

Anti-AIF/AIFM1 Antibody Picoband™ (monoclonal, 2I5)

Catalog Number: M01571-1

About AIFM1

Apoptosis-inducing factor 1, mitochondrial, also known as AIF or PDCD8 is a protein that in humans is encoded by the AIFM1 gene. AIFM1 gene is mapped to Xq26.1 based on an alignment of the AIFM1 sequence with the genomic sequence. This gene encodes a flavoprotein essential for nuclear disassembly in apoptotic cells, and it is found in the mitochondrial intermembrane space in healthy cells. Induction of apoptosis results in the translocation of this protein to the nucleus where it affects chromosome condensation and fragmentation. In addition, this gene product induces mitochondria to release the apoptogenic proteins cytochrome c and caspase-9. Mutations in this gene cause combined oxidative phosphorylation deficiency 6, which results in a severe mitochondrial encephalomyopathy. A related pseudogene has been identified on chromosome 10.

Overview

Product Name	Anti-AIF/AIFM1 Antibody Picoband™ (monoclonal, 2I5)
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-AIF/AIFM1 Antibody Picoband™ (monoclonal, 2I5) catalog # M01571-1. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	Flow Cytometry, IF, IHC, ICC, WB
Clonality	Monoclonal 2I5
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	Mouse
Uniprot ID	O95831

Technical Details

Immunogen	A synthetic peptide corresponding to a sequence at the C-terminus of human AIF, identical to the related mouse and rat sequences.
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Mouse IgG (EK1001) for Western blot, and HRP Conjugated anti-Mouse IgG Super Vision Assay Kit (SV0001-1) for IHC(P) and ICC.
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Mouse IgG1
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.

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

Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml

Immunofluorescence, 2ug/ml

Immunocytochemistry/Immunofluorescence, 5ug/ml

Flow Cytometry, 1-3ug/1x10⁶ cells

Anti-AIF/AIFM1 Antibody Picoband™ (monoclonal, 2I5) (M01571-1) Images

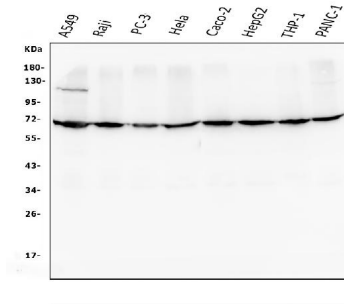


Figure 1. Western blot analysis of AIF using anti-AIF antibody (M01571-1).

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: A549 whole cell lysates,
Lane 2: Raji whole cell lysates,
Lane 3: PC-3 whole cell lysates,
Lane 4: HeLa whole cell lysates,
Lane 5: Caco-2 whole cell lysates,
Lane 6: HepG2 whole cell lysates,
Lane 7: THP-1 whole cell lysates,
Lane 8: PANC-1 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 mouse anti-AIF antigen affinity purified monoclonal antibody (Catalog # M01571-1) 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-mouse 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 # EK1001) with Tanon 5200 system. A specific band was detected for AIF at approximately 70KD. The expected band size for AIF is at 70KD.

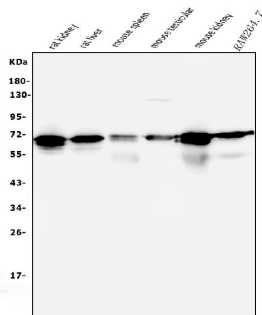


Figure 2. Western blot analysis of AIF using anti-AIF antibody (M01571-1).

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 kidney tissue lysates,
Lane 2: rat liver tissue lysates,
Lane 3: mouse spleen lysates,
Lane 4: mouse testicular tissue lysates,
Lane 5: mouse kidney tissue lysates,
Lane 6: RAW264.7 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 mouse anti-AIF antigen affinity purified monoclonal antibody (Catalog # M01571-1) 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-mouse 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 # EK1001) with Tanon 5200 system. A specific band was detected for AIF at approximately 70KD. The expected band size for AIF is at 70KD.

