

# Product Datasheet

## 5-MethylCytosine Antibody - Azide and BSA Free NB100-744

Unit Size: 0.125 mg

Store at -20C. Avoid freeze-thaw cycles.

[www.novusbio.com](http://www.novusbio.com)



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### Publications: 4

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Updated 9/9/2025 v.20.1

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**NB100-744****5-MethylCytosine Antibody - Azide and BSA Free**

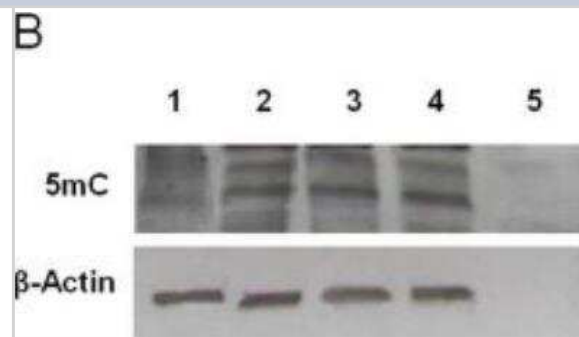
<b>Product Information</b>	
<b>Unit Size</b>	0.125 mg
<b>Concentration</b>	Please see the vial label for concentration. If unlisted please contact technical services.
<b>Storage</b>	Store at -20C. Avoid freeze-thaw cycles.
<b>Clonality</b>	Polyclonal
<b>Preservative</b>	No Preservative
<b>Isotype</b>	IgG
<b>Purity</b>	Protein G purified
<b>Buffer</b>	PBS (pH 7.5)

<b>Product Description</b>	
<b>Description</b>	Novus Biologicals Sheep 5-MethylCytosine Antibody - Azide and BSA Free (NB100-744) is a polyclonal antibody validated for use in IHC, WB, ICC/IF and IP. Anti-5-MethylCytosine Antibody: Cited in 4 publications. All Novus Biologicals antibodies are covered by our 100% guarantee.
<b>Host</b>	Sheep
<b>Species</b>	Human, Rat, All Species
<b>Reactivity Notes</b>	Human reactivity reported in scientific literature (PMID: 20808872).
<b>Specificity/Sensitivity</b>	This is specific for 5-Methyl Cytosine. It was tested using immunoprecipitation against 5-Methyl Cytosine and bromodeoxyuridine (BrdU) or a control (no antigen).
<b>Immunogen</b>	5-Methyl cytosine conjugated to KLH.

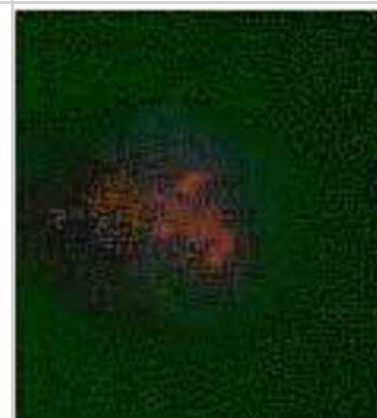
<b>Product Application Details</b>	
<b>Applications</b>	Western Blot, Immunohistochemistry-Paraffin, Immunocytochemistry/Immunofluorescence, Immunohistochemistry, Immunoprecipitation
<b>Recommended Dilutions</b>	Western Blot, Immunohistochemistry 1:10-1:500, Immunocytochemistry/Immunofluorescence 1:10-1:500, Immunoprecipitation 25-100 ug/ml, Immunohistochemistry-Paraffin 10 ug/ml
<b>Application Notes</b>	Western blot images and application from PMID 20808872.

## Images

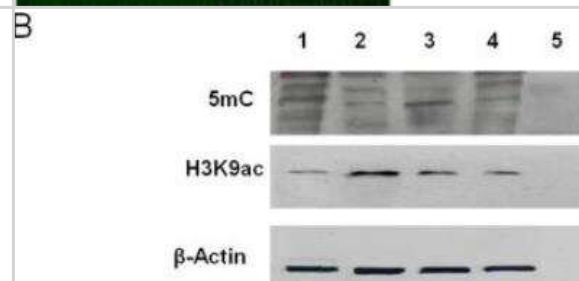
Western Blot: 5-MethylCytosine Antibody [NB100-744] - Global epigenetic changes in HFFs 7 days after hESC extract treatment. (B) Immunoblotting analysis of 5-methyl cytosine. Image collected and cropped by CiteAb from the following publication ([dx.plos.org/10.1371/journal.pone.0012297](https://doi.org/10.1371/journal.pone.0012297)), licensed under a CC-BY license.



