

## Product Datasheet

### SP1 Rabbit Polyclonal Antibody (orb443221)

<b>Catalog Number</b>	orb443221
<b>Category</b>	Antibodies
<b>Description</b>	Anti-SP1 Antibody. Tested in ELISA, Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
<b>Target</b>	Transcription factor Sp1
<b>Clonality</b>	Polyclonal
<b>Species/Host</b>	Rabbit
<b>Isotype</b>	Rabbit IgG
<b>Conjugation</b>	Unconjugated
<b>Reactivity</b>	Human, Mouse, Rat
<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>	Add 0.2ml of distilled water will yield a concentration of 500ug/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

**Biorbyt Ltd.**

7 Signet Court, Swann Road  
Cambridge  
CB5 8LA  
United Kingdom

Email: [info@biorbyt.com](mailto:info@biorbyt.com), [support@biorbyt.com](mailto:support@biorbyt.com)

Phone: [+44 \(0\)1223 859353](tel:+44(0)1223859353) | Fax: [+1 \(415\) 651-8558](tel:+1(415)651-8558)

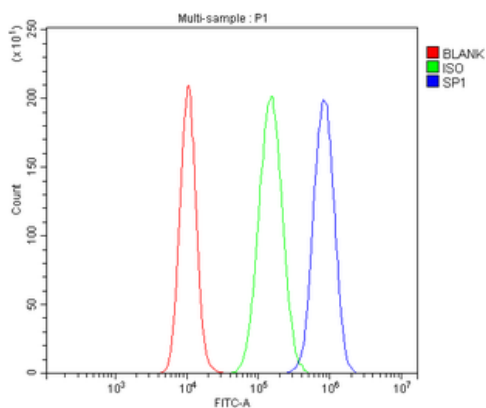
**Biorbyt LLC**

68 TW Alexander Drive  
Research Triangle Park  
Durham  
NC 27713  
United States

Email: [info@biorbyt.com](mailto:info@biorbyt.com), [support@biorbyt.com](mailto:support@biorbyt.com)

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<b>Tested applications</b>	ELISA, FC, ICC, IF, IHC, WB
<b>Dilution range</b>	Western blot, 0.1-0.5µg/ml Immunohistochemistry (Paraffin-embedded Section), 2-5µg/ml Immunocytochemistry/Immunofluorescence, 5µg/ml Immunofluorescence, 5µg/ml Flow Cytometry(Fixed), 1-3 µg/1x10 <sup>6</sup> cells ELISA, 0.1-0.5µg/ml
<b>Specificity</b>	No cross reactivity with other proteins.
<b>Cross Reactivity</b>	No cross-reactivity with other proteins.
<b>Antibody Type</b>	Primary Antibody
<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 rabbit anti-SP1 Antibody (1 µg/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit 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 rabbit 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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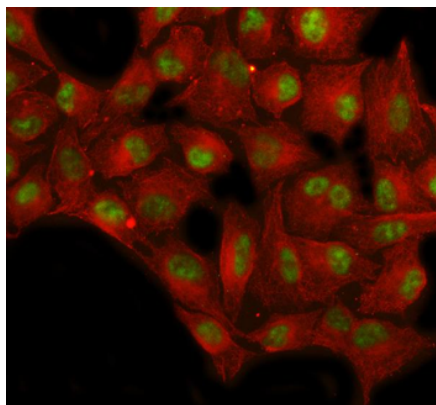
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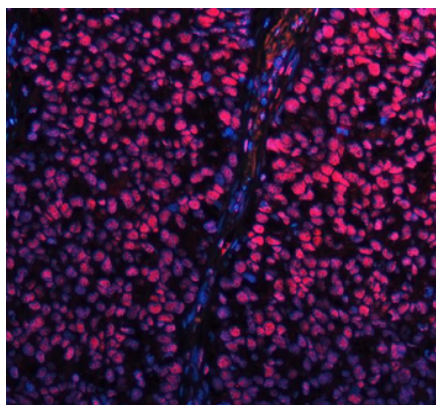
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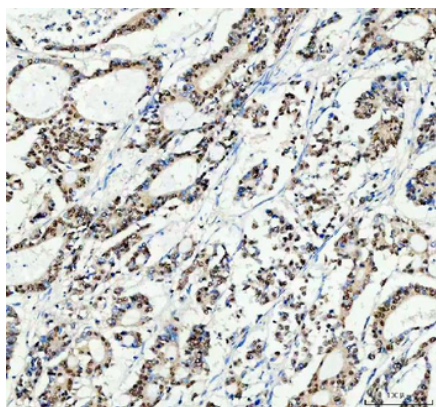
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IF analysis of SP1 using anti-SP1 antibody and anti-Beta Tubulin antibody. SP1 was detected in immunocytochemical section of A549 cell. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5  $\mu\text{g}/\text{mL}$  rabbit anti-SP1 Antibody and mouse anti-Beta Tubulin antibody overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG and DyLight®594 Conjugated Goat Anti-Mouse IgG were used as secondary antibody at 1:500 dilution and incubated for 30 minutes at 37°C. Visualize using a fluorescence microscope and filter sets appropriate for the label used.



