CREATIVE BIOMART®
Assay Kit

Methylated DNA Quantification Kit (Fluorometric)

Product Information

Cat.No. Kit-0554

Product Overview

Methylated DNA Quantification Kit (Fluorometric) is used for measuring methylated DNA.

Description

DNA methylation occurs by the covalent addition of a methyl group at the 5-carbon of the cytosine ring by DNA methyltransferases, resulting in 5-methylcytosine (5-mC). In somatic cells, 5-mC is found almost exclusively in the context of paired symmetrical methylation of the dinucleotide CpG, whereas in embryonic stem (ES) cells, a substantial amount of 5-mC is also observed in non-CpG contexts. The biological importance of 5-mC as a major epigenetic modification in phenotype and gene expression has been recognized widely. For example, decrease in 5-mC content (DNA hypomethylation) is likely caused by methyl-deficiency due to a variety of environmental influences, and has been proposed as a molecular marker in multiple biological processes such as cancer. It has been well demonstrated that the decrease in DNA methylation is one of the most important characteristics of cancer. Thus, the quantification of 5-mC content or methylation in cancer cells could provide very useful information for detection and analysis of this disease.

Applications

Methylated DNA Quantification Kit (Fluorometric) is suitable for detecting DNA methylation status using DNA isolated from any species such as mammals, plants, fungi, bacteria, and viruses in a variety of forms including, but not limited to, cultured cells, fresh and frozen tissues, paraffin-embedded tissues, plasma/serum samples, and body fluid samples. This kit is particularly suitable for samples only available in small amounts such as laser capture microdissection samples and embryos.

Usage

For research use only (RUO)

Storage

Upon receipt: (1) Store MF3, MF4, MF6, MF7, and MF8 at –20°C away from light; (2) Store MF1, MF5,

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MF9, and 8-Well Assay Strips at 4°C away from light; (3) Store MF2 and MF10 at room temperature away from light.All components of the kit are stable for 6 months from date of shipment, when stored properly.Note: Check MF1 (10X Wash Buffer) contains salt precipitates before use. If so, briefly warm at room temperature or 37°C and shake the buffer until the salts are re-dissolved.

Kit Components

Component Amount Storage Upon ReceiptMF1 (10X Wash Buffer) 14 mL 4°CMF2 (Binding Solution)) 5 mL RTMF3 (Negative Control, 20 μg/mL)* 10 µL -20°CMF4 (Positive Control, 20 μg/mL)* 10 µL -20°CMF5 (Capture Antibody, 1000 µg/mL*) 4 µL 4°CMF6 (Detection Antibody, 400 µg/mL)* 8 µL -20°CMF7 (Enhancer Solution)* 8 µL -20°CMF8 (Fluoro Developer) 8 µL -20°CMF9 (Fluoro Enhancer) 8 µL 4°CMF10 (Fluoro Dilutor) 4 mL RT8-Well Assay Strips (With Frame) 6 4°C* Spin the solution down to the bottom prior to use.Note: The MF3 Negative Control is an unmethylated polynucleotide containing 50% of cytosine. The MF4 Positive Control is a methylated polynucleotide containing 50% of 5-methylcytosine.

Detection method Fluorometric

Compatible Sample Types

DNA Extract

Features & Benefits

Fluorometric assay with easy-to-follow steps for convenience and speed. The entire procedure can be finished within 4 hours.Innovative kit composition enables background signals to be extremely low, which eliminates the need for plate blocking and allows the assay to be simple, accurate, reliable, and consistent.High sensitivity, of which the detection limit can be as low as 50 pg of methylated DNA.Optimized antibody and enhancer solutions allow high specificity to 5-mC, with no cross-reactivity to unmethylated cytosine and no or negligible cross-reactivity to hydroxymethylcytosine within the indicated concentration range of the sample DNA.Universal positive and negative controls are included, which are suitable for quantifying methylated DNA from any species.Strip-well microplate format makes the assay flexible: manual or high throughput analysis.

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