10320 Camino Santa Fe, Suite G
San Diego, CA 92121
Tel: 858.875.1900 Fax: 858.875.1999

Anti-DDX4/MVH Antibody Picoband™ (monoclonal, 3C3)

Catalog # ABO15098

Product Information

Application	WB, IHC, IF
Primary Accession	Q9NQI0
Host	Mouse
Isotype	Mouse IgG2b
Reactivity	Rat, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-DDX4/MVH Antibody Picoband™ (monoclonal, 3C3) . Tested in IF, IHC, WB applications. This antibody reacts with Mouse, Rat.
Reconstitution	Adding 0.2 ml of distilled water will yield a concentration of 500 □g/ml.

Additional Information

Gene ID	54514
Other Names	Probable ATP-dependent RNA helicase DDX4, 3.6.4.13, DEAD box protein 4, Vasa homolog, DDX4, VASA
Calculated MW	79308
Application Details	Western blot, 0.25-0.5 □g/ml, Mouse, Rat Immunohistochemistry(Paraffin-embedded Section), 2-5 □g/ml, Mouse, Rat Immunofluorescence, 5 □g/ml, Mouse, Rat
Source	Eukaryota
Contents	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na2HPO4.
Clone Names	Clone: 3C3
Immunogen	E.coli-derived human DDX4/MVH recombinant protein (Position: D3-D666).
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	DDX4

Synonyms	VASA
Function	ATP-dependent RNA helicase required during spermatogenesis (PubMed:10920202, PubMed:21034600). Required to repress transposable elements and preventing their mobilization, which is essential for the germline integrity (By similarity). Acts via the piRNA metabolic process, which mediates the repression of transposable elements during meiosis by forming complexes composed of piRNAs and Piwi proteins and governs the methylation and subsequent repression of transposons (By similarity). Involved in the secondary piRNAs metabolic process, the production of piRNAs in fetal male germ cells through a ping-pong amplification cycle (By similarity). Required for PIWIL2 slicing- triggered piRNA biogenesis: helicase activity enables utilization of one of the slice cleavage fragments generated by PIWIL2 and processing these pre-piRNAs into piRNAs (By similarity).
Cellular Location	Cytoplasm {ECO:0000250|UniProtKB:Q61496}. Cytoplasm, perinuclear region {ECO:0000250|UniProtKB:Q61496} Note=Component of the meiotic nuage, also named P granule, a germ-cell- specific organelle required to repress transposon activity during meiosis. {ECO:0000250|UniProtKB:Q61496}
Tissue Location	Expressed only in ovary and testis. Expressed in migratory primordial germ cells in the region of the gonadal ridge in both sexes.

Background

DDX4(DEAD/H BOX 4), also known as VASA. The deduced 724-amino acid VASA protein contains the 8 conserved domains found in all known DEAD box proteins. The amino acid sequence in this core region shows greater similarity to VASA homologs in other species than to other human DEAD box proteins. By radiation hybrid analysis, Castrillon et al.(2000) mapped the VASA gene to 5q. By fluorescence in situ hybridization, they refined the localization to 5q11.2-q12. This region is syntenic to the distal end of mouse chromosome 13, where the mouse VASA homolog(Ddx4) resides(Abe and Noce, 1997). Using a combination of proteomics, cytology, and functional analysis in C. elegans, Chu et al.(2006) reduced 1,099 proteins copurified with spermatogenic chromatin to 132 proteins for functional analysis.

Images

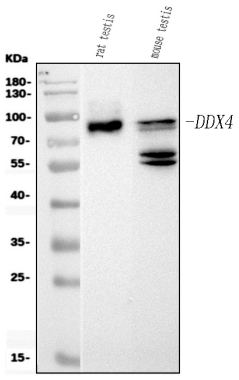


Figure 1. Western blot analysis of DDX4/MVH using anti-DDX4/MVH antibody (M02448-1).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.
Lane 1: rat testis tissue lysates,
Lane 2: mouse testis tissue lysate.
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-DDX4/MVH antigen affinity purified monoclonal antibody (Catalog # M02448-1) 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 DDX4/MVH at approximately 79 kDa. The expected band

size for DDX4/MVH is at 79 kDa.

