Product Datasheet

DNMT3B Antibody
NB100-266

Unit Size: 0.1 ml

Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.

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Reviews: 1 Publications: 17

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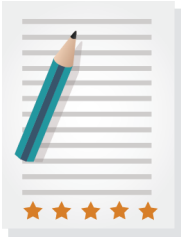

NB100-266

DNMT3B Antibody

Product Information	
Unit Size	0.1 ml
Concentration	This product is unpurified. The exact concentration of antibody is not quantifiable.
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	No Preservative
Isotype	IgG
Purity	Unpurified
Buffer	Whole antisera
Target Molecular Weight	100 kDa

Product Description	
Host	Rabbit
Gene ID	1789
Gene Symbol	DNMT3B
Species	Human, Mouse
Reactivity Notes	Human and mouse.
Immunogen	Amino acids 4-101 of human Dnmt3b [UniProt# Q9UBC3]

Product Application Details	
Applications	Western Blot, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin, Immunoprecipitation
Recommended Dilutions	Western Blot 1:1000, Immunohistochemistry 1:200, Immunocytochemistry/ Immunofluorescence 1:100-1:1000, Immunoprecipitation 1:1000, Immunohistochemistry-Paraffin 1:200
Application Notes	This Dnmt3b antibody is useful for Immunocytochemistry/Immunofluorescence, Immunohistochemistry on paraffin-embedded sections, Immunoprecipitation and Western Blot, where a band is seen at ~100 kDa. This antibody does not neutralize the enzymatic activity of its corresponding methyltransferase. In ICC/IF, cytoplasmic staining was observed in NIH-3T3 cells. In IHC, predominant nuclear staining was observed, as well as some weaker cytoplasmic signal, in mouse testes. Prior to immunostaining paraffin tissues, antigen retrieval with sodium citrate buffer (pH 6.0) is recommended. The observed molecular weight of the protein may vary from the listed predicted molecular weight due to post translational modifications, post translation cleavages, relative charges, and other experimental factors.

Publications

Cardenas H, Fang F, Jiang G et al. Methylomic Signatures of High Grade Serous Ovarian Cancer Epigenetics 2020-12-08 [PMID: 33289590]

Rinaldi L, Datta D, Serrat J et al. Dnmt3a and Dnmt3b Associate with Enhancers to Regulate Human Epidermal Stem Cell Homeostasis. CellPress 2016-07-28 [PMID: 27476967] (WB, Chemotaxis, ICC/IF, Human)

Liu Y, Sun L, Fong P et al. An association between overexpression of DNA methyltransferase 3B4 and clear cell renal cell carcinoma. Oncotarget. 2017-03-21 [PMID: 28160561] (WB, Human)

Hoang TV, Horowitz ER, Chaffee BR et al. Lens development requires DNMT1 but takes place normally in the absence of both DNMT3A and DNMT3B activity. Epigenetics. 2016-11-08 [PMID: 27824296] (Mouse)

Al-Salihi M, Yu M, Burnett DM et al. The depletion of DNA methyltransferase-1 and the epigenetic effects of 5-aza-2'deoxycytidine (decitabine) are differentially regulated by cell cycle progression. Epigenetics 2011-08-01 [PMID: 21725200] (Human)

Quist T, Jin H, Zhu Jf et al. The impact of osteoblastic differentiation on osteosarcomagenesis in the mouse. Oncogene. 2014-10-27 [PMID: 25347737] (IHC-P, Mouse)

Kinney SM, Chin HG, Vaisvila R et al. Tissue-specific distribution and dynamic changes of 5-hydroxymethylcytosine in mammalian genomes. J Biol Chem. 2011-07-01 [PMID: 21610077] (WB, Mouse)

Qiu YY, Mirkin BL, Dwivedi RS et al. Differential expression of DNA-methyltransferases in drug resistant murine neuroblastoma cells. Cancer Detect Prev;26(6):444-53. 2002-01-01 [PMID: 12507229] (WB, ICC/IF, Mouse)

Chen M, Shabashvili D, Nawab A, Yang SX, Dyer LM, Brown KD, Hollingshead M, Hunter KW, Kaye FJ, Hochwald SN, Marquez VE, Steeg P, Zajac-Kaye M. DNA methyltransferase inhibitor, zebularine, delays tumor growth and induces apoptosis in a genetically engineered mouse model of breast cancer. Mol Cancer Ther;11(2):370-82. 2012-02-01 [PMID: 22203734]

Kim DH, Jung YJ, Lee JE et al. SIRT1 activation by resveratrol ameliorates cisplatin-induced renal injury through deacetylation of p53. Am J Physiol Renal Physiol;301(2):F427-35. 2011-08-01 [PMID: 21593185]

