

# Anti-ALPP Antibody Picoband™ (monoclonal, 6C7G3)

Catalog # ABO16233

## Product Information

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|--------------------------|----------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Application</b>       | WB, IHC, IF, ICC, FC                                                                                                                         |
| <b>Primary Accession</b> | <a href="#">P05187</a>                                                                                                                       |
| <b>Host</b>              | Mouse                                                                                                                                        |
| <b>Isotype</b>           | Mouse IgG2b                                                                                                                                  |
| <b>Reactivity</b>        | Human                                                                                                                                        |
| <b>Clonality</b>         | Monoclonal                                                                                                                                   |
| <b>Format</b>            | Lyophilized                                                                                                                                  |
| <b>Description</b>       | Anti-ALPP Antibody Picoband™ (monoclonal, 6C7G3) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human. |
| <b>Reconstitution</b>    | Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.                                                                    |

## Additional Information

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| <b>Gene ID</b>             | 250                                                                                                                                                                                                                                                               |
| <b>Other Names</b>         | Alkaline phosphatase, placental type, 3.1.3.1, Alkaline phosphatase Regan isozyme, Placental alkaline phosphatase 1, PLAP-1, ALPP ( <a href="#">HGNC:439</a> )                                                                                                    |
| <b>Calculated MW</b>       | 57954                                                                                                                                                                                                                                                             |
| <b>Application Details</b> | Western blot, 0.25-0.5 µg/ml, Human<br>Immunohistochemistry(Paraffin-embedded Section), 2-5 µg/ml, Human<br>Immunocytochemistry/Immunofluorescence, 5 µg/ml, Human<br>Immunofluorescence, 5 µg/ml, Human<br>Flow Cytometry, 1-3 µg/1x10 <sup>6</sup> cells, Human |
| <b>Source</b>              | Eukaryota                                                                                                                                                                                                                                                         |
| <b>Contents</b>            | Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na <sub>2</sub> HPO <sub>4</sub> .                                                                                                                                                                      |
| <b>Clone Names</b>         | Clone: 6C7G3                                                                                                                                                                                                                                                      |
| <b>Immunogen</b>           | A synthetic peptide corresponding to a sequence at the C-terminus of human ALPP.                                                                                                                                                                                  |
| <b>Purification</b>        | Immunogen affinity purified.                                                                                                                                                                                                                                      |
| <b>Storage</b>             | 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 freezing and thawing.                                                                 |

## Protein Information

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|--------------------------|----------------------------------------------------------------------|
| <b>Name</b>              | ALPP ( <a href="#">HGNC:439</a> )                                    |
| <b>Function</b>          | Alkaline phosphatase that can hydrolyze various phosphate compounds. |
| <b>Cellular Location</b> | Cell membrane; Lipid-anchor, GPI-anchor                              |
| <b>Tissue Location</b>   | Detected in placenta (at protein level).                             |

## Background

Alkaline phosphatase, placental type also known as placental alkaline phosphatase (PLAP) is an allosteric enzyme that in humans is encoded by the ALPP gene. The protein encoded by this gene is an alkaline phosphatase, a metalloenzyme that catalyzes the hydrolysis of phosphoric acid monoesters. It belongs to a multigene family composed of four alkaline phosphatase isoenzymes. The enzyme functions as a homodimer and has a catalytic site containing one magnesium and two zinc ions, which are required for its enzymatic function. The protein is primarily expressed in placental and endometrial tissue; however, strong ectopic expression has been detected in ovarian adenocarcinoma, serous cystadenocarcinoma, and other ovarian cancer cells.

## Images

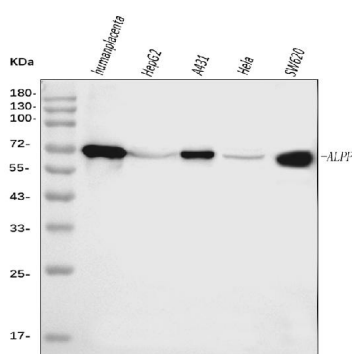


Figure 1. Western blot analysis of ALPP using anti-ALPP antibody (M01718-4).

