## For Research Use Only

## m5C Monoclonal antibody

Catalog Number: 68301-1-Ig



**Basic Information** 

Catalog Number: 68301-1-lg Size: 500 µ g/ml

Mouse Isotype: IgG2b

Source:

GenBank Accession Number:

GeneID (NCBI): Full Name: Purification Method: Protein A purification

CloneNo.: 2C9G9

Recommended Dilutions: IHC 1:2500-1:10000 Dot Blot 1:2500-1:10000

**Applications** 

Tested Applications: Dot Blot, IHC, ELISA Species Specificity: Human, Mouse, Rat, m5C

Note-IHC: suggested antigen retrieval with TE buffer pH 9.0; (\*) Alternatively, antigen retrieval may be performed with citrate

buffer pH 6.0

Positive Controls

IHC: mouse testis tissue, rat brain tissue, rat testis

tissue

Dot Blot: HeLa cells,

## **Background Information**

In eukaryotes, 5-methylcytosine in DNA (5mC) and its oxidized derivatives (5-hydroxymethylcytosine (5hmC), 5-formylcytosine (5fC), and 5-carboxylcytosine (5caC)) are the most prominent modifications and have been suggested to contribute to epigenetic gene regulation through a variety of different mechanisms. m5C has also been found in mRNA, rRNA and tRNA of representative organisms from all kinds of species. As reversible epigenetic modifications, m5C modifications of RNA affect the fate of the modified RNA molecules and play important roles in various biological processes including RNA stability control, protein synthesis, and transcriptional regulation (PMID: 35365216).

Storage

Storage:

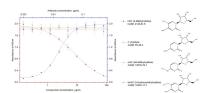
Store at -20°C. Stable for one year after shipment.

Storage Buffer

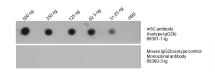
PBS with 0.02% sodium azide and 50% glycerol pH 7.3.

Aliquoting is unnecessary for -20°C storage

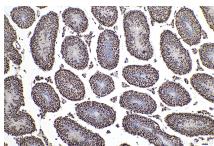
## **Selected Validation Data**

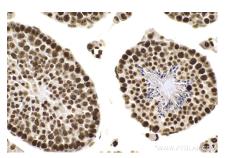


Indirect ELISA and competitive ELISA results show that this antibody is specific to m5C. Indirect ELISA (blue curve, refer to top X-right Y axis) was performed by coating BSA conjugated m5C at 0.4ng/well followed by blocking with 1% BSA. Serial diluted primary antibody was added to the plates and incubated at 37°C. HRP-goat anti-mouse was used for detection. Competitive ELISA was performed similarly except that different concentration of m5C or its structure analogue

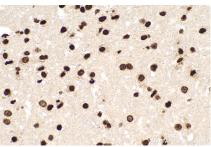


Total DNA was isolated from HeLa cell line and was dotted to NC membrane at different amount as indicated above the dots. The membrane was blocked with BSA and blotted with m5C antibody 68301-1-lg at 1:5000 followed by incubation of HRP-goat anti-mouse secondary antibody. Signal was developed by ECL substrate. A parallel dot blot was performed using Mouse IgG2b isotype control Monoclonal antibody 66360-3-lg at the same dose.

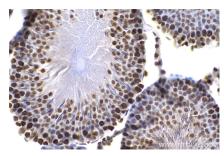




Immunohistochemical analysis of paraffinembedded mouse testis tissue slide using 68301-1-Ig (m5C antibody) at dilution of 1:5000 (under 40x lens). Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0).



Immunohistochemical analysis of paraffinembedded rat brain tissue slide using 68301-1-lg (m5C antibody) at dilution of 1:5000 (under 40x lens). Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0).



Immunohistochemical analysis of paraffinembedded rat testis tissue slide using 68301-1-lg (m5C antibody) at dilution of 1:5000 (under 40x lens). Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0).