

Product Information

Anti-MBD2a,b (RA-18)

Developed in Rabbit
Affinity Isolated Antibody

Product Number **M 7318**

Product Description

Anti-MBD2a,b (RA-18) is developed in rabbit using as immunogen a synthetic peptide corresponding to amino acids 394-411 of human MBD2a conjugated to KLH via an N-terminal added lysine residue. This sequence is conserved in MBD2a and MBD2b of human origin, and is not found in MBD3. The antibody is affinity-purified using the immunizing peptide immobilized on agarose.

Anti MBD2a,b (RA-18) recognizes MBD2a and MBD2b. Applications include immunoblotting (MBD2a, approx. 45 kDa; MBD2b, approx. 29 kDa) and immunofluorescence. In immunoblotting, the antibody also detects an unidentified band at approx. 43 kDa. Staining of the MBD2 a and b bands in immunoblotting is specifically inhibited by the immunizing peptide.

Chromatin, the physiological packaging structure of histone proteins and DNA, is a key element in the regulation of gene expression. Histones are subjected to post-translational modifications such as acetylation, phosphorylation, and methylation, and play a major role in the regulation of transcription.^{1,2} DNA methylation is the major modification of eukaryotic genomes, which occurs at the fifth position of cytosine in CpG dinucleotide sequences, and is associated with transcriptional repression.³⁻⁶ Silencing of transcription units have been found to occur in genes located on the inactive X-chromosome, genes silenced by genomic imprinting, and genes silenced in transformed cell lines and tumors.^{3,7-9} The DNA methylation system is composed of methyl-CpG-binding proteins, as well as of DNA cytosine methyl transferases.^{3,10} Five methyl-CpG binding proteins were isolated: MeCP2, MBD1, MBD2, MBD3, and MBD4.^{10,11} With the exceptions of MBD2 and MBD3, sequence similarity is limited to the methyl-CpG binding domains themselves (MBD). MBD2 consists of two isoforms, MBD2a and MBD2b, which are generated from a single gene; MBD2a is a 414 amino acids protein, whereas MBD2b lacks the 152 amino acids N-terminal extension.¹²

MBD2a is a component of the MeCP1 corepressor complex, which is a 400-800 kDa complex containing as components MBD2, Mi-2, MTA2, MBD3, and HDAC1/2.¹³ Although MBD2 is highly similar to MBD3 and both are components of the MeCP1 complex, there are functional differences between both proteins. MBD2, but not MBD3, binds to methylated DNA *in vitro* or *in vivo*, suggesting that MBD2 targets the MeCP1 complex to methylated DNA.¹²⁻¹⁴ Likewise MBD2 isoforms have different roles *in vivo*. Thus, MBD2a, but not MBD2b, interacts with RNA Helicase A (RHA).¹⁵ In connection to the ability of MBD2 to bind to methylated DNA, and its function as part of a corepressor complex, high levels of MBD2 protein appear to protect against some cancers.^{15,16}

Reagent

Anti-MBD2a,b (RA-18) is supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide.

Antibody Concentration: approx. 1.0 mg/ml

Precautions and Disclaimer

Due to the sodium azide content, a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazardous and safe handling practices.

Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For extended storage, freeze in working aliquots. Repeated freezing and thawing is not recommended. Storage in frost-free freezers is also not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilutions should be discarded if not used within 12 hours.

Product Profile

By immunoblotting, a working antibody concentration of 0.5-1.0 µg/ml is recommended using nuclear extracts of HeLa cells.

By indirect immunofluorescence, a working antibody concentration of 6-8 µg/ml is recommended using NIH-3T3 cells.

Recommendation: For immunoblotting, dilute the antibody in phosphate buffered saline containing 0.5% non-fat dry milk and 0.05% Tween™ 20.

Note: In order to obtain the best results using various techniques and preparations, we recommend determining the optimal working dilutions by titration.

References

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