

Anti-SP1 Antibody Picoband™ (monoclonal, 3C4C3)

Catalog # ABO16623

Product Information

Application	WB, IHC, IF, ICC, FC
Primary Accession	P08047
Host	Mouse
Isotype	IgG1
Reactivity	Human
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-SP1 Antibody Picoband™ (monoclonal, 3C4C3) . Tested in Flow Cytometry, IF, ICC, IHC, WB applications. This antibody reacts with Human.
Reconstitution	Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.

Additional Information

Gene ID	6667
Other Names	Transcription factor Sp1, SP1, TSFP1
Calculated MW	80693
Application Details	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, 1-3 µg/1x10 ⁶ cells, Human
Source	Eukaryota
Contents	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na ₂ HPO ₄ .
Clone Names	Clone: 3C4C3
Immunogen	E. coli-derived human SP1 recombinant protein (Position: Q384-A603).
Purification	Immunogen affinity purified.
Storage	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

Name	SP1
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Synonyms

TSFP1

Function

Transcription factor that can activate or repress transcription in response to physiological and pathological stimuli. Binds with high affinity to GC-rich motifs and regulates the expression of a large number of genes involved in a variety of processes such as cell growth, apoptosis, differentiation and immune responses. Highly regulated by post-translational modifications (phosphorylations, sumoylation, proteolytic cleavage, glycosylation and acetylation). Also binds the PDGFR-alpha G-box promoter. May have a role in modulating the cellular response to DNA damage. Implicated in chromatin remodeling. Plays an essential role in the regulation of FE65 gene expression. In complex with ATF7IP, maintains telomerase activity in cancer cells by inducing TERT and TERC gene expression. Isoform 3 is a stronger activator of transcription than isoform 1. Positively regulates the transcription of the core clock component BMAL1 (PubMed:[10391891](#), PubMed:[11371615](#), PubMed:[11904305](#), PubMed:[14593115](#), PubMed:[16377629](#), PubMed:[16478997](#), PubMed:[16943418](#), PubMed:[17049555](#), PubMed:[18171990](#), PubMed:[18199680](#), PubMed:[18239466](#), PubMed:[18513490](#), PubMed:[18619531](#), PubMed:[19193796](#), PubMed:[20091743](#), PubMed:[21046154](#), PubMed:[21798247](#)). Plays a role in the recruitment of SMARCA4/BRG1 on the c-FOS promoter. Plays a role in protecting cells against oxidative stress following brain injury by regulating the expression of RNF112 (By similarity).

Cellular Location

Nucleus. Cytoplasm. Note=Nuclear location is governed by glycosylated/phosphorylated states. Insulin promotes nuclear location, while glucagon favors cytoplasmic location

Tissue Location

Up-regulated in adenocarcinomas of the stomach (at protein level). Isoform 3 is ubiquitously expressed at low levels

Background

Transcription factor Sp1, also known as specificity protein 1* is a protein that in humans is encoded by the SP1 gene. The protein encoded by this gene is a zinc finger transcription factor that binds to GC-rich motifs of many promoters. The encoded protein is involved in many cellular processes, including cell differentiation, cell growth, apoptosis, immune responses, response to DNA damage, and chromatin remodeling. Post-translational modifications such as phosphorylation, acetylation, glycosylation, and proteolytic processing significantly affect the activity of this protein, which can be an activator or a repressor. Three transcript variants encoding different isoforms have been found for this gene.

Images

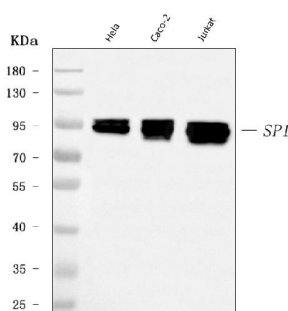


Figure 1. Western blot analysis of SP1 using anti-SP1 antibody (M00110-2).

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

(Catalog # M00110-2) 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 SP1 at approximately 90 kDa. The expected band size for SP1 is at 81 kDa.

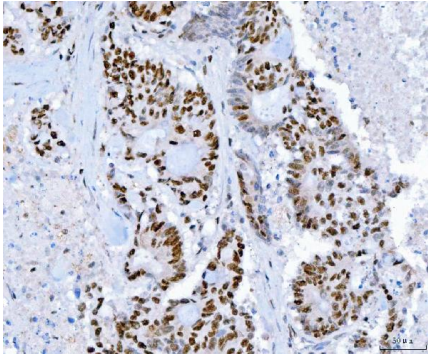


Figure 2. IHC analysis of SP1 using anti-SP1 antibody (M00110-2).

SP1 was detected in a paraffin-embedded section of human colorectal adenocarcinoma 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-SP1 Antibody (M00110-2) 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.

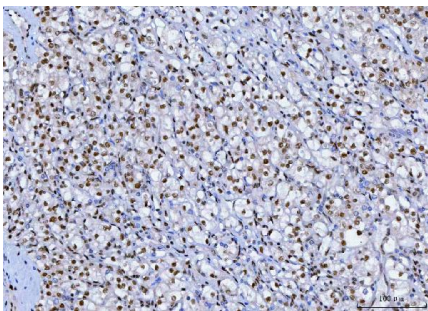


Figure 3. IHC analysis of SP1 using anti-SP1 antibody (M00110-2).

SP1 was detected in a paraffin-embedded section of human glioma 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-SP1 Antibody (M00110-2) 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.

