

Anti-PPCS Antibody Picoband™ (monoclonal, 7G13)

Catalog # ABO14871

Product Information

Application	WB, IHC, FC
Primary Accession	Q9HAB8
Host	Mouse
Isotype	Mouse IgG2a
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-PPCS Antibody Picoband™ (monoclonal, 7G13) . Tested in Flow Cytometry, IHC, WB applications. This antibody reacts with Human, Mouse, Rat.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500 µg/ml.

Additional Information

Gene ID	79717
Other Names	Phosphopantothenate--cysteine ligase, 6.3.2.51, Phosphopantothenoylcysteine synthetase, PPC synthetase, PPCS, COAB
Calculated MW	34005
Application Details	Western blot, 0.1-0.5 µg/ml, Human Immunohistochemistry (Paraffin-embedded Section), 0.5-1 µg/ml, Human, Mouse, Rat, By Heat Flow Cytometry, 1-3 µg/1x10 ⁶ cells, Human
Subcellular Localization	Cytosol.
Source	Eukaryota
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na ₂ HPO ₄ , 0.05mg NaN ₃ .
Clone Names	Clone: 7G13
Immunogen	A synthetic peptide corresponding to a sequence at the C-terminus of human PPCS, which shares 73.9% and 78.2% amino acid (aa) sequence identity with mouse and rat PPCS, respectively.
Cross Reactivity	No cross-reactivity with other proteins.
Storage	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.

Protein Information

Name	PPCS
Synonyms	COAB
Function	Catalyzes the second step in the biosynthesis of coenzyme A from vitamin B5, where cysteine is conjugated to 4'-phosphopantothenate to form 4-phosphopantothenoylcysteine (PubMed: 11923312 , PubMed: 12906824 , PubMed: 29754768). Has a preference for ATP over CTP as a cosubstrate (PubMed: 11923312).

Background

Phosphopantothenoylcysteine synthetase in humans is encoded by the PPCS gene. Biosynthesis of coenzyme A (CoA) from pantothenic acid (vitamin B5) is an essential universal pathway in prokaryotes and eukaryotes. PPCS, one of the last enzymes in this pathway, converts phosphopantothenate to phosphopantothenoylcysteine. By genomic sequence analysis, this PPCS gene is mapped to chromosome 1.

Images

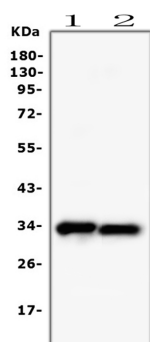


Figure 1. Western blot analysis of PPCS using anti-PPCS antibody (M05567).

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 A375 whole cell lysates

Lane 2: human COLO-320 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-PPCS antigen affinity purified monoclonal antibody (Catalog # M05567) at 0.5 µg/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 1:10000 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 PPCS at approximately 34KD. The expected band size for PPCS is at 34KD.

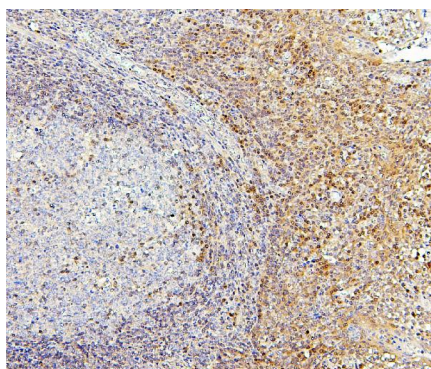


Figure 2. IHC analysis of PPCS using anti-PPCS antibody (M05567).

PPCS was detected in paraffin-embedded section of human tonsil tissues. 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 1 µg/ml mouse anti-PPCS Antibody (M05567) 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.

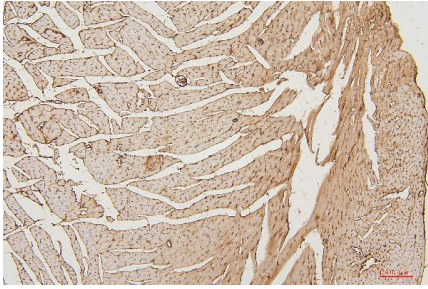


Figure 3. IHC analysis of PPCS using anti-PPCS antibody (M05567).

PPCS was detected in paraffin-embedded section of mouse cardiac muscle tissues. 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 1 $\mu\text{g}/\text{ml}$ mouse anti-PPCS Antibody (M05567) 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.

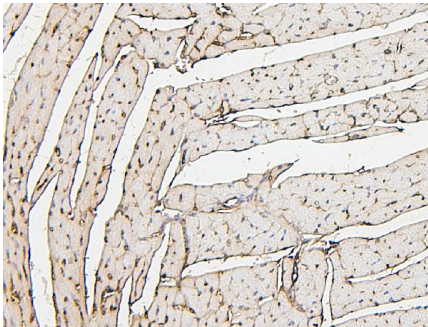


Figure 4. IHC analysis of PPCS using anti-PPCS antibody (M05567).

PPCS was detected in paraffin-embedded section of mouse cardiac muscle tissues. 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 1 $\mu\text{g}/\text{ml}$ mouse anti-PPCS Antibody (M05567) 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.

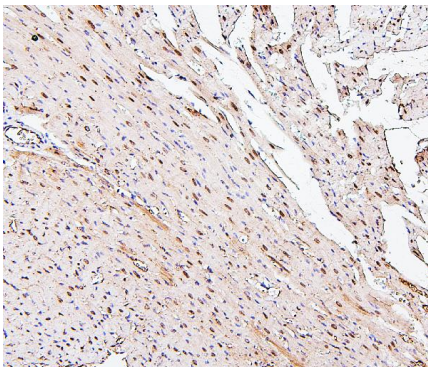


Figure 5. IHC analysis of PPCS using anti-PPCS antibody (M05567).

PPCS was detected in paraffin-embedded section of rat cardiac muscle tissues. 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 1 $\mu\text{g}/\text{ml}$ mouse anti-PPCS Antibody (M05567) 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.

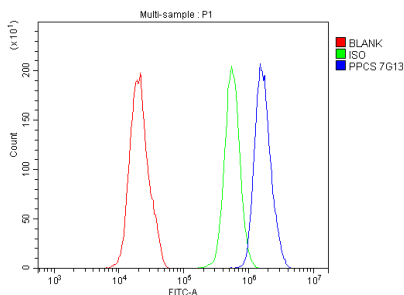


Figure 6. Flow Cytometry analysis of PC-3 cells using anti-PPCS antibody (M05567).

Overlay histogram showing PC-3 cells stained with M05567 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-PPCS Antibody (M05567, 1 $\mu\text{g}/1 \times 10^6$ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 $\mu\text{g}/1 \times 10^6$ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 $\mu\text{g}/1 \times 10^6$) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

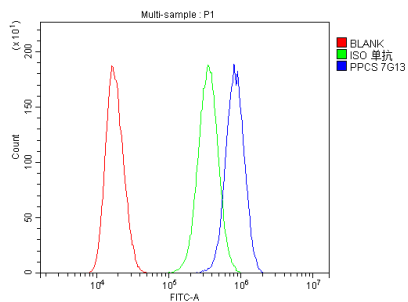


Figure 7. Flow Cytometry analysis of SiHa cells using anti-PPCS antibody (M05567). Overlay histogram showing SiHa cells stained with M05567 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-PPCS Antibody (M05567, 1 μ g/ 1×10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 μ g/ 1×10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 μ g/ 1×10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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