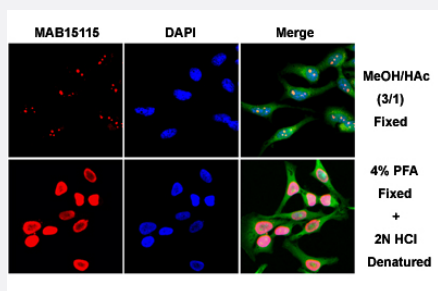


RecomAb™

# 5-methylcytosine (5-mC) monoclonal antibody, clone RM231

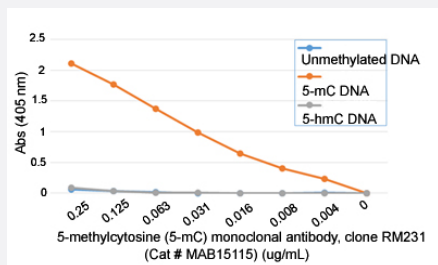
Catalog # MAB15115      Size 50 ug

## Applications



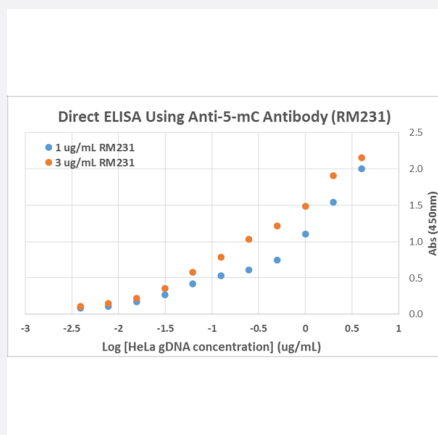
### Immunocytochemistry

Immunocytochemistry staining of HeLa cells with 5-methylcytosine (5-mC) monoclonal antibody, clone RM231 (Cat # MAB15115) (Red). Actin filaments have been labeled with fluorescein phalloidin (Green), and nuclei stained with DAPI (Blue).



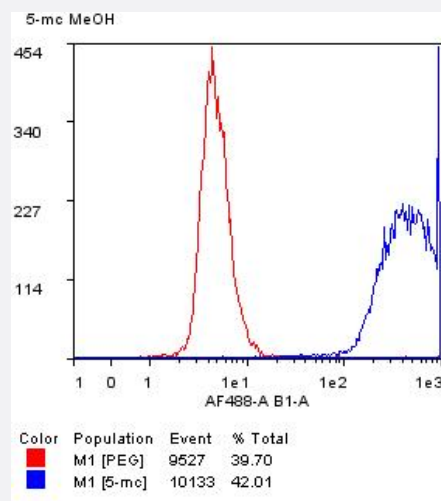
### Enzyme-linked Immunoabsorbent Assay

ELISA analysis of single stranded DNA with 5-methylcytosine (5-mC) monoclonal antibody, clone RM231 (Cat # MAB15115). The plate was coated with streptavidin and then biotinylated single stranded unmethylated DNA, 5-Methylcytosine (5-mC) DNA, and 5-Hydroxymethylcytosine (5-hmC) DNA. A serial dilution of MAB15115 was used as the primary antibody, and an alkaline phosphatase conjugated anti-rabbit IgG as the secondary antibody.



### Enzyme-linked Immunoabsorbent Assay

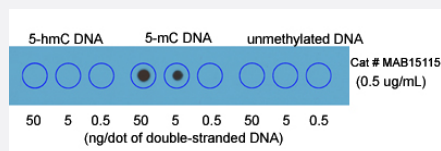
Direct ELISA of HeLa cell genomic DNA using 5-methylcytosine (5-mC) monoclonal antibody, clone RM231 (Cat# MAB15115). The plate was directly coated with different concentrations of genomic DNA isolated from HeLa cells. 1 ug/mL or 3 ug/mL of Cat# MAB15115 was used as the primary antibody, and a HRP conjugated anti-rabbit IgG as the secondary antibody.



## Enzyme-linked Immunoabsorbent Assay

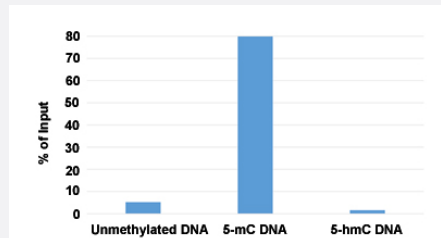
Flow Cytometry analysis of 5-mC expression in HEK293 cells using 5-methylcytosine (5-mC) monoclonal antibody, clone RM231 (Cat# MAB15115). The cells were fixed with ice-cold MeOH, permeabilized with 0.5% Triton X-100, denatured with 2N HCl, then stained with Cat# MAB15115 (anti-5-mC, Blue) or with a negative control antibody (RM105, Red).

## Dot Blot



Dot blot analysis of double stranded DNA with 5-methylcytosine (5-mC) monoclonal antibody, clone RM231 (Cat # MAB15115). The membrane was pre-spotted with 50, 5, and 0.5 ng/dot of double stranded 5-Hydroxymethylcytosine (5-hmC) DNA, 5-Methylcytosine (5-mC) DNA, and unmethylated DNA. The pre-spotted membrane was then blotted with MAB15115.