IF analysis of SP1 using anti-SP1 antibody. SP1 was detected in a paraffin-embedded section of human non-small cell lung 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 5  $\mu\text{g}/\text{mL}$  rabbit anti-SP1 Antibody overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG was used as secondary antibody at 1:500 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 1  $\mu\text{g}/\text{mL}$  rabbit anti-SP1 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.

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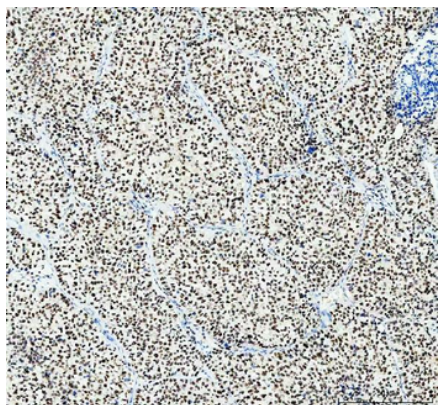
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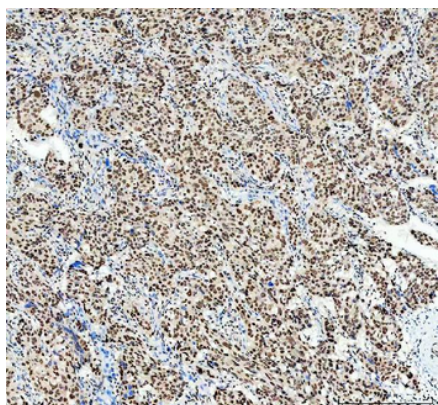
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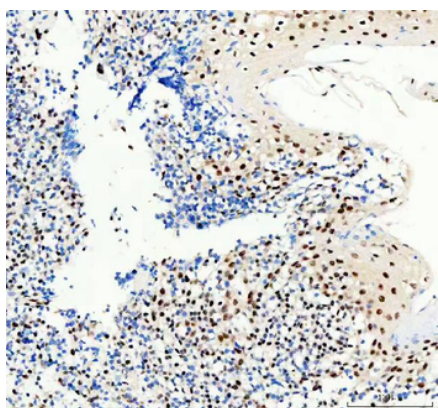
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IHC analysis of SP1 using anti-SP1 antibody. SP1 was detected in a paraffin-embedded section of human non-small cell lung carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1  $\mu$ g/ml rabbit anti-SP1 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit 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 1  $\mu$ g/ml rabbit anti-SP1 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit 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 tonsillitis 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  $\mu$ g/ml rabbit anti-SP1 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.

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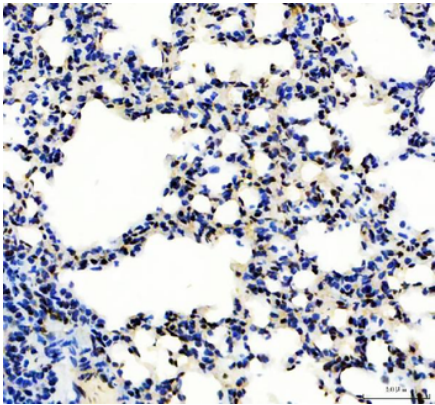
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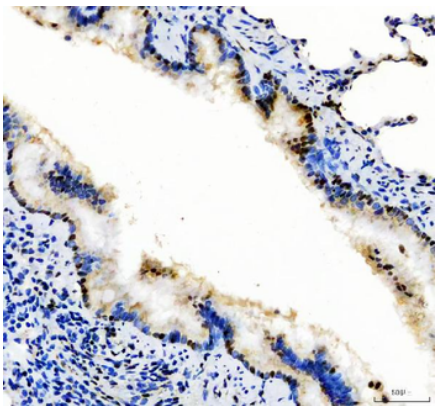
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IHC analysis of SP1 using anti-SP1 antibody. SP1 was detected in a paraffin-embedded section of mouse lung tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml rabbit anti-SP1 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit 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 rat lung 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 rabbit anti-SP1 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.

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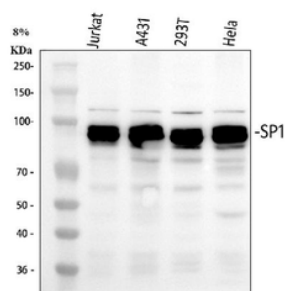
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Western blot analysis of SP1 using anti-SP1 antibody.

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 Jurkat whole cell

lysates, Lane 2: human A431 whole cell lysates, Lane 3: human

293T whole cell lysates, Lane 4: human HeLa 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 rabbit anti-SP1 antigen affinity

purified polyclonal antibody 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-rabbit IgG-HRP secondary antibody at a

dilution of 1:5000 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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