocytochemistry/Immunofluorescence, 5 μg/ml, Human Flow Cytometry (Fixed), 1-3 μg/1x10^6 cells, Human Direct ELISA, 0.1-0.5 μg/ml, Human

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Anti-SIX4 Antibody Picoband™ (A05695-1) Images

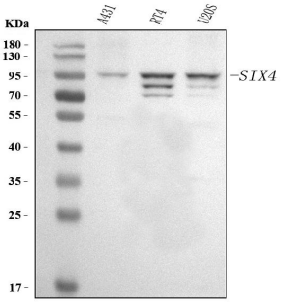

Figure 1. Western blot analysis of SIX4 using anti-SIX4 antibody (A05695-1).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.
Lane 1: human A431 whole cell lysates,
Lane 2: human RT4 whole cell lysates,
Lane 3: human U20S 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-SIX4 antigen affinity purified polyclonal antibody (Catalog # A05695-1) 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 SIX4 at approximately 95 kDa. The expected band size for SIX4 is at 95 kDa.

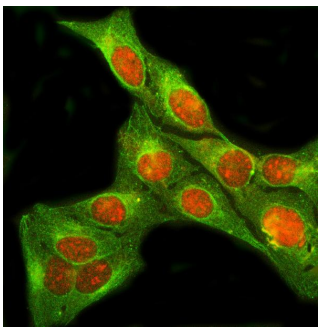

Figure 2. IF analysis of SIX4 using anti-SIX4 antibody (A05695-1) and anti-Beta Tubulin antibody (M01857-3).
SIX4 was detected in immunocytochemical section of U2OS cell. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 ug/mL rabbit anti-SIX4 Antibody (A05695-1) and mouse anti-Beta Tubulin antibody (M01857-3) overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG (BA1032) and DyLight®488 Conjugated Goat Anti-Mouse IgG (BA1126) were used as secondary antibody at 1:500 dilution and incubated for 30 minutes at 37°C. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

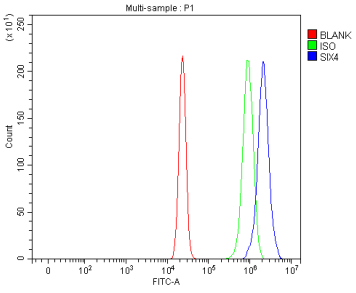

Figure 3. Flow Cytometry analysis of U20S cells using anti-SIX4 antibody (A05695-1).
Overlay histogram showing U20S cells stained with A05695-1 (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-SIX4 Antibody (A05695-1, 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.

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