

Anti-MCM6 Antibody Picoband™ (monoclonal, 3I4C8)

Catalog # ABO15109

Product Information

Application	WB, IHC, IF, ICC, FC
Primary Accession	Q14566
Host	Mouse
Isotype	Mouse IgG2b
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-MCM6 Antibody Picoband™ (monoclonal, 3I4C8) . Tested in Flow Cytometry, IF, IHC, ICC, 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	4175
Other Names	DNA replication licensing factor MCM6, 3.6.4.12, p105MCM, MCM6 (HGNC:6949)
Calculated MW	92889
Application Details	Western blot, 0.25-0.5 µg/ml, Human, Mouse, Rat Immunohistochemistry(Paraffin-embedded Section), 2-5 µg/ml, Human, Mouse, Rat 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: 3I4C8
Immunogen	E.coli-derived human MCM6 recombinant protein (Position: Q14-D821).
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	MCM6 (HGNC:6949)
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: 16899510 , PubMed: 32453425 , PubMed: 34694004 , PubMed: 34700328 , PubMed: 35585232 , PubMed: 9305914). 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=Binds to chromatin during G1 and detaches from it during S phase.

Background

MCM6(Minichromosome maintenance, s. pombe, homolog of, 6) is a protein that in humans is encoded by the MCM6 gene. MCM6 is one of the highly conserved mini-chromosome maintenance proteins (MCM) that are essential for the initiation of eukaryotic genome replication. The MCM genes were originally identified in yeast defective in minichromosome maintenance and have since been shown to play roles in the progression of the cell cycle; many are cell division control genes. The MCM6 gene is mapped on 2q21.3. Mcm 6 has recently been shown to interact strongly Cdt1 at defined residues, by mutating these target residues Wei et al. observed lack of Cdt1 recruitment of Mcm2-7 to the pre-RC. An approximately 200-kb region surrounding the C/T(-13910) polymorphism in MCM6 intron 13 functioned as an enhancer of the lactase gene promoter in intestinal cell culture.

Images

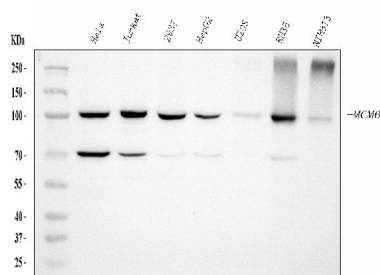


Figure 1. Western blot analysis of MCM6 using anti-MCM6 antibody (M02755-1).

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 Jurkat whole cell lysates,
Lane 3: human 293T whole cell lysates,
Lane 4: human HepG2 whole cell lysates,
Lane 5: human U2OS whole cell lysates,
Lane 6: rat RH35 whole cell lysates,
Lane 7: mouse NIH/3T3 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-MCM6 antigen affinity purified monoclonal antibody (Catalog # M02755-1) 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 MCM6 at

approximately 105 kDa. The expected band size for MCM6 is at 93 kDa.

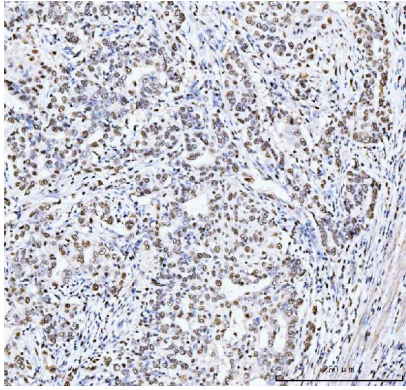


Figure 2. IHC analysis of MCM6 using anti-MCM6 antibody (M02755-1).

MCM6 was detected in a paraffin-embedded section of human metaplasia of squamous cells of the renal pelvis 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-MCM6 Antibody (M02755-1) 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.

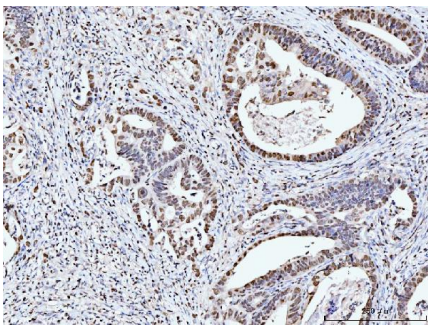


Figure 3. IHC analysis of MCM6 using anti-MCM6 antibody (M02755-1).

MCM6 was detected in a paraffin-embedded section of human rectal moderately differentiated 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-MCM6 Antibody (M02755-1) 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.