## Methylated DNA Immunoprecipitation



Methylated DNA Immunoprecipitation (MeDIP) analysis of 5-methylcytosine (5-mC) monoclonal antibody, clone RM231 (Cat # MAB15115) at a 2:1 DNA:Ab ratio. 1 ng of unmethylated, 5-Methylcytosine (5-mC) or 5-Hydroxymethylcytosine (5-hmC) DNA standard (897 bp) was spiked in 1 ug of genomic DNA isolated from HeLa cells as the control. Realtime PCR was then performed to determine the capture of DNA standard as in % of input.

## Specification

<b>Product Description</b>	Rabbit recombinant monoclonal antibody raised against 5-methylcytosine (5-mC).
<b>Antibody Species</b>	Rabbit
<b>Immunogen</b>	Original antibody is raised against 5-methylcytosine conjugated with BSA.
<b>Sequence</b>	N/A
<b>Specificity</b>	This antibody reacts to 5-methylcytosine in both single-stranded and double-stranded DNA. No cross reactivity with non-methylated cytosine and hydroxymethylcytosine in DNA.

<b>Form</b>	Liquid
<b>Purification</b>	Protein A affinity purification
<b>Isotype</b>	IgG
<b>Recommend Usage</b>	Dot Blot (0.5-2 ug/mL) ELISA (0.1-1 ug/mL) Immunocytochemistry (0.5-2 ug/mL) Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) Methylated DNA Immunoprecipitation (0.2-2 ug/mL) The optimal working dilution should be determined by the end user.
<b>Storage Buffer</b>	In PBS (50% glycerol, 1% BSA, 0.09% sodium azide)
<b>Storage Instruction</b>	Store at -20°C. Aliquot to avoid repeated freezing and thawing.
<b>Note</b>	This product contains sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.

## Applications

- Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections)

- Immunocytochemistry

Immunocytochemistry staining of HeLa cells with 5-methylcytosine (5-mC) monoclonal antibody, clone RM231 (Cat # MAB15115) (Red). Actin filaments have been labeled with fluorescein phalloidin (Green), and nuclei stained with DAPI (Blue).

- Enzyme-linked Immunoabsorbent Assay

ELISA analysis of single stranded DNA with 5-methylcytosine (5-mC) monoclonal antibody, clone RM231 (Cat # MAB15115). The plate was coated with streptavidin and then biotinylated single stranded unmethylated DNA, 5-Methylcytosine (5-mC) DNA, and 5-Hydroxymethylcytosine (5-hmC) DNA. A serial dilution of MAB15115 was used as the primary antibody, and an alkaline phosphatase conjugated anti-rabbit IgG as the secondary antibody.

- Enzyme-linked Immunoabsorbent Assay

Direct ELISA of HeLa cell genomic DNA using 5-methylcytosine (5-mC) monoclonal antibody, clone RM231 (Cat# MAB15115). The plate was directly coated with different concentrations of genomic DNA isolated from HeLa cells. 1 ug/mL or 3 ug/mL of Cat# MAB15115 was used as the primary antibody, and a HRP conjugated anti-rabbit IgG as the secondary antibody.

- Enzyme-linked Immunoabsorbent Assay

Flow Cytometry analysis of 5-mC expression in HEK293 cells using 5-methylcytosine (5-mC) monoclonal antibody, clone RM231 (Cat# MAB15115). The cells were fixed with ice-cold MeOH, permeabilized with 0.5% Triton X-100, denatured with 2N HCl, then stained with Cat# MAB15115 (anti-5-mC, Blue) or with a negative control antibody (RM105, Red).

- Dot Blot

Dot blot analysis of double stranded DNA with 5-methylcytosine (5-mC) monoclonal antibody, clone RM231 (Cat # MAB15115). The membrane was pre-spotted with 50, 5, and 0.5 ng/dot of double stranded 5-Hydroxymethylcytosine (5-hmC) DNA, 5-Methylcytosine (5-mC) DNA, and unmethylated DNA. The pre-spotted membrane was then blotted with MAB15115.

- Methylated DNA Immunoprecipitation

Methylated DNA Immunoprecipitation (MeDIP) analysis of 5-methylcytosine (5-mC) monoclonal antibody, clone RM231 (Cat # MAB15115) at a 2:1 DNA:Ab ratio. 1 ng of unmethylated, 5-Methylcytosine (5-mC) or 5-Hydroxymethylcytosine (5-hmC) DNA standard (897 bp) was spiked in 1 ug of genomic DNA isolated from HeLa cells as the control. Realtime PCR was then performed to determine the capture of DNA standard as in % of input.

## Publication Reference

- [Discovery of a new predominant cytosine DNA modification that is linked to gene expression in malaria parasites.](#)

Hammam E, Ananda G, Sinha A, Scheidig-Benatar C, Bohec M, Preiser PR, Dedon PC, Scherf A, Vembar SS.

Nucleic Acids Research 2020 Jan; 48(1):184.

Application: IP, WB, Parasites, DNA, Parasites