

Anti-PC4/SUB1 Antibody Picoband™ (monoclonal, 8D9D1)

Catalog # ABO16597

Product Information

Application	WB, IHC, IF, FC
Primary Accession	P53999
Host	Mouse
Isotype	Mouse IgG2b
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-PC4/SUB1 Antibody Picoband™ (monoclonal, 8D9D1) . Tested in Flow Cytometry, IF, IHC, WB applications. This antibody reacts with Human, Mouse, Rat.
Reconstitution	Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.

Additional Information

Gene ID	10923
Other Names	Activated RNA polymerase II transcriptional coactivator p15, Positive cofactor 4, PC4, SUB1 homolog, p14, SUB1, PC4, RPO2TC1
Calculated MW	14395
Application Details	Western blot, 0.25-0.5 µg/ml, Human, Mouse, Rat Immunohistochemistry(Paraffin-embedded Section), 2-5 µg/ml, Human, Mouse, Rat 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: 8D9D1
Immunogen	E.coli-derived human PC4/SUB1 recombinant protein (Position: N62-L127).
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	SUB1
Synonyms	PC4, RPO2TC1
Function	General coactivator that functions cooperatively with TAFs and mediates functional interactions between upstream activators and the general transcriptional machinery. May be involved in stabilizing the multiprotein transcription complex. Binds single-stranded DNA. Also binds, in vitro, non-specifically to double-stranded DNA (ds DNA).
Cellular Location	Nucleus.

Background

Activated RNA polymerase II transcriptional coactivator p15, also known as positive cofactor 4 (PC4) or SUB1 homolog, is a protein that in humans is encoded by the SUB1 gene. This gene is mapped to 5p13.3. The transcriptional cofactor PC4 is an ancient single-strand DNA (ssDNA)-binding protein that has a homologue in bacteriophage T5 where it is likely the elusive replicative ssDNA-binding protein. The recombinant PC4 is shown to function identically to the native protein through its interaction with TAFs.

Images

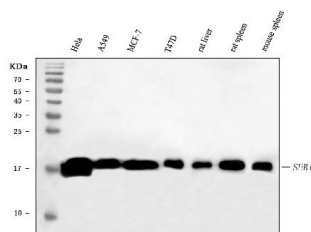


Figure 1. Western blot analysis of PC4/SUB1 using anti-PC4/SUB1 antibody (M02698-3).

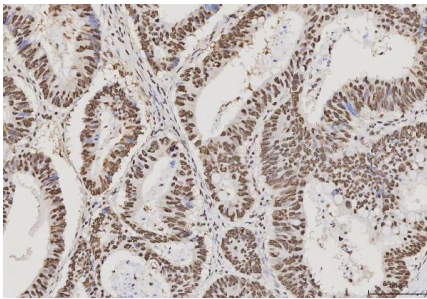
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

Lane 1: human Hela whole cell lysates,
Lane 2: human A549 whole cell lysates,
Lane 3: human MCF-7 whole cell lysates,
Lane 4: human T47D whole cell lysates,
Lane 5: rat liver tissue lysates,
Lane 6: rat spleen tissue lysates,
Lane 7: mouse spleen tissue 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-PC4/SUB1 antigen affinity purified monoclonal antibody (Catalog # M02698-3) 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 PC4/SUB1 at approximately 19 kDa. The expected band size for PC4/SUB1 is at 14 kDa.

Figure 2. IHC analysis of PC4/SUB1 using anti-PC4/SUB1 antibody (M02698-3).

PC4/SUB1 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 μ g/ml mouse anti-PC4/SUB1 Antibody (M02698-3) 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.

