

## Anti-C9ORF72 Antibody Picoband™

Catalog Number: A00419-1

### About C9ORF72

C9orf72 (chromosome 9 open reading frame 72) is a protein which in humans is encoded by the gene C9orf72. It is mapped to 9p21.2. The protein encoded by this gene plays an important role in the regulation of endosomal trafficking, and has been shown to interact with Rab proteins that are involved in autophagy and endocytic transport. Expansion of a GGGGCC repeat from 2-22 copies to 700-1600 copies in the intronic sequence between alternate 5' exons in transcripts from this gene is associated with 9p-linked ALS (amyotrophic lateral sclerosis) and FTD (frontotemporal dementia) (PMID: 21944778, 21944779). Studies suggest that hexanucleotide expansions could result in the selective stabilization of repeat-containing pre-mRNA, and the accumulation of insoluble dipeptide repeat protein aggregates that could be pathogenic in FTD-ALS patients (PMID: 23393093). Alternative splicing results in multiple transcript variants encoding different isoforms.

### Overview

Product Name	Anti-C9ORF72 Antibody Picoband™
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-C9ORF72 Antibody Picoband™ catalog # A00419-1. Tested in Flow Cytometry, IHC-P, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	Flow Cytometry, IHC-P, 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	Q96LT7

### Technical Details

Immunogen	A synthetic peptide corresponding to a sequence at the N-terminus of human C9ORF72, which shares 96.4% amino acid (aa) sequence identity with both mouse and rat C9ORF72.
Predicted Reactive Species	Chicken
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 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	<p>Dilute the sample so that the expected range of concentrations fall within the detection range of this kit.</p> <p>If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples.</p> <p>Some PubMed article(s) citing the expression level of this target are as follows:</p> <p>Boster Bio's internal QC testing used:</p> <p>"Western blot, 0.25-0.5ug/ml, Human, Mouse, Rat</p> <p>Flow Cytometry, 1-3ug/1x10<sup>6</sup> cells, Human</p> <p>"</p>

## Anti-C9ORF72 Antibody Picoband™ (A00419-1) Images

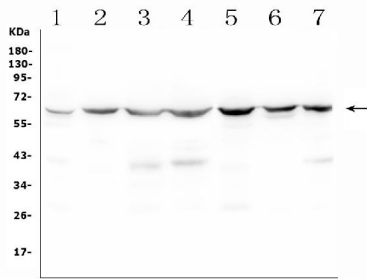


Figure 1. Western blot analysis of C9ORF72 using anti-C9ORF72 antibody (A00419-1).

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 placenta tissue lysates,  
Lane 2: human U2OS whole cell lysates,  
Lane 3: human PC-3 whole cell lysates,  
Lane 4: human K562 whole cell lysates,  
Lane 5: human Hela whole cell lysates,  
Lane 6: human A431 whole cell lysates,  
Lane 7: human A549 whole 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-C9ORF72 antigen affinity purified polyclonal antibody (Catalog # A00419-1) 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 C9ORF72 at approximately 65KD. The expected band size for C9ORF72 is at 65KD.

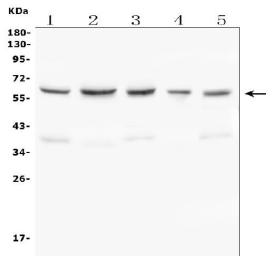


Figure 2. Western blot analysis of C9ORF72 using anti-C9ORF72 antibody (A00419-1).

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: rat brain tissue lysates,  
Lane 2: rat liver tissue lysates,  
Lane 3: rat C6 whole cell lysates,  
Lane 4: mouse liver tissue lysates,  
Lane 5: mouse HEPA1-6 whole 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-C9ORF72 antigen affinity purified polyclonal antibody (Catalog # A00419-1) 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 C9ORF72 at approximately 65KD. The expected band size for C9ORF72 is at 65KD.

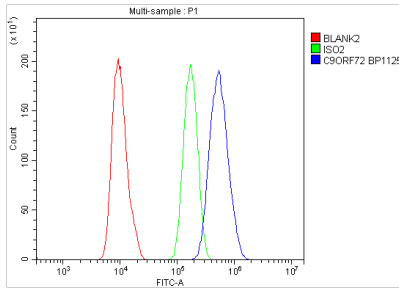


Figure 3. Flow Cytometry analysis of U2OS cells using anti-C9ORF72 antibody (A00419-1).

Overlay histogram showing U2OS cells stained with A00419-1 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-C9ORF72 Antibody (A00419-1, 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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Anti-C9ORF72 Antibody <sup>™</sup>