

Anti-FACL4/ACSL4 Antibody Picoband™

Catalog Number: A04372-2

About ACSL4

Long-chain-fatty-acid—CoA ligase 4 is an enzyme that in humans is encoded by the ACSL4 gene. It is mapped to Xq23. The protein encoded by this gene is an isozyme of the long-chain fatty-acid-coenzyme A ligase family. Although differing in substrate specificity, subcellular localization, and tissue distribution, all isozymes of this family convert free long-chain fatty acids into fatty acyl-CoA esters, and thereby play a key role in lipid biosynthesis and fatty acid degradation. This isozyme preferentially utilizes arachidonate as substrate. The absence of this enzyme may contribute to the cognitive disability or Alport syndrome. Alternative splicing of this gene generates multiple transcript variants.

Overview

| Product Name | Anti-FACL4/ACSL4 Antibody Picoband™ |
|----------------------|---|
| Reactive Species | Human, Mouse, Rat |
| Description | Boster Bio Anti-FACL4/ACSL4 Antibody Picoband™ catalog # A04372-2. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat. |
| Application | Flow Cytometry, IF, IHC, ICC, WB |
| Clonality | Polyclonal |
| Formulation | Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.01mg 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 | O60488 |

Technical Details

| Immunogen | A synthetic peptide corresponding to a sequence at the C-terminus of human FACL4/ACSL4. |
|-------------------------------|---|
| 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, 0.1-0.25ug/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 2-5ug/ml, Mouse, Rat Immunocytochemistry/Immunofluorescence, 5ug/ml, Human Flow Cytometry, 1-3ug/1x10 ⁶ cells, Human |
|---------------------|---|
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Anti-FACL4/ACSL4 Antibody Picoband™ (A04372-2) Images

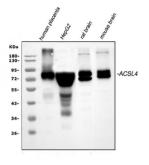


Figure 1. Western blot analysis of FACL4/ACSL4 using anti-FACL4/ACSL4 antibody (A04372-2).

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 placenta tissue lysates,

Lane 2: human HepG2 whole cell lysates,

Lane 3: rat brain tissue lysates,

Lane 4: mouse brain tissue 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-FACL4/ACSL4 antigen affinity purified polyclonal antibody (Catalog # A04372-2) at 0.25 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 FACL4/ACSL4 at approximately 79 kDa. The expected band size for FACL4/ACSL4 is at 79 kDa.

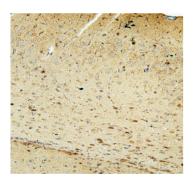


Figure 2. IHC analysis of FACL4/ACSL4 using anti-FACL4/ACSL4 antibody (A04372-2).

FACL4/ACSL4 was detected in a paraffin-embedded section of mouse brain tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-FACL4/ACSL4 Antibody (A04372-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

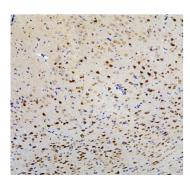


Figure 3. IHC analysis of FACL4/ACSL4 using anti-FACL4/ACSL4 antibody (A04372-2).

FACL4/ACSL4 was detected in a paraffin-embedded section of rat brain tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-FACL4/ACSL4 Antibody (A04372-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



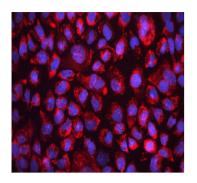


Figure 4. IF analysis of FACL4/ACSL4 using anti-FACL4/ACSL4 antibody (A04372-2).

FACL4/ACSL4 was detected in an 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 5 ug/mL rabbit anti-FACL4/ACSL4 Antibody (A04372-2) overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG (BA1032) was used as secondary antibody at 1:500 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.

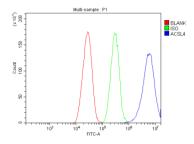


Figure 5. Flow Cytometry analysis of HepG2 cells using anti-FACL4/ACSL4 antibody (A04372-2).

Overlay histogram showing HepG2 cells stained with A04372-2 (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-FACL4/ACSL4 Antibody (A04372-2, 1 ug/1x10 6 cells) for 30 min at 20 $^\circ$ C. DyLight 6 488 conjugated goat anti-rabbit IgG (BA1127, 5-10 ug/1x10 6 cells) was used as secondary antibody for 30 minutes at 20 $^\circ$ 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.

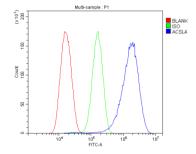


Figure 6. Flow Cytometry analysis of U87 cells using anti-FACL4/ACSL4 antibody (A04372-2).

Overlay histogram showing U87 cells stained with A04372-2 (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-FACL4/ACSL4 Antibody (A04372-2, 1 ug/1x10 6 cells) for 30 min at 20 $^\circ$ 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 $^\circ$ 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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