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m6A

Cat.No. 202 111; Monoclonal mouse antibody, 100 µg purified IgG (lyophilized)

Data Sheet

Reconstitution/ Storage	100 μ g purified IgG, lyophilized. Albumin and azide were added for stabilization. For reconstitution add 100 μ l H ₂ O to get a 1mg/ml solution in PBS. Then aliquot and store at -20°C to -80°C until use. Antibodies should be stored at +4°C when still lyophilized. Do not freeze! For detailed information, see back of the data sheet.
Applications	Dot blot: 1:1000 IP: yes ICC: not tested yet IHC: not tested yet IHC-P: not tested yet ELISA: yes MeRIP: yes
Clone	212B11
Subtype	IgG2b (κ light chain)
Immunogen	N6-methyladenosine fused to BSA.
Reactivity	Reacts with: human, rat, mouse, eukaryotes, prokaryotes. Other species not tested yet.
Specificity	Specific for N6-methyladenosine (m6A) with some cross-reactivity to m6Am.

TO BE USED IN VITRO / FOR RESEARCH ONLY NOT TOXIC, NOT HAZARDOUS, NOT INFECTIOUS, NOT CONTAGIOUS

Background

m6A (N6-methyladenosine) is a posttranscriptional RNA-modification found throughout all kingdoms, e.g. in vertebrate snRNAs U2, U4, U6, in viral and eukaryotic mRNAs, and in E. coli 16S rRNA. Recent studies have found that mRNA is predominately m6A modified at stop codons and long internal exons, which are conserved between mouse and human. The so-called RNA methylome probably plays an important role in in the regulation of gene expression.

In E. coli Dam methylase introduces m6A modifications on the DNA level at the 5'-GATC-3' motif. This allows the cell to differentiate between the parental and the daughter strand during mismatch repair.

Selected References for 202 111

Enterotoxigenic Escherichia coli infection promotes enteric defensin expression via FOXO6-METTL3-m6A-GPR161 signalling axis.

Zong X, Wang H, Xiao X, Zhang Y, Hu Y, Wang F, Wang Y, Lu Z RNA biology (2020): 1-11.. **DOTBLOT, MERIP; tested species: pig**

A lncRNA from the FTO locus acts as a suppressor of the m6A writer complex and p53 tumor suppression signaling. Zhang J, Wei J, Sun R, Sheng H, Yin K, Pan Y, Jimenez R, Chen S, Cui XL, Zou Z, Yue Z, et al. Molecular cell (2023) 8315: 2692-2708.e7.. **DOTBLOT, MERIP; tested species: mouse**

Light-induced LLPS of the CRY2/SPA1/FIO1 complex regulating mRNA methylation and chlorophyll homeostasis in Arabidopsis. Jiang B, Zhong Z, Gu L, Zhang X, Wei J, Ye C, Lin G, Qu G, Xiang X, Wen C, Gateas M, et al. Nature plants (2023):.. ELISA

N6-Adenosine Methylation in RNA and a Reduced m3G/TMG Level in Non-Coding RNAs Appear at Microirradiation-Induced DNA Legions

Svobodová Kovaříková A, Stixová L, Kovařík A, Komůrková D, Legartová S, Fagherazzi P, Bártová E Cells (2020) 92: . . ICC; tested species: mouse

Identification of Methylated Deoxyadenosines in Genomic DNA by dA6m DNA Immunoprecipitation.

Koziol MJ, Bradshaw CR, Allen GE, Costa AS, Frezza C

Bio-protocol (2016) 621:.. IP

N6-Methyladenosine Promotes TNF mRNA Degradation In CD4+ T Lymphocytes.

van Vroonhoven ECN, Picavet LW, Scholman RC, Sijbers LJPM, Kievit CRE, van den Dungen NAM, Mokry M, Evers A, Lebbink RJ, Mocholi E, Coffer PJ, et al.

Journal of leukocyte biology (2024):.. MERIP; tested species: human

Evaluation of N 6-methyldeoxyadenosine antibody-based genomic profiling in eukaryotes.

Debo BM, Mallory BJ, Stergachis AB

Genome research (2023) 333: 427-434. . DOTBLOT

A Neuroligin-1 mutation associated with Alzheimer's disease produces memory and age-dependent impairments in hippocampal plasticity.

Arias-Aragón F, Tristán-Clavijo E, Martínez-Gallego I, Robles-Lanuza E, Coatl-Cuaya H, Martín-Cuevas C, Sánchez-Hidalgo AC, Rodríguez-Moreno A, Martinez-Mir A, Scholl FG

iScience (2023) 266: 106868. . DOTBLOT

mRNA stability and m6A are major determinants of subcellular mRNA localization in neurons.

Loedige I, Baranovskii A, Mendonsa S, Dantsuji S, Popitsch N, Breimann L, Zerna N, Cherepanov V, Milek M, Ameres S, Chekulaeva M, et al.