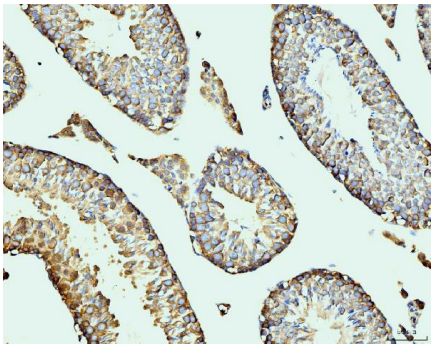


Figure 2. IHC analysis of DDX4/MVH using anti-DDX4/MVH antibody (M02448-1).
DDX4/MVH was detected in a paraffin-embedded section of mouse testis tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-DDX4/MVH Antibody (M02448-1) 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 Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

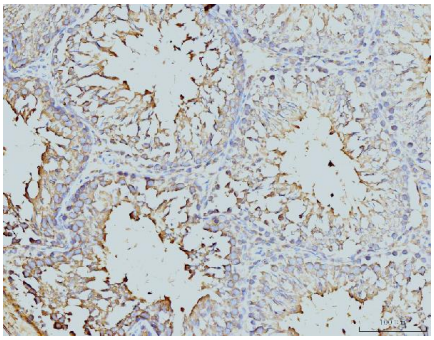


Figure 3. IHC analysis of DDX4/MVH using anti-DDX4/MVH antibody (M02448-1).
DDX4/MVH was detected in a paraffin-embedded section of rat testis tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-DDX4/MVH Antibody (M02448-1) 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 Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

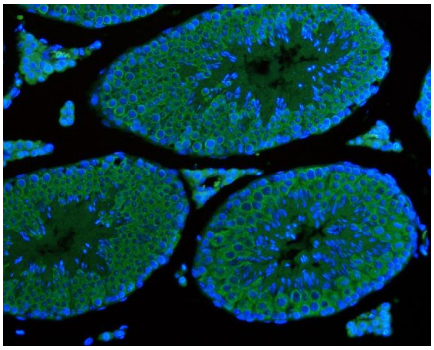


Figure 4. IF analysis of DDX4/MVH using anti-DDX4/MVH antibody (M02448-1).
DDX4/MVH was detected in a paraffin-embedded section of mouse testis tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 5 µg/mL mouse anti-DDX4/MVH Antibody (M02448-1) overnight at 4°C. Biotin conjugated goat anti-mouse IgG (BA1001) was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using DyLight®488 Conjugated Avidin (BA1128). The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

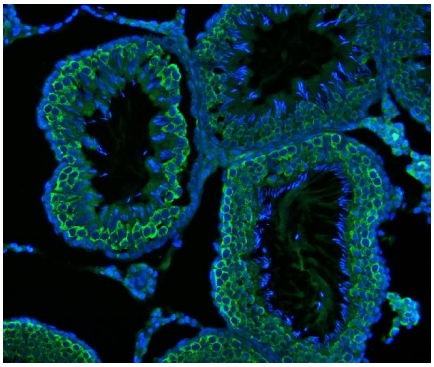


Figure 5. IF analysis of DDX4/MVH using anti-DDX4/MVH antibody (M02448-1).
DDX4/MVH was detected in a paraffin-embedded section of rat testis tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 5 µg/mL mouse anti-DDX4/MVH Antibody (M02448-1) overnight at 4°C. Biotin conjugated goat anti-mouse IgG (BA1001) was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using DyLight®488 Conjugated Avidin (BA1128). The section was counterstained with DAPI. Visualize using a

fluorescence microscope and filter sets appropriate for the label used.

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