Immunocytochemistry/Immunofluorescence: 5-MethylCytosine Antibody [NB100-744] - Positive staining of rat neuron cells performed with 2% BSA/0.1% Tween 20 in PBS blocking and incubated at 4 degrees C overnight using NB100-744 at a dilution of 1:200.



Western Blot: 5-MethylCytosine Antibody [NB100-744] - Immunoblotting analysis shows decrease in 5-methyl cytosine and increase in H3K9 acetylation levels after combined treatment. Lanes 1 to 5 were loaded with proteins (2 ug) from HFFs, hESCs, inhibitor treated HFFs and combined treated HFFs. Image collected and cropped by CiteAb from the following publication ([dx.plos.org/10.1371/journal.pone.0012297](https://doi.org/10.1371/journal.pone.0012297)), licensed under a CC-BY license.



## Publications

Kim S, Hill A, Warman ML, Smits P. Golgi Disruption and Early Embryonic Lethality in Mice Lacking USO1 PLoS One 2012-01-01 [PMID: 23185636]

Sahaboglu A, Barth M et al. Olaparib significantly delays photoreceptor loss in a model for hereditary retinal degeneration. Sci Rep 2016-12-22 [PMID: 28004814] (IF/IHC, Mouse)

Farinelli P, Perera A, Arango-Gonzalez B et al. DNA methylation and differential gene regulation in photoreceptor cell death. Cell Death Dis 2014-12-04 [PMID: 25476906] (IP)

Han J, Sachdev PS, Sidhu KS. A combined epigenetic and non-genetic approach for reprogramming human somatic cells. PLoS One 2010-01-01 [PMID: 20808872] (WB, Human)

## Procedures

### Protocol specific for Cytosine (5-Methyl) Antibody (NB100-744)

Protocol specific for Cytosine (5-Methyl) Antibody (NB100-744):

Use 0.5 to 1 ug of sonicated or enzyme-digested genomic DNA

IP:

1) Dilute to a final volume of 100 ul and a final concentration of 0.15% SDS, 1% triton x-100, 150mM NaCl, 1mM EDTA pH8.0, 0.5 mM EGTA pH8.0, 10 mM Tris pH8.0, 0.1 % BSA, 7 mM NaOH (crucial), anti-5mC (up to 30 ug of antibody for saturating conditions), Prot A/G beads.

2) Rotate o/n at 4C.

Wash:

1) 2x in: 0.1% SDS, 0.1% sodium deoxycholate, 1% triton, 150 mM NaCl, 1mM EDTA pH8.0, 0.5 mM, EGTA pH8.0, 10 mM Tris pH8.0

2) 1x in: same as above, but NaCl is 500mM

3) 1x in: 0.25 M LiCl, 0.5% sodium deoxycholate, 0.5% NP-40, 1mM EDTA pH8.0, 0.5 mM, EGTA pH8.0, 10 mM Tris pH8.0.

4) 2x in: 1mM EDTA pH8.0, 0.5 mM EGTA pH8.0, 10 mM Tris pH8.0.

Elute:

1) Elute in 1% SDS, 100mM NaHCO<sub>3</sub>.

2) Purify DNA using phenol/chloroform extraction and EtOH precipitation.

3) Analyze using real-time PCR.



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### **Products Related to NB100-744**

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NBP2-33376H	Blue Marker Antibody (6F4-F6) [HRP]
HAF016	Donkey anti-Sheep IgG Secondary Antibody [HRP]
NL010	Donkey anti-Sheep IgG Secondary Antibody [NL557]
NBP1-97055-10mg	Sheep IgG Isotype Control

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### **Limitations**

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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