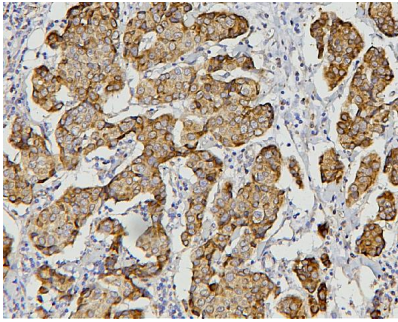


Figure 3. IHC analysis of AIF using anti-AIF antibody (M01571-1).

AIF was detected in paraffin-embedded section of human mammary cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml mouse anti-AIF Antibody (M01571-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

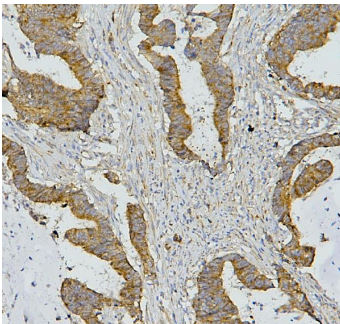


Figure 4. IHC analysis of AIF using anti-AIF antibody (M01571-1).

AIF was detected in paraffin-embedded section of human colon cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml mouse anti-AIF Antibody (M01571-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

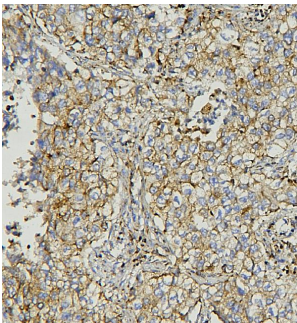


Figure 5. IHC analysis of AIF using anti-AIF antibody (M01571-1).

AIF was detected in paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml mouse anti-AIF Antibody (M01571-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

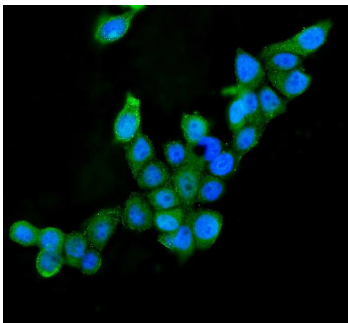
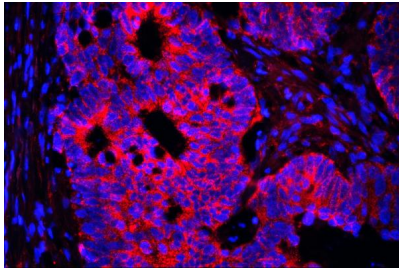


Figure 5. IF analysis of AIF using anti-AIF antibody (M01571-1).

AIF was detected in immunocytochemical section of MCF-7 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 5ug/mL mouse anti-AIF Antibody (M01571-1) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Mouse IgG (BA1126) 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 6. IF analysis of AIF using anti-AIF antibody (M01571-1).

AIF was detected in paraffin-embedded section of human intestinal cancer tissue. Heat mediated antigen retrieval was



performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/mL mouse anti-AIF Antibody (M01571-1) overnight at 4°C. DyLight®550 Conjugated Goat Anti-Mouse IgG (BA1133) 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.

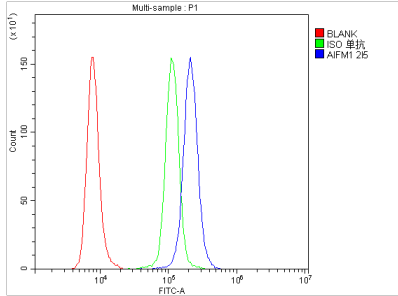


Figure 7. Flow Cytometry analysis of Raji cells using anti-AIF antibody (M01571-1). Overlay histogram showing Raji cells stained with M01571-1 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-AIF Antibody (M01571-1, 1ug/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10ug/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1ug/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

3 Publications Citing This Product

1. PubMed ID: 27895670, Wnt5a Increases Properties of Lung Cancer Stem Cells and Resistance to Cisplatin through Activation of Wnt5a/PKC Signaling Pathway
2. PubMed ID: 26645545, Toll-like receptor 4 contributes to chronic itch, allodynia, and spinal astrocyte activation in male mice
3. PubMed ID: 27330750, Mechanism of apoptosis induction in human hepatocellular carcinoma cells following treatment with a gecko peptides mixture

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