

DNMT Activity/Inhibition Colorimetric Assay Ultra Kit

Product Information

Cat.No.

Kit-0306

Product Overview

DNMT Activity/Inhibition Assay Ultra Kit (Colorimetric) 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 (Colorimetric) is a further 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)

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DNMT Activity/Inhibition Colorimetric Assay Ultra Kit

Storage

Upon receipt: (1) Store MU3, MU4, MU6, and MU7 at -20°C away from light; (2) Store MU1, MU5, MU8, and the 8-Well Assay Strips at 4°C away from light; (3) Store all remaining components (MU2, MU9, 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 MU1 (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; and (2) transfer the amount of MU8 required into a secondary container (tube or vial) before adding MU8 into the assay wells in order to avoid contamination. Check if a blue color is present in MU8 (Developer Solution) before each use, as this would indicate contamination of the solution and should not be used.

Kit Components

MU1 (10X Wash Buffer) 14 ml 4°C MU2 (DNMT Assay Buffer) 4 ml RT MU3 (Adomet, 50X)* 60 µl -20°C MU4 (DNMT Enzyme Control, 50 µg/ml)* 6 µl -20°C MU5 (Capture Antibody, 1000 µg/ml*) 5 µl 4°C MU6 (Detection Antibody, 400 µg/ml)* 6 µl -20°C MU7 (Enhancer Solution)* 6 µl -20°C MU8 (Developer Solution) 5 ml 4°C MU9 (Stop Solution) 5 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 Colorimetric

Compatible Sample Types

Nuclear Extract, Purified Enzyme