

DNMT Activity/Inhibition Fluorometric Assay Ultra Kit

Product Information

Cat.No.

Kit-0307

Product Overview

DNMT Activity/Inhibition Assay Ultra Kit (Fluorometric) is use for screening DNMT inhibitors.

Description

DNA methylation occurs by a covalent addition of a methyl group at the 5-carbon of the cytosine ring, resulting in 5-methylcytosine. These methyl groups project into the major grooves of DNA and inhibit transcription. In human DNA, 5-methylcytosine is found in approximately 1.5% of genomic DNA, primarily at CpG sites. There are clusters of CpG sites at 0.3 to 2 kb stretches of DNA known as CpG islands that are typically found in or near promoter regions of genes, where transcription is initiated. In the bulk of genomic DNA, most CpG sites are heavily methylated. However, CpG islands in germ-line tissue and promoters of normal somatic cells remain unmethylated, allowing gene expression to occur. When a CpG island in the promoter region of a gene is methylated, the expression of the gene is repressed. The repression can be caused by directly inhibiting the binding of specific transcription factors, and indirectly by recruiting methyl-CpG-binding proteins and their associated repressive chromatin remodeling activity. In addition to the effect on gene transcription, DNA methylation is also involved in genomic imprinting, which refers to a parental origin specific expression of a gene, and the formation of a chromatin domain.

Applications

DNMT Activity/Inhibition Assay Ultra Kit (Fluorometric) is suitable for measuring total DNMT activity or inhibition using nuclear extracts or purified enzymes from a broad range of species such as mammals, plants, fungi, bacteria, and viruses in a variety of forms including, but not limited to, cultured cells and fresh and frozen tissues. Nuclear extracts can be prepared by using your own successful method.

Usage

For research use only (RUO)

Tel: 1-631-559-9269 1-516-512-3133

Fax:1-631-938-8127

Email:info@creative-biomart.org

45-1 Ramsey Road, Shirley, NY 11967, USA

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Storage

Upon receipt: (1) Store MK3, MK4, MK6, MK7 and MK8 at -20°C away from light; (2) Store MK1, MK5, MK9, and the 8-Well Assay Strips at 4°C away from light; (3) Store all remaining components (MK2, MK10, and the Adhesive Covering Film) at room temperature away from light. All components of the kit are stable for 6 months from the date of shipment, when stored properly. Note: (1) Check if MK1 (10X Wash Buffer) contains salt precipitates before use. If so, warm (at room temperature or 37°C) and shake the buffer until the salts are re-dissolved.

Kit Components

MK1 (10X Wash Buffer) 14 ml 4°C MK2 (DNMT Assay Buffer) 4 ml RT MK3 (Adomet, 50X)* 60 µl -20°C MK4 (DNMT Enzyme Control, 50 µg/ml)* 6 µl -20°C MK5 (Capture Antibody, 1000 µg/ml)* 5 µl 4°C MK6 (Detection Antibody, 400 µg/ml)* 6 µl -20°C MK7 (Enhancer Solution)* 6 µl -20°C MK8 (Fluoro Developer)* 6 µl -20°C MK9 (Fluoro Enhancer)* 6 µl 4°C MK10 (Fluoro Dilutor) 4 ml RT 8-Well Assay Strips (With Frame) 6 4°C Adhesive Covering Film 1 RT* Spin the solution down to the bottom prior to use.

Detection method Fluorometric

Compatible Sample Types

Nuclear Extract, Purified Enzyme