

## Product Datasheet

### SP1 Mouse Monoclonal Antibody (orb1474870)

|                             |   |
|-----------------------------|---|
| <b>Catalog Number</b>       | orb1474870  |
| <b>Category</b>             | Antibodies  |
| <b>Description</b>          | Anti-SP1 Antibody (monoclonal, B5E6E5). Tested in Flow Cytometry, IF, ICC, IHC, WB applications. This antibody reacts with Human. |
| <b>Target</b>               | Transcription factor Sp1  |
| <b>Clonality</b>            | Monoclonal  |
| <b>Species/Host</b>         | Mouse   |
| <b>Isotype</b>              | IgG1  |
| <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>            | E. coli-derived human SP1 recombinant protein (Position: Q384-A603).  |
| <b>UniProt ID</b>           | <b>P08047</b>   |
| <b>MW</b>                   | 90 kDa  |

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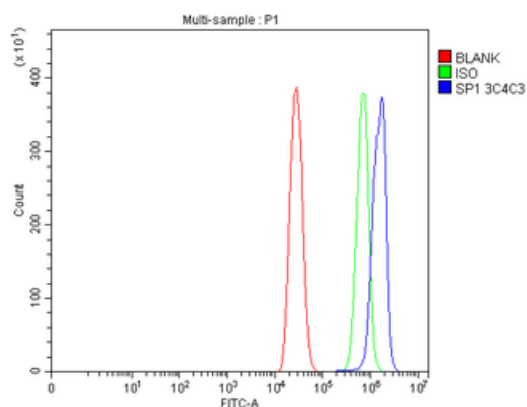
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|                            |  |
|----------------------------|--|
| <b>Tested applications</b> | FC, ICC, IF, IHC, WB   |
| <b>Dilution range</b>      | Western blot, 0.25-0.5 µg/ml, Human Immunohistochemistry(Paraffin-embedded Section), 2-5 µg/ml, Human Immunocytochemistry/Immunofluorescence, 5 µg/ml, Human 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>        | B5E6E5   |
| <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 A431 cells using anti-SP1 antibody. Overlay histogram showing A431 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-SP1 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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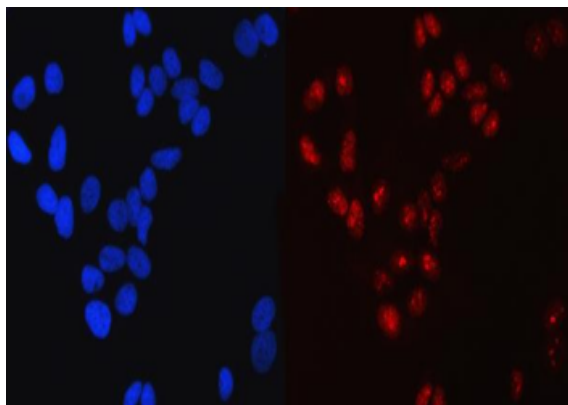
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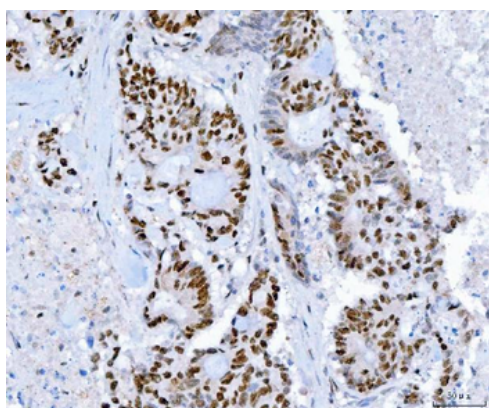
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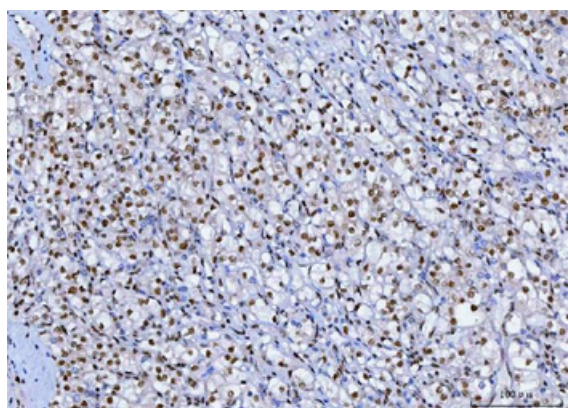
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IF analysis of SP1 using anti-SP1 antibody. SP1 was detected in an immunocytochemical section of HeLa cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 µg/mL mouse anti-SP1 Antibody overnight at 4°C. DyLight®550 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 SP1 using anti-SP1 antibody. SP1 was detected in a paraffin-embedded section of human colorectal adenocarcinoma 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-SP1 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.