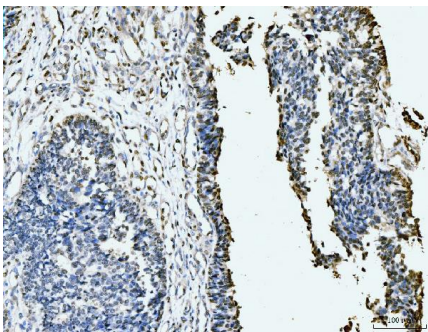


Figure 4. IHC analysis of MCM6 using anti-MCM6 antibody (M02755-1).

MCM6 was detected in a paraffin-embedded section of human bladder epithelial 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-MCM6 Antibody (M02755-1) 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.

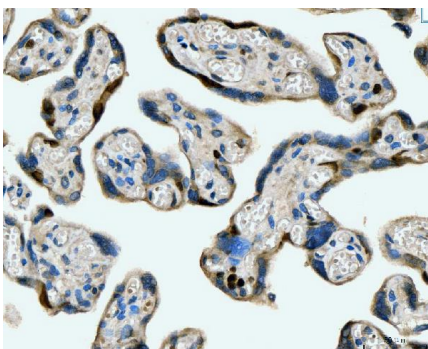


Figure 5. IHC analysis of MCM6 using anti-MCM6 antibody (M02755-1).

MCM6 was detected in a paraffin-embedded section of human placenta 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-MCM6 Antibody (M02755-1) 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.

with DAB as the chromogen.

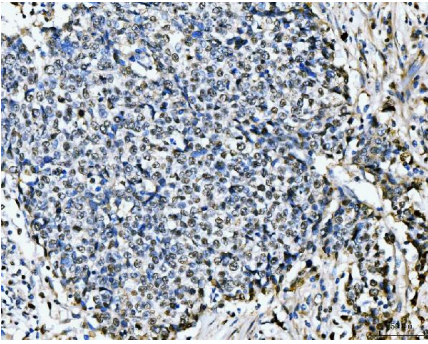


Figure 6. IHC analysis of MCM6 using anti-MCM6 antibody (M02755-1).

MCM6 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-MCM6 Antibody (M02755-1) 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.

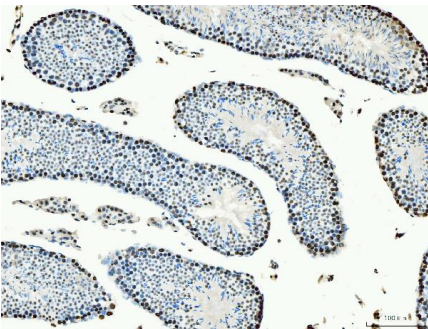


Figure 7. IHC analysis of MCM6 using anti-MCM6 antibody (M02755-1).

MCM6 was detected in a paraffin-embedded section of mouse testis 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-MCM6 Antibody (M02755-1) 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.

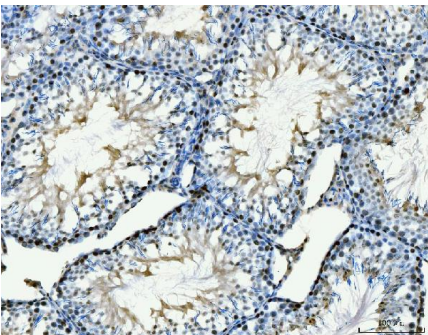


Figure 8. IHC analysis of MCM6 using anti-MCM6 antibody (M02755-1).

MCM6 was detected in a paraffin-embedded section of rat testis 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-MCM6 Antibody (M02755-1) 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.

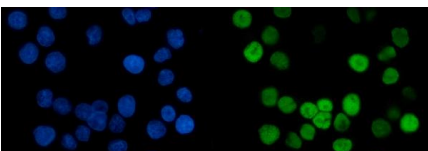


Figure 9. IF analysis of MCM6 using anti-MCM6 antibody (M02755-1).

MCM6 was detected in an immunocytochemical section of CACO-2 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 $\mu\text{g}/\text{mL}$ mouse anti-MCM6 Antibody (M02755-1) 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.

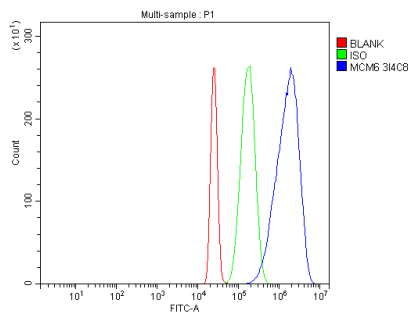


Figure 10. Flow Cytometry analysis of HL-60 cells using anti-MCM6 antibody (M02755-1).

Overlay histogram showing HL-60 cells stained with M02755-1 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-MCM6 Antibody (M02755-1, 1 μ g/ 1×10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 μ g/ 1×10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 μ g/ 1×10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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