

Anti-PPT1 Antibody Picoband™ (monoclonal, 10F3)

Catalog # ABO14860

Product Information

Application	WB, IHC, FC
Primary Accession	P50897
Host	Mouse
Isotype	Mouse IgG2b
Reactivity	Human
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-PPT1 Antibody Picoband™ (monoclonal, 10F3) . Tested in Flow Cytometry, IHC, WB applications. This antibody reacts with Human.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500 µg/ml.

Additional Information

Gene ID	5538
Other Names	Palmitoyl-protein thioesterase 1, PPT-1, 3.1.2.22, Palmitoyl-protein hydrolase 1, PPT1, CLN1 {ECO:0000303 PubMed:19941651}, PPT
Calculated MW	34193
Application Details	Western blot, 0.1-0.5 µg/ml Immunohistochemistry (Paraffin-embedded Section), 0.5-1 µg/ml Flow Cytometry, 1-3 µg/1x10 ⁶ cells
Subcellular Localization	Lysosome. Secreted.
Source	Eukaryota
Protein Name	Palmitoyl-protein thioesterase 1
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na ₂ HPO ₄ , 0.05mg NaN ₃ .
Clone Names	Clone: 10F3
Immunogen	A synthetic peptide corresponding to a sequence at the C-terminus of human PPT1, different from the related mouse and rat sequences by four amino acids.
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	PPT1
Synonyms	CLN1 {ECO:0000303 PubMed:19941651}, PPT
Function	Has thioesterase activity against fatty acid thioesters with 14 -18 carbons, including palmitoyl-CoA, S-palmitoyl-N- acetylcysteamine, and palmitoylated proteins (PubMed: 12855696 , PubMed: 26731412 , PubMed: 8816748). In contrast to PPT2, PPT1 can hydrolyze palmitoylated proteins and palmitoylcysteine (PubMed: 12855696).
Cellular Location	Lysosome. Secreted Golgi apparatus. Endoplasmic reticulum

Background

Palmitoyl-protein thioesterase 1 (PPT-1), also known as palmitoyl-protein hydrolase 1, is an enzyme that in humans is encoded by the PPT1 gene. PPT-1 is a member of the palmitoyl protein thioesterase family. The protein encoded by this gene is a small glycoprotein involved in the catabolism of lipid-modified proteins during lysosomal degradation. The encoded enzyme removes thioester-linked fatty acyl groups such as palmitate from cysteine residues. Defects in this gene are a cause of infantile neuronal ceroid lipofuscinosis 1 (CLN1, or INCL) and neuronal ceroid lipofuscinosis 4 (CLN4). Two transcript variants encoding different isoforms have been found for this gene.

Images

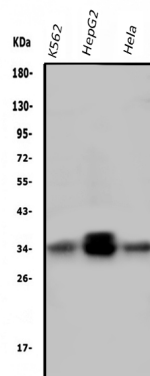


Figure 1. Western blot analysis of PPT1 using anti-PPT1 antibody (M02690).

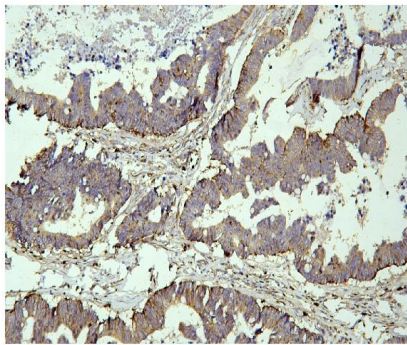
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.

Lane 1: human K562 whole cell lysates,
Lane 2: human HepG2 whole cell lysates,
Lane 3: human HeLa 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-PPT1 antigen affinity purified monoclonal antibody (Catalog # M02690) 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 PPT1 at approximately 34KD. The expected band size for PPT1 is at 34KD.

Figure 2. IHC analysis of PPT1 using anti PPT1 antibody (M02690).

PPT1 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 1 µg/ml mouse anti-PPT1 Antibody (M02690) 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.

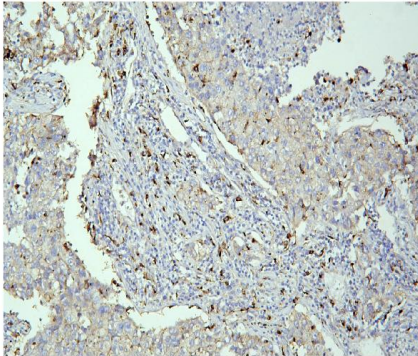


Figure 3. IHC analysis of PPT1 using anti PPT1 antibody (M02690). PPT1 was detected in paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-PPT1 Antibody (M02690) 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.

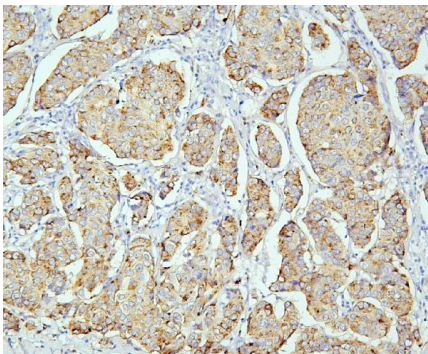


Figure 4. IHC analysis of PPT1 using anti PPT1 antibody (M02690). PPT1 was detected in paraffin-embedded section of human mammary cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-PPT1 Antibody (M02690) 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.

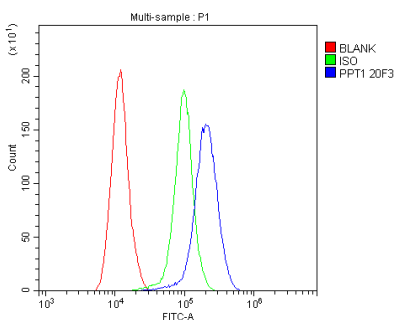


Figure 5. Flow Cytometry analysis of THP-1 cells using anti-PPT1 antibody (M02690). Overlay histogram showing THP-1 cells stained with M02690 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-PPT1 Antibody (M02690, 1 µg/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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