Molecular cell (2023) 8315: 2709-2725.e10. . DOTBLOT; tested species: mouse

Identification and comparison of m6A modifications in glioblastoma non-coding RNAs with MeRIP-seq and Nanopore dRNA-seq. Krusnauskas R, Stakaitis R, Steponaitis G, Almstrup K, Vaitkiene P

Epigenetics (2023): 1-14. . MERIP; tested species: human

Downregulation of Fat Mass and Obesity-Related Protein in the Anterior Cingulate Cortex Participates in Anxiety- and Depression-Like Behaviors Induced by Neuropathic Pain.

Wang XL, Wei X, Yuan JJ, Mao YY, Wang ZY, Xing N, Gu HW, Lin CH, Wang WT, Zhang W, Xing F, et al.

Frontiers in cellular neuroscience (2022) 16: 884296. . MERIP; tested species: mouse

Access the online factsheet including applicable protocols at https://sysy.com/product/202111 or scan the QR-code.



FAQ - How should I store my antibody?

Shipping Conditions

 All our antibodies and control proteins / peptides are shipped lyophilized (vacuum freezedried) and are stable in this form without loss of quality at ambient temperatures for several weeks.

Storage of Sealed Vials after Delivery

- Unlabeled and biotin-labeled antibodies and control proteins should be stored at 4°C before reconstitution. They must not be stored in the freezer when still lyophilized!
 Temperatures below zero may cause loss of performance.
- Fluorescence-labeled antibodies should be reconstituted immediately upon receipt. Long term storage (several months) may lead to aggregation.
- **Control peptides** should be kept at -20°C before reconstitution.

Long Term Storage after Reconstitution (General Considerations)

- The storage freezer must not be of the frost-free variety ("no-frost freezer"). This cycle
 between freezing and thawing (to reduce frost-build-up), which is exactly what should be
 avoided. For the same reason, antibody vials should be placed in an area of the freezer that
 has minimal temperature fluctuations, for instance towards the back rather than on a door
 shelf.
- Aliquot the antibody and store frozen (-20°C to -80°C). Avoid very small aliquots (below 20 µl)
 and use the smallest storage vial or tube possible. The smaller the aliquot, the more the stock
 concentration is affected by evaporation and adsorption of the antibody to the surface of the
 storage vial or tube. Adsorption of the antibody to the surface leads to a substantial loss of
 activity.
- The addition of glycerol to a final concentration of 50% lowers the freezing point of your stock and keeps your antibody at -20°C in liquid state. This efficiently avoids freeze and thaw cycles.

Product Specific Hints for Storage

Control proteins / peptides

Store at -20°C to -80°C.

Monoclonal Antibodies

- Ascites and hybridoma supernatant should be stored at -20°C up to -80°C. Prolonged storage at 4°C is not recommended! Unlike serum, ascites may contain proteases that will degrade the antibodies.
- **Purified IgG** should be stored at -20°C up to -80°C. Adding a carrier protein like BSA will increase long term stability. Many of our antibodies already contain carrier proteins. Please refer to the data-sheet for detailed information.

Polyclonal Antibodies

- Crude antisera: With anti-microbials added, they may be stored at 4°C. However, frozen storage (-20°C up to -80°C) is preferable.
- Affinity purified antibodies: Less robust than antisera. Storage at -20°C up to -80°C is
 recommended. Adding a carrier protein like BSA will increase long term stability. Most of our
 antibodies already contain carrier proteins. Please refer to the data-sheet for detailed
 information.

Fluorescence-labeled Antibodies

• Store as a liquid with 1:1 (v/v) glycerol at -20°C. Protect these antibodies from light exposure.

Avoid repeated freeze-thaw cycles for all antibodies!

FAQ - How should I reconstitute my antibody?

Reconstitution

- All our purified antibodies are lyophilized from PBS. To reconstitute the antibody in PBS, add
 the amount of deionized water given in the respective datasheet. If higher volumes are
 preferred, add water as mentioned above and then the desired amount of PBS and a
 stabilizing carrier protein (e.g. BSA) to a final concentration of 2%. Some of our antibodies
 already contain albumin. Take this into account when adding more carrier protein.
 For complete reconstitution, carefully remove the lid. After adding water, briefly vortex the
 solution. You can spin down the liquid by placing the vial into a 50 ml centrifugation tube filled
 with paper.
- If desired, add small amounts of azide or thimerosal to prevent microbial growth. This is especially recommended if you want to keep an aliquot a 4°C.
- After reconstitution of fluorescence-labeled antibodies, add 1:1 (v/v) glycerol to a final
 concentration of 50%. This lowers the freezing point of your stock and keeps your antibody in
 liquid state at -20°C.
- Glycerol may also be added to unlabeled primary antibodies. It is a suitable way to avoid freezethaw cycles.
- Please refer to our tips and hints for subsequent storage of reconstituted antibodies and control peptides and proteins.