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Anti-Nanog Antibody Picoband™

Catalog Number: A00153-4

About Nanog

NANOG (pron. nanOg) is a transcription factor critically involved with self-renewal of undifferentiated embryonic stem cells. In humans, this protein is encoded by the NANOG gene. It is mapped to 12p13.31. NANOG is thought to be a key factor in maintaining pluripotency. Moreover, NANOG is also thought to function in concert with other factors such as POU5F1 (Oct-4) and SOX2 to establish ESC identity. The NANOG protein has been found to be a transcriptional activator for the Rex1 promoter, playing a key role in sustaining Rex1 expression. Knockdown of NANOG in embryonic stem cells results in a reduction of Rex1 expression, while forced expression of NANOG stimulates Rex1 expression.

Overview

Product Name	Anti-Nanog Antibody Picoband™
Reactive Species	Mouse, Rat
Description	Boster Bio Anti-Nanog Antibody Picoband™ catalog # A00153-4. Tested in ELISA, Flow Cytometry, IF, ICC, WB applications. This antibody reacts with Mouse, Rat.
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	Q80Z64

Technical Details

Immunogen	E.coli-derived mouse Nanog recombinant protein (Position: M1-A191).
Predicted Reactive Species	Chicken
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, 0.25-0.5 ug/ml, Mouse, Rat Immunocytochemistry/Immunofluorescence, 5 ug/ml, Rat Flow Cytometry, 1-3 ug/1x10^6 cells, Mouse Direct ELISA, 0.1-0.5 ug/ml, Mouse

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Anti-Nanog Antibody Picoband™ (A00153-4) Images

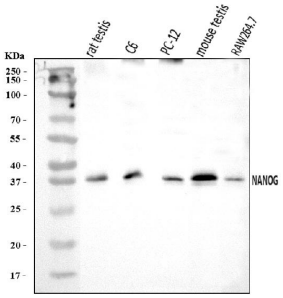

Figure 1. Western blot analysis of Nanog using anti-Nanog antibody (A00153-4).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.
Lane 1: rat testis tissue lysates,
Lane 2: rat C6 whole cell lysates,
Lane 3: rat PC-12 whole cell lysates,
Lane 4: mouse testis tissue lysates,
Lane 5: mouse RAW264.7 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-Nanog antigen affinity purified polyclonal antibody (Catalog # A00153-4) 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 Nanog at approximately 37 kDa. The expected band size for Nanog is at 37 kDa.

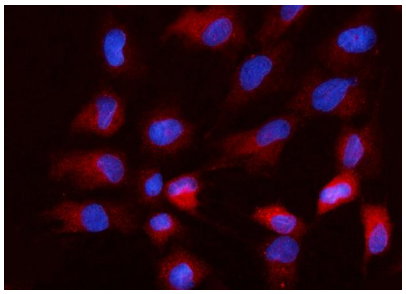

Figure 2. IF analysis of Nanog using anti-Nanog antibody (A00153-4).
Nanog was detected in an immunocytochemical section of NRK cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 ug/mL rabbit anti-Nanog Antibody (A00153-4) overnight at 4°C. DyLight®594 Conjugated Goat Anti-Rabbit IgG (BA1142) 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.

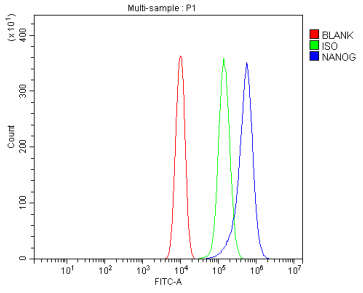

Figure 3. Flow Cytometry analysis of RAW264.7 cells using anti-Nanog antibody (A00153-4).
Overlay histogram showing RAW264.7 cells stained with A00153-4 (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-Nanog Antibody (A00153-4, 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 without incubation with primary antibody and secondary antibody (Red line) was

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used as a blank control.

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