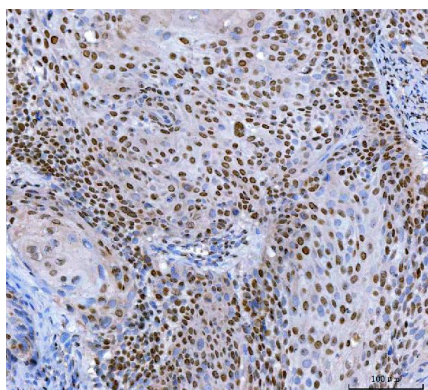
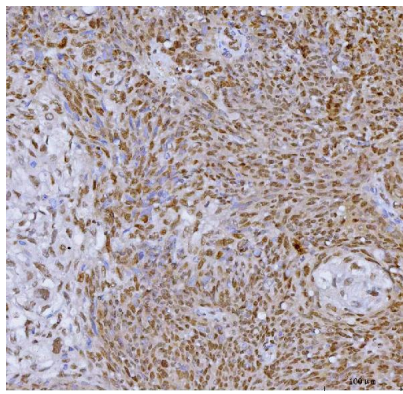


Figure 4. IHC analysis of SP1 using anti-SP1 antibody (M00110-2).

SP1 was detected in a paraffin-embedded section of human laryngeal squamous cell carcinoma 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-SP1 Antibody (M00110-2) 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 5. IHC analysis of SP1 using anti-SP1 antibody (M00110-2).

SP1 was detected in a paraffin-embedded section of human lung cancer 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 $\mu\text{g}/\text{ml}$ mouse anti-SP1 Antibody (M00110-2) 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.

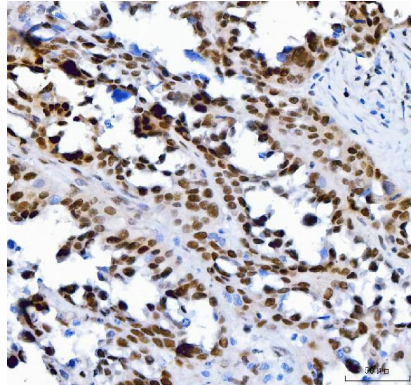


Figure 6. IHC analysis of SP1 using anti-SP1 antibody (M00110-2). SP1 was detected in a paraffin-embedded section of human ovarian cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 $\mu\text{g}/\text{ml}$ mouse anti-SP1 Antibody (M00110-2) 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.

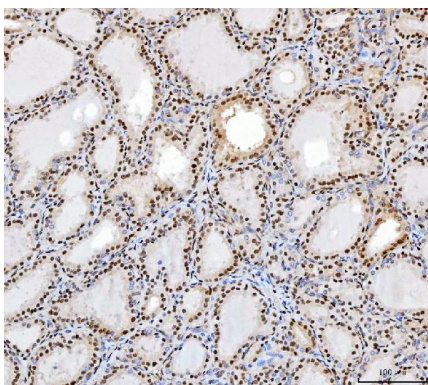


Figure 7. IHC analysis of SP1 using anti-SP1 antibody (M00110-2). SP1 was detected in a paraffin-embedded section of human thyroid cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 $\mu\text{g}/\text{ml}$ mouse anti-SP1 Antibody (M00110-2) 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.

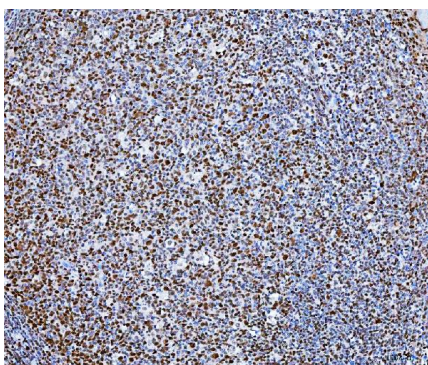
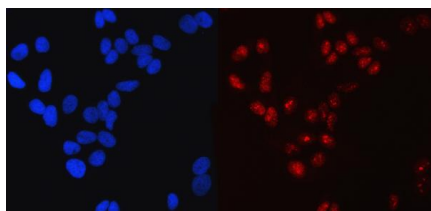


Figure 8. IHC analysis of SP1 using anti-SP1 antibody (M00110-2). SP1 was detected in a paraffin-embedded section of human tonsil tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 $\mu\text{g}/\text{ml}$ mouse anti-SP1 Antibody (M00110-2) 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 9. IF analysis of SP1 using anti-SP1 antibody (M00110-2). SP1 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 $\mu\text{g/mL}$ mouse anti-SP1 Antibody (M00110-2) overnight at 4°C. DyLight®550 Conjugated Goat Anti-Mouse IgG (BA1133) 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.

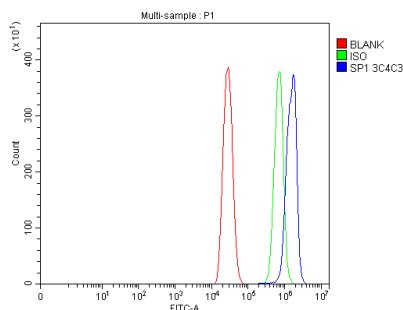


Figure 10. Flow Cytometry analysis of A431 cells using anti-SP1 antibody (M00110-2). Overlay histogram showing A431 cells stained with M00110-2 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-SP1 Antibody (M00110-2, 1 $\mu\text{g}/1 \times 10^6$ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 $\mu\text{g}/1 \times 10^6$ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 $\mu\text{g}/1 \times 10^6$) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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