# Product Datasheet

## 5-MethylCytosine Antibody (15H61) - BSA Free
## NBP2-59166

Unit Size: 50 ug

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

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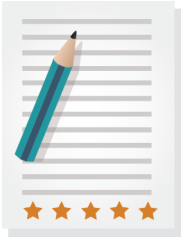
Updated 2/23/2025 v.20.1





# NBP2-59166

5-MethylCytosine Antibody (15H61) - BSA Free

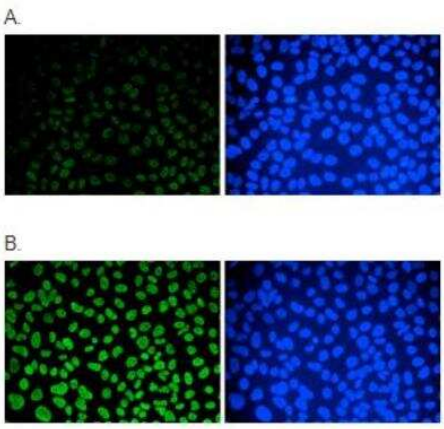
| Product Information | |
|---|---|
| **Unit Size** | 50 ug |
| **Concentration** | Please see the vial label for concentration. If unlisted please contact technical services. |
| **Storage** | Store at -20C. Avoid freeze-thaw cycles. |
| **Clonality** | Monoclonal |
| **Clone** | 15H61 |
| **Preservative** | 0.05% Sodium Azide |
| **Isotype** | IgG1 |
| **Purity** | Protein A purified |
| **Buffer** | PBS |

| Product Description | |
|---|---|
| **Description** | Monoclonal antibody raised in mouse against 5-mC (5-methylcytosine) conjugated to BSA. |
| **Host** | Mouse |
| **Species** | Human, Mouse, Drosophila |
| **Immunogen** | The exact immunogen is propietary information. |

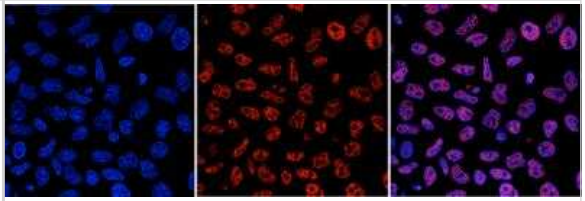
| Product Application Details | |
|---|---|
| **Applications** | Dot Blot, Immunocytochemistry/ Immunofluorescence, In-situ Hybridization, Methylated DNA Immunoprecipitation |
| **Recommended Dilutions** | Immunocytochemistry/ Immunofluorescence 1:1000, In-situ Hybridization 1:200 - 1:1000, Dot Blot 1:600, Methylated DNA Immunoprecipitation 1 - 2 ug/IP |

## Images

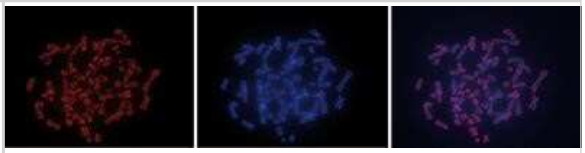
Immunocytochemistry/Immunofluorescence: 5-MethylCytosine Antibody (C.15200003) [NBP2-59166] - Human osteosarcoma (U2OS) cells were stained with the monoclonal antibody against 5-mC. Cells were fixed with 2.5% PFA in PBS for 30 minutes, permeabilized with 0.5% Triton X-100 for 1 hour and treated with 2N HCl for 1 hour followed by 2 x 5 minutes with 0.1 M borate buffer to depurinate the DNA. After blocking with PBS containing 0.1% Triton X-100 and 1% BSA, the cells were immunofluorescently labelled with the 5-mC antibody diluted 1:500 in blocking solution, followed by a goat anti-mouse antibody conjugated to Alexa488. Figure 3A: cells were immunofluorescently labelled with the 5-mC antibody after incubation of the antibody with 50 uM mCTP (left) or with DAPI (right). Figure 3B: staining of the cells with the 5-mC antibody after incubation of the antibody with 50 uM hmCTP and with DAPI.



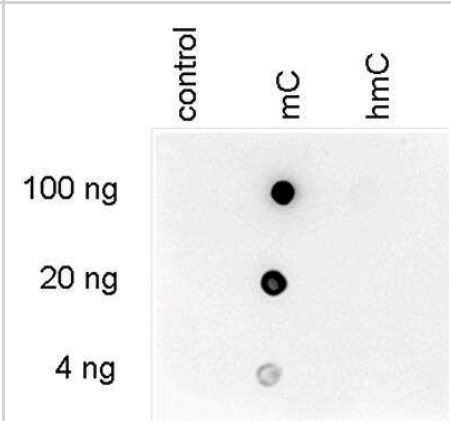

Immunocytochemistry/Immunofluorescence: 5-MethylCytosine Antibody (C.15200003) [NBP2-59166] - HeLa cells were stained with the antibody against 5-mC and with DAPI. Cells were fixed with 4% formaldehyde for 10 minutes and blocked with PBS/Triton X-100 containing 1% BSA. The cells were immunofluorescently labelled with the 5-mC antibody (middle) diluted 1:1000 in blocking solution followed by an anti-mouse antibody conjugated to Alexa594. The left panel shows staining of the nuclei with DAPI. A merge of the two stainings is shown on the right.



In-situ Hybridization: 5-MethylCytosine Antibody (C.15200003) [NBP2-59166] - To detect methylated chromosomal regions, FISH was performed on metaphase chromosomes from HeLa cells using the antibody against 5-mC. The cells were blocked in metaphase by treatment with colcemid (0.1 ug/mL) for 1 - 2 hours, fixed overnight at -20C with ethanol/glacial acetic acid and treated with 2N HCl for 30 minutes at room temperature. Subsequently, the cells were blocked with PBS containing 1% BSA and 0.1% Triton X-100 and stained with the 5-mC antibody (left) diluted 1:1,000 in blocking solution, followed by an anti-mouse antibody conjugated to Alexa594. The middle panel shows staining of the chromosomes with DAPI. A merge of the two stainings is shown on the right.



Dot Blot: 5-MethylCytosine Antibody (C.15200003) [NBP2-59166] - To demonstrate the specificity of the antibody against 5-mC, a Dot blot analysis was performed using hmC, mC and C controls. 100 to 4 ng (equivalent of 5 to 0.2 pmol of C-bases) of the controls were spotted on a membrane (Amersham Hybond-N+). The antibody was used at a dilution of 1:600. Dot blot shows a high specificity of the antibody for the methylated control.



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## Products Related to NBP2-59166

| | |
|---|---|
| HAF007 | Goat anti-Mouse IgG Secondary Antibody [HRP] |
| NB720-B | Rabbit anti-Mouse IgG (H+L) Secondary Antibody [Biotin] |
| NBP1-97005-0.5mg | Mouse IgG1 Isotype Control (MG1) |
| NBP2-62131 | 5-MethylCytosine ELISA Kit (Colorimetric) |

### 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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