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ProductInformation

Anti-DNMT1

Developed in Rabbit IgG Fraction of Antiserum

Product Number **D4567**

Product Description

Anti-DNMT1 is developed in rabbit using as immunogen a synthetic peptide corresponding to amino acids 1581-1595 of rat DNMT1, conjugated to KLH via an N-terminal added lysine residue. The sequence is conserved in human and mouse. Whole antiserum is fractionated and then further purified by ion-exchange chromatography to provide the IgG fraction of antiserum that is essentially free of other rabbit serum proteins.

Anti-DNMT1 recognizes DNMT1 by immunoblotting (approx. 180 kDa) and indirect immunofluorescence. Staining of the DNMT1 band in immunoblotting is specifically inhibited by the DNMT1 immunizing peptide. An additional faint band of approx. 70 kDa may be observed in some preparations; this band is 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. DNA methylation, the major modification of eukaryotic genomes, occurs at the fifth position of cytosine in CpG dinucleotide sequences. DNA methylation is associated with transcriptional repression. 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.

To date, the DNA methylation system is composed of methyl-CpG-binding proteins, as well as of DNA cytosine methyl transferases.^{3, 10} Three DNA (cytosine-5)-methyltransferases have been isolated: DNMT1, -2, and -3 isoforms. The main DNA methyltransferase DNMT1 is suggested to be important for the maintenance of methylation, as well as for the

novo methylation activities occurring in somatic cells of vertebrates. 11 Human DNMT1 is a 1616 amino acids protein; the N-terminal two-thirds of the protein is considered to be the regulatory domain, while the C-terminal region contains the catalytic domain. The catalytic domain shares sequence homologies with all DNMTs. Two isoforms of DNMT1 have been isolated, DNMT1a and DNMT1b, the difference residing in 16 extra amino acids within the latter. DNMT1b is the minor form, representing about 2% of the total DNMT1 protein. DNMT1 is part of different complexes; it can establish a repressive transcription complex consisting of the histone deacetylase HDAC4, DMAP1, and DNMT1 itself. 12 Consistent with the role of DNA methylation in gene silencing, DNMT1 can associate with HDAC1 and MeCP2. Interestingly, in vivo co-expression of hDNMT1 and hDNMT3a or hDNMT3b leads to methylation spreading in the genome, suggesting cooperation between *de novo* and maintenance enzymes during DNA methylation. 13

Antibodies reacting specifically with DNMT1 may be useful for the study of chromatin remodeling effects on gene expression.

Reagent

Anti-DNMT1 is supplied as IgG fraction of antiserum in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide.

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 hazards and safe handling practices.

Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For prolonged storage, freeze in working aliquots at -20 °C. 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 dilution of 1:1,000-1:2,000 is recommended using nuclear extracts of HEK 293T cells.

By indirect immunofluorescence, a working antibody dilution of 1:100-1:200 is recommended using COS-7 cells.

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

References

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