Datasheet for ABIN6971329

anti-5-Hydroxymethylcytosine antibody

4 Images

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Overview

Quantity:	100 µg
Target:	5-Hydroxymethylcytosine (5-hmC)
Reactivity:	Human, Mouse
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This 5-Hydroxymethylcytosine antibody is un-conjugated
Application:	Dot Blot (DB), Immunocytochemistry (ICC), Methylated DNA Immunoprecipitation (MeDIP), Immunofluorescence (IF), Immunohistochemistry (IHC)

Product Details

Immunogen:	This 5-Hydroxymethylcytosine antibody was raised against 5-hydroxymethylcytidine conjugated to KLH and recognizes 5-hydroxymethylcytosine.
Clone:	59-1
Isotype:	IgG2a
Characteristics:	In addition to this monoclonal, We offer two polyclonal antibodies that recognize 5-hydroxymethylcytosine, a whole serum version and a purified IgG version .All are validated for use in methyl DNA immunoprecipitation (MeDIP). For customers that require the ability to quantitate the amount of IgG in the MeDIP reaction, we recommend either this monoclonal or the purified IgG polyclonal . The whole serum version is very high titre and should be used carefully (0.1 - 0.5 µL per IP) as not to generate high non-specific background. The whole serum version has been used successfully in immunofluorescence (IF, Ito et al, 2010), and the purified IgG version is likely to work in this application as well. DNA methylation is an epigenetic event in

Product Details

	which DNA methyltransferases (DNMTs) catalyze the reaction of a methyl group to the fifth carbon of cytosine in a CpG dinucleotide. This modification helps to control gene expression and is also involved in genomic imprinting, while aberrant DNA methylation is often associated with disease. 5-methylcytosine is a modified base that is found in the DNA of plants and vertebrates. A second type of DNA methylation exists, 5-hydroxymethylcytosine (5-hydroxy methylcytosine, 5-hmC). This results from the enzymatic conversion of 5-methylcytosine into 5-hydroxymethylcytosine by the TET family of cytosine oxygenases. This antibody was developed specifically to distinguish 5-hydroxymethylcytosine from 5-methylcytosine as conventional methods (enrichment by antibody or methyl DNA binding protein, enzymatic digestion and bisulfite sequencing) cannot do so. It is possible that 5-hydroxymethylcytosine (5-hmC) represents a pathway to demethylate DNA, as 5-hydroxymethylcytosine is repaired as mismatched DNA and replaced with unmethylated cytosine. 5-Hydroxymethylcytosine (5-hmC) antibody (mAb) (Clone 59.1) was raised in a Mouse host. It has been validated for use in Dot blot, Immunocytochemistry, Immunofluorescence, Immunohistochemistry and Methyl DNA Immunoprecipitation, it has been shown to react with Human and Mouse samples, but the sequence is not species specific so it should react with a wide range of sample types.
Purification:	Protein G Chromatography

Target Details

Target:	5-Hydroxymethylcytosine (5-hmC)
Abstract:	5-hmC Products
Target Type:	Chemical

Application Details

Application Notes:	Optimal working dilution should be determined by the investigator.
Restrictions:	For Research Use only

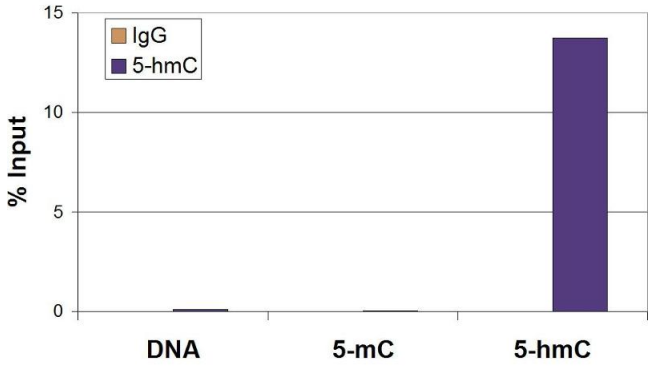
Handling

Buffer:	Purified IgG in 70 mM Tris (pH 8), 105 mM NaCl, 31 mM glycine, 0.07 mM EDTA, 30 % glycerol and 0.035 % sodium azide.
Preservative:	Sodium azide
Precaution of Use:	This product contains Sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which