IHC analysis of SP1 using anti-SP1 antibody. SP1 was detected in a paraffin-embedded section of human glioma 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-SP1 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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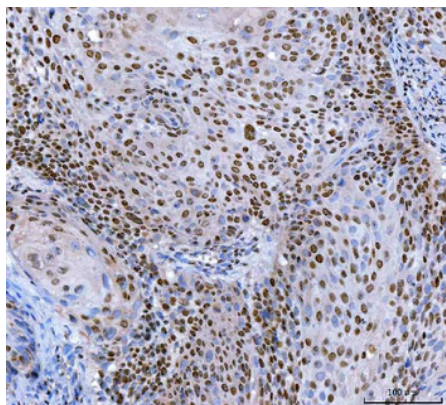
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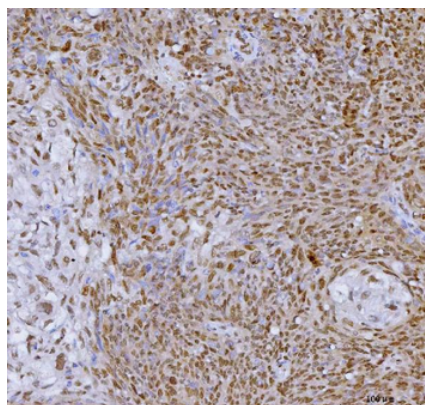
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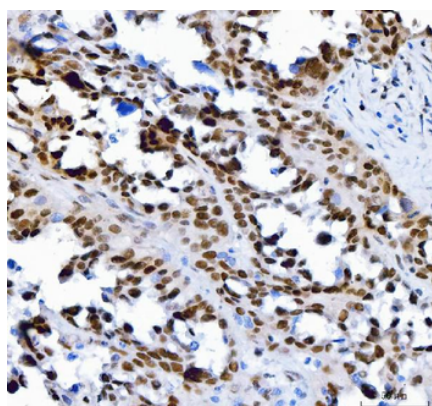
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IHC analysis of SP1 using anti-SP1 antibody. SP1 was detected in a paraffin-embedded section of human laryngeal squamous cell carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μg/ml mouse anti-SP1 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.



IHC analysis of SP1 using anti-SP1 antibody. SP1 was detected in a 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 2 μg/ml mouse anti-SP1 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.



IHC analysis of SP1 using anti-SP1 antibody. SP1 was detected in a paraffin-embedded section of human ovarian 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 2 μg/ml mouse anti-SP1 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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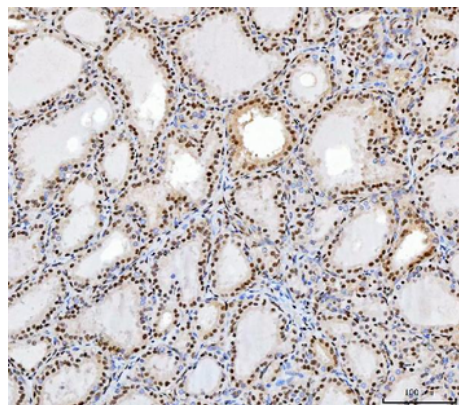
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IHC analysis of SP1 using anti-SP1 antibody. SP1 was detected in a paraffin-embedded section of human thyroid cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-SP1 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.



IHC analysis of SP1 using anti-SP1 antibody. SP1 was detected in a paraffin-embedded section of human tonsil 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-SP1 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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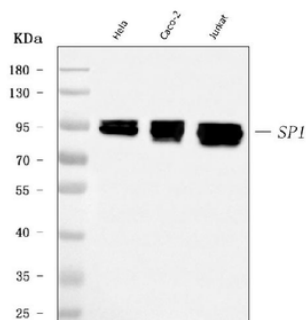
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Western blot analysis of SP1 using anti-SP1 antibody.

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 Hela whole cell lysates, Lane 2: human Caco-2 whole cell lysates, Lane 3: human Jurkat 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-SP1 antigen affinity purified monoclonal antibody at 0.5  $\mu$ g/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 SP1 at approximately 90 kDa. The expected band size for SP1 is at 81 kDa.

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