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## Anti-NOV/CCN3 Antibody Picoband<sup>™</sup>

Catalog Number: A06319-1

## About NOV

NOV (nephroblastoma overexpressed), also known as CCN3, is a matricellular protein that in humans is encoded by the NOV gene. The protein encoded by this gene is a small secreted cysteine-rich protein and a member of the CCN family of regulatory proteins. CNN family proteins associate with the extracellular matrix and play an important role in cardiovascular and skeletal development, fibrosis and cancer development.

## Overview

Product Name	Anti-NOV/CCN3 Antibody Picoband™
Reactive Species	Rat
Description	Boster Bio Anti-NOV/CCN3 Antibody Picoband <sup>™</sup> catalog # A06319-1. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Rat.
Application	Flow Cytometry, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.
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	P48745

## **Technical Details**

Immunogen	A synthetic peptide corresponding to a sequence at the C-terminus of human NOV/CCN3, different from the related mouse sequence by seven amino acids, and from the related rat sequence by four amino acids.
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 IHC(P) and 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.
Purification	Immunogen affinity purified.

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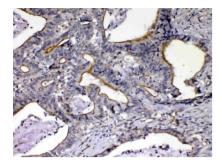
Suggested Dilutions	Dilute the sample so that the expected range of concentrations fall within the detection range of this kit. If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples. Some PubMed article(s) citing the expression level of this target are as follows: Boster Bio's internal QC testing used: Western blot, 0.1-0.5ug/ml, Human Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml, Human, Rat, By Heat Immunocytochemistry/Immunofluorescence, 2ug/ml, Human Flow Cytometry, 1-3ug/1x10 <sup>6</sup> cells, Human
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## Anti-NOV/CCN3 Antibody Picoband<sup>™</sup> (A06319-1) Images



#### Figure 2. IHC analysis of NOV/CCN3 using anti-NOV/CCN3 antibody (A06319-1).

NOV/CCN3 was detected in paraffin-embedded section of human intestinal cancer tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-NOV/CCN3 Antibody (A06319-1) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

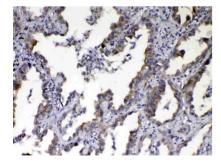


Figure 1. Western blot analysis of NOV/CCN3 using anti-NOV/CCN3 antibody (A06319-1).

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 50ug of sample under reducing conditions.

Lane 1: human Hela whole cell lysate.

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-NOV/CCN3 antigen affinity purified polyclonal antibody (Catalog # A06319-1) 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 1:10000 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 NOV/CCN3 at approximately 47KD. The expected band size for NOV/CCN3 is at 39KD.



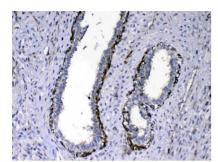
#### Figure 3. IHC analysis of NOV/CCN3 using anti-NOV/CCN3 antibody (A06319-1).

NOV/CCN3 was detected in paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-NOV/CCN3 Antibody (A06319-1) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

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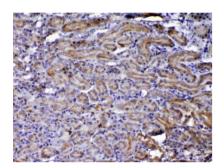
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#### NOV/CCN3 antibody (A06319-1).

NOV/CCN3 was detected in paraffin-embedded section of human mammary cancer tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-NOV/CCN3 Antibody (A06319-1) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.



#### Figure 5. IHC analysis of NOV/CCN3 using anti-NOV/CCN3 antibody (A06319-1).

NOV/CCN3 was detected in paraffin-embedded section of rat kidney tissue . Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-NOV/CCN3 Antibody (A06319-1) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

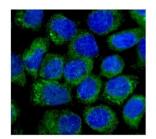


Figure 6. IF analysis of NOV/CCN3 using anti-NOV/CCN3 antibody (A06319-1).

NOV/CCN3 was detected in 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 2ug/mL rabbit anti-NOV/CCN3 Antibody (A06319-1) 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.

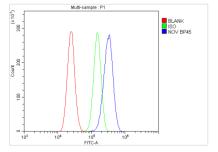


Figure 7. Flow Cytometry analysis of U87 cells using anti-NOV/CCN3 antibody (A06319-1). Overlay histogram showing U87 cells stained with A06319-1 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-NOV/CCN3 Antibody (A06319-1,1ug/1x10<sup>6</sup> cells) for 30 min at 20°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°C. Isotype control antibody (Green line) was rabbit IgG (1ug/1x10<sup>6</sup>) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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