

# Anti-MCM7 Antibody Picoband<sup>™</sup> (monoclonal, 3H11)

Catalog # ABO16570

Specification

## Anti-MCM7 Antibody Picoband<sup>™</sup> (monoclonal, 3H11) - Product Information

Application Primary Accession Host Isotype Reactivity Clonality Format Description WB, IHC, IF, ICC, FC <u>P33993</u> Mouse Mouse IgG1 Rat, Human Monoclonal Lyophilized

Anti-MCM7 Antibody Picoband<sup>™</sup> (monoclonal, 3H11) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Rat.

Reconstitution

Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.

### Anti-MCM7 Antibody Picoband<sup>™</sup> (monoclonal, 3H11) - Additional Information

Gene ID 4176

**Other Names** DNA replication licensing factor MCM7, 3.6.4.12, CDC47 homolog, P1.1-MCM3, MCM7 (<a href="http://www.genenames.org/cgi-bin/gene\_symbol\_report?hgnc\_id=6950" target="\_blank">HGNC:6950</a>), CDC47, MCM2

Calculated MW 81 kDa KDa

**Application Details** Western blot, 0.25-0.5 μg/ml, Human, Rat<br> Immunohistochemistry(Paraffin-embedded Section), 2-5 μg/ml, Human<br> Immunocytochemistry/Immunofluorescence, 5 μg/ml, Human<br> Flow Cytometry, 1-3 μg/1x10^6 cells, Human<br>

**Contents** Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na2HPO4.

Immunogen E.coli-derived human MCM7 recombinant protein (Position: D526-V719). Human MCM7 shares 94% amino acid (aa) sequence identity with MCM7.

**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.

## Anti-MCM7 Antibody Picoband<sup>™</sup> (monoclonal, 3H11) - Protein Information

Name MCM7 (<u>HGNC:6950</u>)

Synonyms CDC47, MCM2

Function

Acts as a component of the MCM2-7 complex (MCM complex) which is the replicative helicase essential for 'once per cell cycle' DNA replication initiation and elongation in eukaryotic cells. Core component of CDC45-MCM-GINS (CMG) helicase, the molecular machine that unwinds template DNA during replication, and around which the replisome is built (PubMed:<a href="http://www.uniprot.org/citations/25661590" target="\_blank">25661590</a>, PubMed:<a href="http://www.uniprot.org/citations/32453425" target=" blank">32453425</a>, PubMed:<a href="http://www.uniprot.org/citations/34694004" target=" blank">34694004</a>, PubMed:<a href="http://www.uniprot.org/citations/34700328" target=" blank">34700328</a>, PubMed:<a href="http://www.uniprot.org/citations/35585232" target=" blank">35585232</a>, PubMed:<a href="http://www.uniprot.org/citations/9305914" target=" blank">9305914</a>). The active ATPase sites in the MCM2-7 ring are formed through the interaction surfaces of two neighboring subunits such that a critical structure of a conserved arginine finger motif is provided in trans relative to the ATP-binding site of the Walker A box of the adjacent subunit. The six ATPase active sites, however, are likely to contribute differentially to the complex helicase activity (PubMed:<a href="http://www.uniprot.org/citations/32453425" target=" blank">32453425</a>). Required for S-phase checkpoint activation upon UV-induced damage.

#### **Cellular Location**

Nucleus. Chromosome. Note=Associated with chromatin before the formation of nuclei and detaches from it as DNA replication progresses.

#### Anti-MCM7 Antibody Picoband<sup>™</sup> (monoclonal, 3H11) - Protocols

Provided below are standard protocols that you may find useful for product applications.

- <u>Western Blot</u>
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- <u>Cell Culture</u>

Anti-MCM7 Antibody Picoband<sup>™</sup> (monoclonal, 3H11) - Images



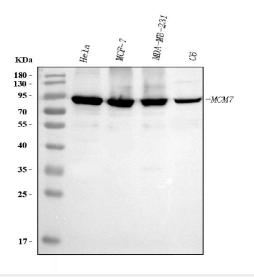


Figure 1. Western blot analysis of MCM7 using anti-MCM7 antibody (M01649-3).

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 MCF-7 whole cell lysates,

Lane 3: human MDA-MB-231 whole cell lysates,

Lane 4: rat C6 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-MCM7 antigen affinity purified monoclonal antibody (Catalog # M01649-3) 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 (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for MCM7 at approximately 81 kDa. The expected band size for MCM7 is at 81 kDa.