Handling

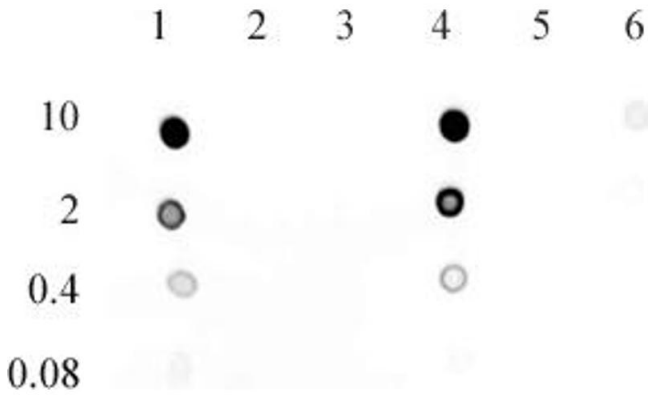
	should be handled by trained staff only.
Storage:	-20 °C
Storage Comment:	Avoid repeated freeze/thaw cycles by aliquoting items into single-use fractions for storage at -20°C for up to 2 years. Keep all reagents on ice when not in storage.

Images



Immunoprecipitation

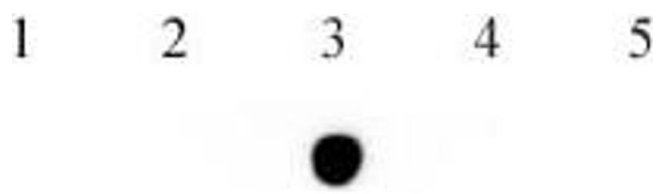
Image 1. 5-Hydroxymethylcytosine (5-hmC, 5-hydroxymethylcytidine) antibody tested by Methyl DNA immunoprecipitation. DNA (25 pg) derived from the promoter of the APC gene was spiked into 500 ng of human genomic DNA and subjected to the MeDIP procedure using 2 µg of 5-Hydroxymethylcytidine antibody (5hmC, maroon bars) or 2 µg of control rabbit IgG (IgG, blue bars). Real time quantitative PCR was performed on the immunoprecipitated DNA and results plotted as % of input DNA. The spiked APC DNA contained either no methylation (DNA), 5-methylcytosine methylation (5-mC) or 5-hydroxymethylcytosine methylation (5-hmC).



Dot Blot

Image 2. 5-Hydroxymethylcytosine (5-hmC) antibody (mAb) tested by dot blot analysis. DNA samples were spotted (indicated in ng on the left) on to a positively charged nylon membrane and blotted with 5-Hydroxymethylcytidine antibody at a 0.2 mg/mL dilution. Lane 1: double-stranded DNA containing 5-hydroxymethylcytosine. Lane 2: double-stranded DNA containing 5-methylcytosine. Lane 3: unmethylated double-stranded DNA. Lane 4: single-stranded DNA containing 5-hydroxymethylcytosine. Lane 5: single-stranded DNA containing 5-methylcytosine. Lane 6: unmethylated single-stranded DNA.

Images



Dot Blot

Image 3. Dot blot of 5-Hydroxymethylcytosine (5-hmC) mAb. Dot blot analysis was used to confirm the specificity of 5-Hydroxymethylcytosine antibody for 5-hydroxymethylcytidine. 10 ng of single-stranded 38 nt DNA oligonucleotides were spotted onto nitrocellulose and probed with the antibody at 0.2 mg/mL. Lane 1: oligo containing unmodified cytidine. Lane 2: oligo containing 5-methylcytidine. Lane 3: oligo containing 5-hydroxymethylcytidine. Lane 4: oligo containing 5-formylcytidine. Lane 5: oligo containing 5-carboxylcytidine.

Please check the product details page for more images. Overall 4 images are available for ABIN6971329.