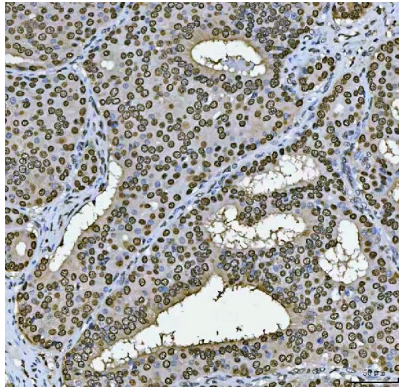


Figure 3. IHC analysis of PC4/SUB1 using anti-PC4/SUB1 antibody (M02698-3). PC4/SUB1 was detected in a paraffin-embedded section of human breast cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-PC4/SUB1 Antibody (M02698-3) 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.

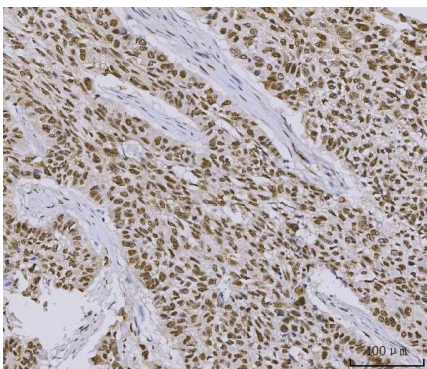


Figure 4. IHC analysis of PC4/SUB1 using anti-PC4/SUB1 antibody (M02698-3). PC4/SUB1 was detected in a paraffin-embedded section of human laryngeal squamous cell carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-PC4/SUB1 Antibody (M02698-3) 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.

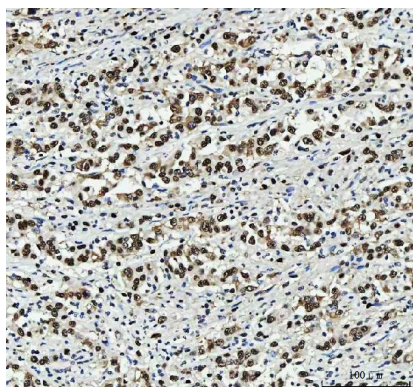
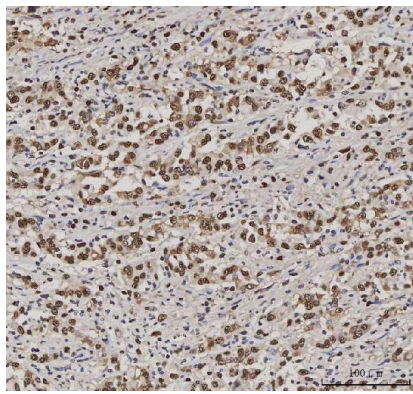


Figure 5. IHC analysis of PC4/SUB1 using anti-PC4/SUB1 antibody (M02698-3). PC4/SUB1 was detected in a paraffin-embedded section of human hepatocellular carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-PC4/SUB1 Antibody (M02698-3) 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 PC4/SUB1 using anti-PC4/SUB1 antibody (M02698-3). PC4/SUB1 was detected in a paraffin-embedded section



of human hepatocellular 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-PC4/SUB1 Antibody (M02698-3) 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.

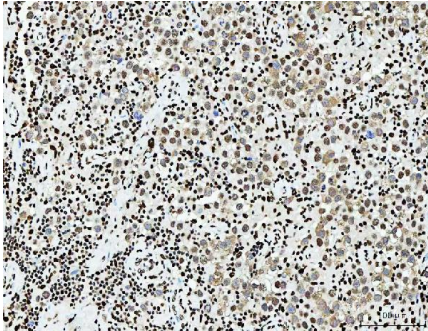


Figure 7. IHC analysis of PC4/SUB1 using anti-PC4/SUB1 antibody (M02698-3). PC4/SUB1 was detected in a paraffin-embedded section of human testicular germ cell tumor tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-PC4/SUB1 Antibody (M02698-3) 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.

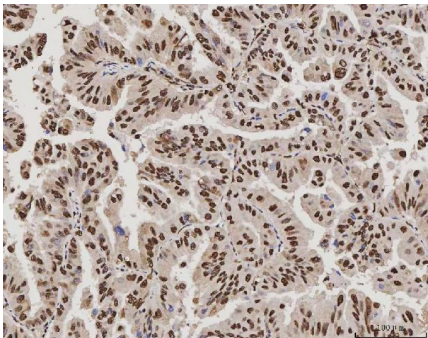


Figure 8. IHC analysis of PC4/SUB1 using anti-PC4/SUB1 antibody (M02698-3). PC4/SUB1 was detected in a paraffin-embedded section of human lung adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-PC4/SUB1 Antibody (M02698-3) 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.