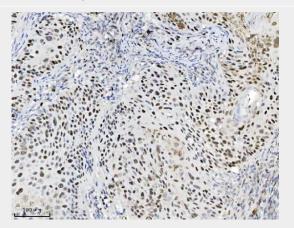


Figure 3. IHC analysis of MCM7 using anti-MCM7 antibody (M01649-3).

MCM7 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  $\mu$ g/ml mouse anti-MCM7 Antibody (M01649-3) 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.



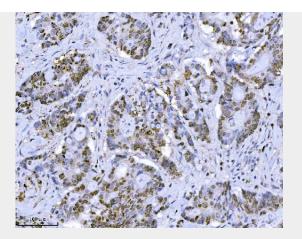


Figure 4. IHC analysis of MCM7 using anti-MCM7 antibody (M01649-3).

MCM7 was detected in a paraffin-embedded section of human adenocarcinoma of the colon 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  $\mu$ g/ml mouse anti-MCM7 Antibody (M01649-3) 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.

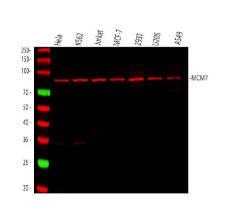


Figure 2. Western blot analysis of MCM7 using anti-MCM7 antibody (M01649-3).

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 K562 whole cell lysates,

Lane 3: human Jurkat whole cell lysates,

Lane 4: human MCF-7 whole cell lysates,

Lane 5: human 293T whole cell lysates,

Lane 6: human U2OS whole cell lysates,

Lane 7: human A549 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-MCM7 antigen affinity purified monoclonal antibody (Catalog # M01649-3) 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-DyLight 647 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 MCM7 at approximately 81 kDa. The expected band size for MCM7 is at 81 kDa.

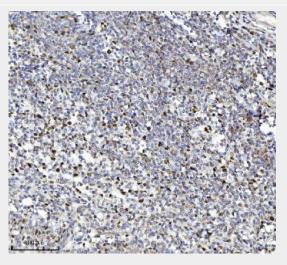


Figure 5. IHC analysis of MCM7 using anti-MCM7 antibody (M01649-3).

MCM7 was detected in a paraffin-embedded section of human chronic tonsillitis 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  $\mu$ g/ml mouse anti-MCM7 Antibody (M01649-3) 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.

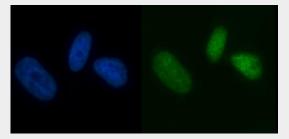


Figure 6. IF analysis of MCM7 using anti-MCM7 antibody (M01649-3).

MCM7 was detected in an immunocytochemical section of PC-3 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  $\mu$ g/mL mouse anti-MCM7 Antibody (M01649-3) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Mouse IgG (BA1126) 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.

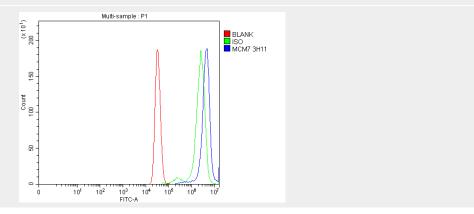




Figure 7. Flow Cytometry analysis of MCF-7 cells using anti-MCM7 antibody (M01649-3). Overlay histogram showing MCF-7 cells stained with M01649-3 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-MCM7 Antibody (M01649-3, 1  $\mu$ g/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10  $\mu$ g/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  $\mu$ g/1x10<sup>6</sup>) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

### Anti-MCM7 Antibody Picoband<sup>™</sup> (monoclonal, 3H11) - Background

MCM7(Minichromosome Maintenance, s. Cerevisiae, homolog of, 7), also called CDC47, FORMERLY, is one of the highly conserved mini-chromosome maintenance proteins (MCM) that are essential for the initiation of eukaryotic genome replication. The MCM7 gene is mapped to 7q22.1. MCM7 plays a pivotal role in the G1/S phase transition, orchestrating the correct assembly of replication forks on chromosomal DNA and ensuring that all the genome is replicated once and not more than once at each cell cycle. The MCM7 gene contains 15 exons. The miRNAs MIR106B, MIR93, and MIR25 are clustered in a 5-prime to 3-prime orientation within intron 13. It has been found that MCM7 and the precursors of microRNAs (miRNAs) MIR106B, MIR93, and MIR25, all of which arise from intron 13 of the MCM7 gene, were overexpressed with almost perfect correlation in 5 of 10 human gastric tumors.