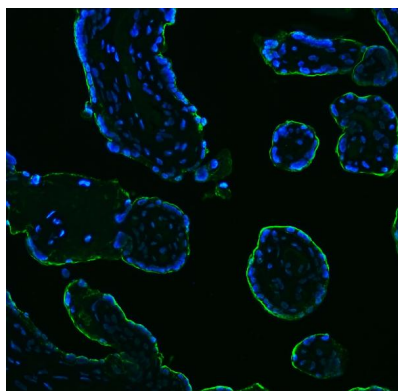
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

Lane 1: human placenta tissue lysates,  
 Lane 2: human HepG2 whole cell lysates,  
 Lane 3: human A431 whole cell lysates,  
 Lane 4: human Hela whole cell lysates,  
 Lane 5: human SW620 whole cell 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 mouse anti-ALPP antigen affinity purified monoclonal antibody (Catalog # M01718-4) 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 ALPP at approximately 70 kDa. The expected band size for ALPP is at 70 kDa.

Figure 2. IF analysis of ALPP using anti-ALPP antibody (M01718-4).

ALPP was detected in a paraffin-embedded section of human placenta 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 5 µg/mL mouse anti-ALPP Antibody (M01718-4) overnight at 4°C. Biotin conjugated goat anti-mouse IgG (BA1001) was used as secondary antibody and incubated for 30



minutes at 37°C. The tissue section was developed using DyLight®488 Conjugated Avidin (BA1128). The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

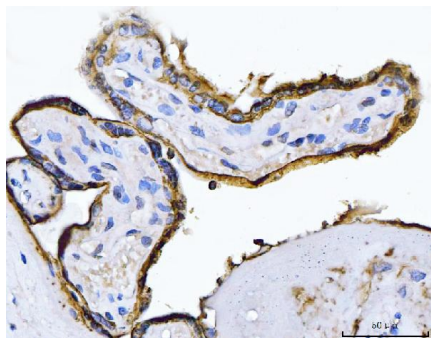


Figure 3. IHC analysis of ALPP using anti-ALPP antibody (M01718-4). ALPP was detected in a paraffin-embedded section of human placenta tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-ALPP Antibody (M01718-4) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.

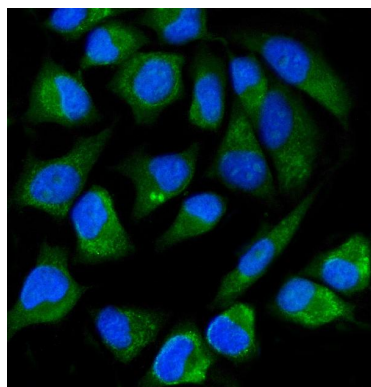


Figure 4. IF analysis of ALPP using anti-ALPP antibody (M01718-4). ALPP was detected in an immunocytochemical section of HeLa cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 µg/mL mouse anti-ALPP Antibody (M01718-4) 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.

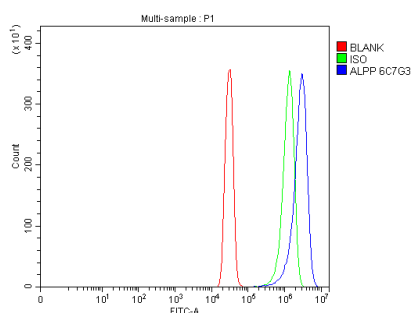


Figure 5. Flow Cytometry analysis of SiHa cells using anti-ALPP antibody (M01718-4). Overlay histogram showing SiHa cells stained with M01718-4 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-ALPP Antibody (M01718-4, 1 µg/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10<sup>6</sup> cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10<sup>6</sup>) used under the same conditions. Unlabelled sample (Red line) was also used as a control.