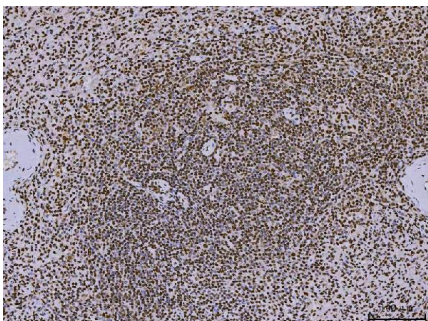
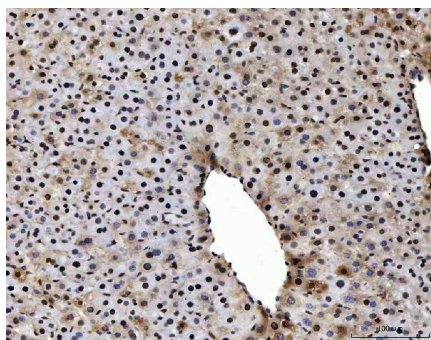


Figure 9. IHC analysis of PC4/SUB1 using anti-PC4/SUB1 antibody (M02698-3). PC4/SUB1 was detected in a paraffin-embedded section of human spleen tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-PC4/SUB1 Antibody (M02698-3) 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 10. IHC analysis of PC4/SUB1 using anti-PC4/SUB1 antibody (M02698-3). PC4/SUB1 was detected in a paraffin-embedded section of mouse liver 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-PC4/SUB1 Antibody (M02698-3) 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.

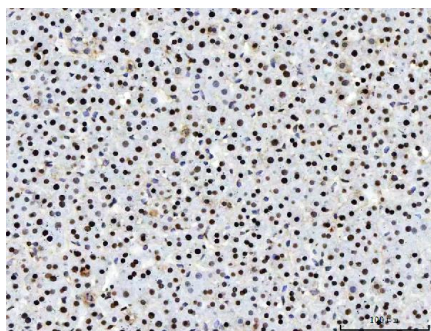


Figure 11. IHC analysis of PC4/SUB1 using anti-PC4/SUB1 antibody (M02698-3). PC4/SUB1 was detected in a paraffin-embedded section of rat liver tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-PC4/SUB1 Antibody (M02698-3) 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.

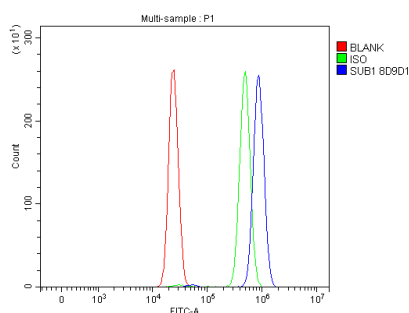


Figure 12. Flow Cytometry analysis of HepG2 cells using anti-PC4/SUB1 antibody (M02698-3). Overlay histogram showing HepG2 cells stained with M02698-3 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-PC4/SUB1 Antibody (M02698-3, 1 µg/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

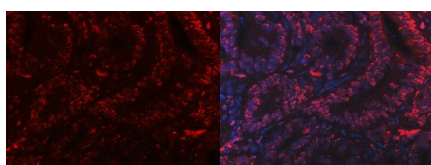


Figure 13. IF analysis of PC4/SUB1 using anti-PC4/SUB1 antibody (M02698-3). PC4/SUB1 was detected in a paraffin-embedded section of human intestine cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 5 µg/mL mouse anti-PC4/SUB1 Antibody (M02698-3) overnight at 4°C. Cy3 Conjugated Goat Anti-Mouse IgG (BA1031) 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.