

Anti-MCM5 Picoband™ Antibody (monoclonal, 4G10)

Catalog # ABO15010

Product Information

Application	WB, IHC, IF, ICC, FC
Primary Accession	P33992
Host	Mouse
Isotype	Mouse IgG2a
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-MCM5 Picoband™ Antibody (monoclonal, 4G10) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500ug/ml.

Additional Information

Gene ID	4174
Other Names	DNA replication licensing factor MCM5 {ECO:0000303 Ref.3}, 3.6.4.12, CDC46 homolog, P1-CDC46, MCM5, CDC46
Calculated MW	82286
Application Details	Western blot, 0.25-0.5 µg/ml, Human, Mouse, Rat 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 4mg Trehalose, 0.9mg NaCl and 0.2mg Na2HPO4.
Clone Names	Clone: 4G10
Immunogen	E.coli-derived human MCM5 recombinant protein (Position: K206-H723).
Purification	Immunogen affinity purified.
Storage	Store 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 freeze-thaw cycles.

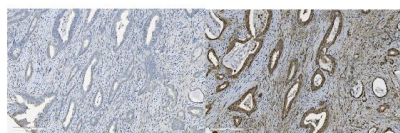
Protein Information

Name	MCM5
Synonyms	CDC46
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 (PubMed: 40940420). 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: 16899510 , PubMed: 32453425 , PubMed: 34694004 , PubMed: 34700328 , PubMed: 35585232). 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: 32453425).
Cellular Location	Nucleus. Chromosome Note=Associated with chromatin before the formation of nuclei and detaches from it as DNA replication progresses

Background

DNA replication licensing factor MCM5 is a protein that in humans is encoded by the MCM5 gene. It is mapped to 22q12.3. The protein encoded by this gene is structurally very similar to the CDC46 protein from *S. cerevisiae*, a protein involved in the initiation of DNA replication. The encoded protein is a member of the MCM family of chromatin-binding proteins and can interact with at least two other members of this family. The encoded protein is upregulated in the transition from the G0 to G1/S phase of the cell cycle and may actively participate in cell cycle regulation.

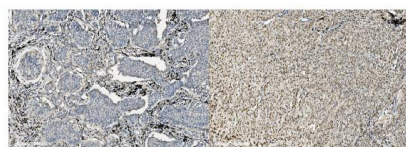
Images



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Figure 2. IHC analysis of MCM5 using anti-MCM5 antibody (M03642-2).

MCM5 was detected in paraffin-embedded section of human gallbladder 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-MCM5 Antibody (M03642-2) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.



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Figure 3. IHC analysis of MCM5 using anti-MCM5 antibody (M03642-2).

MCM5 was detected in 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-MCM5 Antibody (M03642-2) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021)

with DAB as the chromogen.

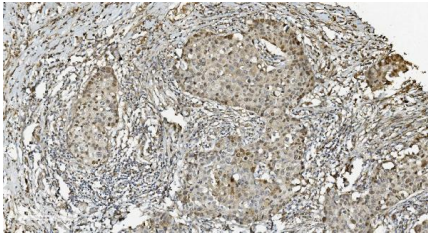


Figure 4. IHC analysis of MCM5 using anti-MCM5 antibody (M03642-2).

MCM5 was detected in paraffin-embedded section of human pancreatic 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-MCM5 Antibody (M03642-2) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

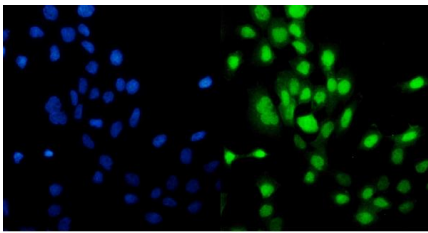


Figure 5. IF analysis of MCM5 using anti-MCM5 antibody (M03642-2).

MCM5 was detected in immunocytochemical section of A431 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 µg/mL mouse anti-MCM5 Antibody (M03642-2) 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.

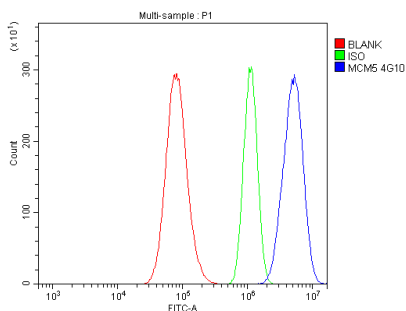


Figure 6. Flow Cytometry analysis of A549 cells using anti-MCM5 antibody (M03642-2).

Overlay histogram showing A549 cells stained with M03642-2 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-MCM5 Antibody (M03642-2, 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.

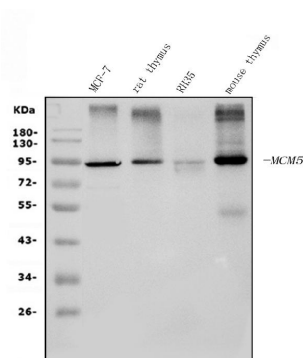


Figure 1. Western blot analysis of MCM5 using anti-MCM5 antibody (M03642-2).

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 50ug of sample under reducing conditions.

Lane 1: human MCF-7 whole cell lysates,
Lane 2: rat thymus tissue lysates,
Lane 3: rat RH-35 whole cell lysates,
Lane 4: mouse thymus tissue lysates.

After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA 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- MCM5 antigen affinity purified monoclonal antibody (Catalog # M03642-2) 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 MCM5 at approximately 95KD. The expected band size for MCM5 is at 95KD.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.