Morey Kinney SR, Smiraglia DJ, James SR et al. Stage-specific alterations of DNA methyltransferase expression, DNA hypermethylation, and DNA hypomethylation during prostate cancer progression in the transgenic adenocarcinoma of mouse prostate model. Mol Cancer Res;6(8):1365-74. 2008-08-01 [PMID: 18667590] (WB, Mouse)

Robert MF, Morin S, Beaulieu N et al. DNMT1 is required to maintain CpG methylation and aberrant gene silencing in human cancer cells. Nat Genet;33(1):61-5. 2003-01-01 [PMID: 12496760] (WB, Human)

More publications at http://www.novusbio.com/NB100-266

Procedures

Protocol Specific for NB100-266
Western Blot Protocol
1. Perform SDS-PAGE on samples to be analyzed, loading 40 ug of total protein per lane.
2. Transfer proteins to membrane according to the instructions provided by the manufacturer of the membrane and transfer apparatus.
3. Stain according to standard Ponceau S procedure (or similar product) to assess transfer success, and mark molecular weight standards where appropriate.
4. Rinse the blot.
5. Block the membrane using standard blocking buffer for at least 1 hour.
6. Wash the membrane in wash buffer three times for 10 minutes each.
7. Dilute primary antibody in blocking buffer and incubate 1 hour at room temperature.
8. Wash the membrane in wash buffer three times for 10 minutes each.
9. Apply the diluted HRP conjugated secondary antibody in blocking buffer (as per manufacturers instructions) and incubate 1 hour at room temperature.
10. Wash the blot in wash buffer three times for 10 minutes each (this step can be repeated as required to reduce background).
11. Apply the detection reagent of choice in accordance with the manufacturers instructions.
Note: Tween-20 can be added to the blocking or antibody dilution buffer at a final concentration of 0.05-0.2%.

Immunohistochemistry-Paraffin Embedded Sections

Antigen Unmasking:
Bring slides to a boil in 10 mM sodium citrate buffer (pH 6.0) then maintain at a sub-boiling temperature for 10 minutes. Cool slides on bench-top for 30 minutes.

Staining:
1. Wash sections in deionized water three times for 5 minutes each.
2. Wash sections in wash buffer for 5 minutes.
3. Block each section with 100-400 ul blocking solution for 1 hour at room temperature.
4. Remove blocking solution and add 100-400 ul diluted primary antibody. Incubate overnight at 4 degrees C.
5. Remove antibody solution and wash sections in wash buffer three times for 5 minutes each.
6. Add 100-400 ul biotinylated diluted secondary antibody. Incubate 30 minutes at room temperature.
7. Remove secondary antibody solution and wash sections three times with wash buffer for 5 minutes each.
8. Add 100-400 ul Streptavidin-HRP reagent to each section and incubate for 30 minutes at room temperature.
9. Wash sections three times in wash buffer for 5 minutes each.
10. Add 100-400 ul DAB substrate to each section and monitor staining closely.
11. As soon as the sections develop, immerse slides in deionized water.
12. Counterstain sections in hematoxylin.
13. Wash sections in deionized water two times for 5 minutes each.
14. Dehydrate sections.
15. Mount coverslips.

Immunocytochemistry Protocol

Culture cells to appropriate density in 35 mm culture dishes or 6-well plates.

1. Remove culture medium and add 10% formalin to the dish. Fix at room temperature for 30 minutes.
2. Remove the formalin and add ice cold methanol. Incubate for 5-10 minutes.
3. Remove methanol and add washing solution (i.e. PBS). Be sure to not let the specimen dry out. Wash three times for 10 minutes.
4. To block nonspecific antibody binding incubate in 10% normal goat serum from 1 hour to overnight at room temperature.
5. Add primary antibody at appropriate dilution and incubate at room temperature from 2 hours to overnight at room temperature.
6. Remove primary antibody and replace with washing solution. Wash three times for 10 minutes.
7. Add secondary antibody at appropriate dilution. Incubate for 1 hour at room temperature.
8. Remove antibody and replace with wash solution, then wash for 10 minutes. Add Hoechst 33258 to wash solution

at 1:25,0000 and incubate for 10 minutes. Wash a third time for 10 minutes.
9. Cells can be viewed directly after washing. The plates can also be stored in PBS containing Azide covered in Parafilm (TM). Cells can also be cover-slipped using Fluoromount, with appropriate sealing.

*The above information is only intended as a guide. The researcher should determine what protocol best meets their needs. Please follow safe laboratory procedures.

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