

## Product Datasheet

### ALPP Mouse Monoclonal Antibody (orb1145867)

<b>Catalog Number</b>	orb1145867
<b>Category</b>	Antibodies
<b>Description</b>	Anti-ALPP Antibody (monoclonal, 6C7G3). Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human.
<b>Target</b>	Alkaline phosphatase, placental type
<b>Clonality</b>	Monoclonal
<b>Species/Host</b>	Mouse
<b>Isotype</b>	Mouse IgG2b
<b>Conjugation</b>	Unconjugated
<b>Reactivity</b>	Human
<b>Form/Appearance</b>	Lyophilized
<b>Concentration</b>	Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.
<b>Buffer/Preservatives</b>	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na <sub>2</sub> HPO <sub>4</sub> .
<b>Reconstitution</b>	Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.
<b>Purification</b>	Immunogen affinity purified.
<b>Immunogen</b>	A synthetic peptide corresponding to a sequence at the C-terminus of human ALPP.
<b>UniProt ID</b>	<b>P05187</b>
<b>MW</b>	70 kDa

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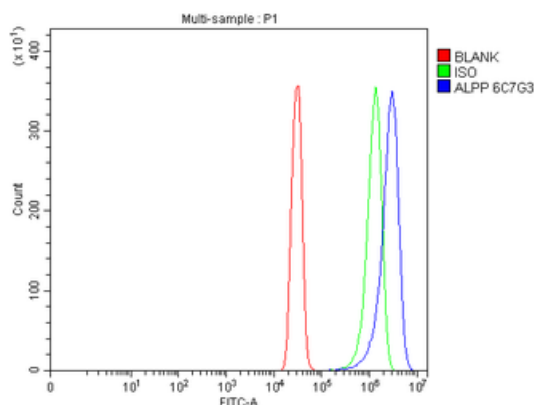
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<b>Tested applications</b>	FC, ICC, IF, IHC, WB
<b>Dilution range</b>	Western blot, 0.25-0.5 µg/ml, Human Immunohistochemistry(Paraffin-embedded Section), 2-5 µg/ml, Human Immunocytochemistry/Immunofluorescence, 5 µg/ml, Human Immunofluorescence, 5 µg/ml, Human Flow Cytometry (Fixed), 1-3 µg/1x10 <sup>6</sup> cells, Human
<b>Cross Reactivity</b>	No cross-reactivity with other proteins.
<b>Antibody Type</b>	Primary Antibody
<b>Clone Number</b>	6C7G3
<b>Storage</b>	Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at -20°C in small aliquots to prevent freeze-thaw cycles.
<b>Note</b>	For research use only
<b>Expiration Date</b>	12 months from date of receipt.



Flow Cytometry analysis of SiHa cells using anti-ALPP antibody. Overlay histogram showing SiHa cells (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 mouse anti-ALPP Antibody (1 µg/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (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 without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

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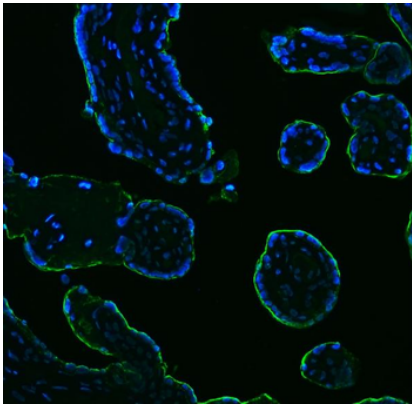
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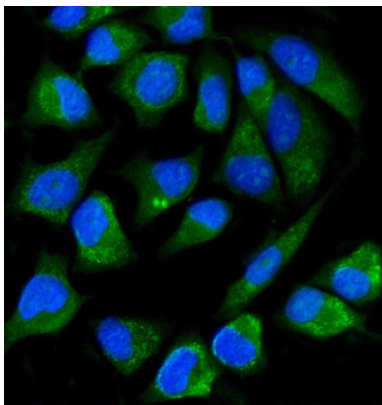
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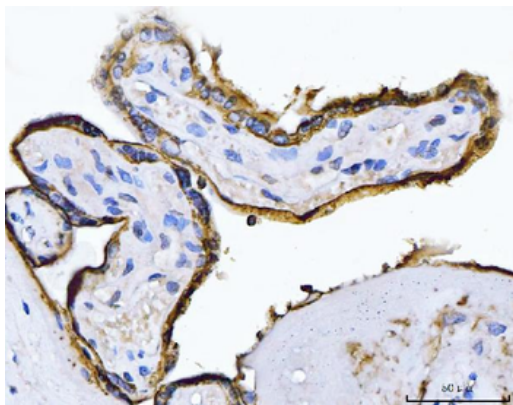
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IF analysis of ALPP using anti-ALPP antibody. ALPP was detected in a paraffin-embedded section of human placenta tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 5 µg/mL mouse anti-ALPP Antibody overnight at 4°C. Biotin conjugated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using DyLight®488 Conjugated Avidin. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.



IF analysis of ALPP using anti-ALPP antibody. ALPP was detected in an immunocytochemical section of HeLa cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 µg/mL mouse anti-ALPP Antibody overnight at 4°C. DyLight®488 Conjugated Goat Anti-Mouse IgG 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.



IHC analysis of ALPP using anti-ALPP antibody. ALPP was detected in a paraffin-embedded section of human placenta 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 2 µg/ml mouse anti-ALPP Antibody 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 with DAB as the chromogen.

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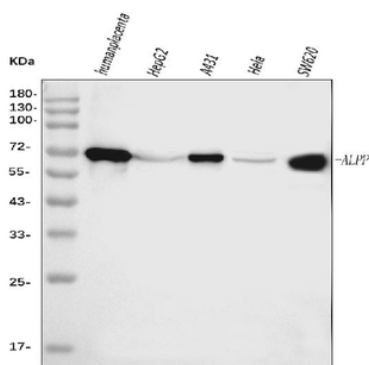
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Western blot analysis of ALPP using anti-ALPP antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. 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 at 0.5  $\mu\text{g}/\text{mL}$  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 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.

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