SYSY Antibodies

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m3G-cap, m7G-cap

Cat.No. 201 001; Monoclonal mouse antibody, 100 µl ascites (lyophilized)

Data Sheet

Reconstitution/ Storage	100 µl ascites, lyophilized. For **reconstitution** add 100 µl H_2O, then aliquot and store at -20°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	**WB**: not recommended **IP**: yes **IHC**: not tested yet **IHC-P (FFPE)**: not tested yet
Clone	H20
Subtype	IgG1 (κ light chain)
Immunogen	Synthetic m_3G-cap conjugated to human serum albumin.
Reactivity	Reacts with: human, rat, mouse, eukaryotes. Other species not tested yet.
Specificity	Recognizes m_3G-cap and m^7G-cap.
Remarks	This antibody can be used to detect capped RNAs (e.g. in viruses) or to identify and purify proteins associated with capped RNAs (see reference #2).

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

Background

Polymerase II transcripts contain a 5´-terminal **m^7G-cap** that is required for the export of these transcripts from the nucleus to the cytoplasm and eucaryotic translation initiation. The Polymerase II transcribed spliceosomal snRNAs U1, U2, U4 and U5 assemble with the eight Sm proteins B/B´, D1, D2, D3, E, F, and G thus forming a core-UsnRNP. The core-UsnRNP is recognized by a methyltransferase that introduces two additional methyl groups to the m^7G-cap thus forming the **m_3G-cap** (hypermethylation). The m_3G-cap forms one part of the bipartite nuclear localisation signal (NLS) of the UsnRNPs. It is thus necessary for the nuclear re-import of the core-UsnRNPs. Also certain snoRNAs that are involved in the processing of pre-rRNAs contain an m_3G-cap.

Selected References for 201 001

mRNA Capping by Venezuelan Equine Encephalitis Virus nsP1: Functional Characterization and Implications for Antiviral Research.
Li C, Guillén J, Rabah N, Blanjoie A, Debart F, Vasseur JJ, Canard B, Decroly E, Coutard B
Journal of virology (2015) 8916: 8292-303. . **WB**

RNA-methylation-dependent RNA processing controls the speed of the circadian clock.
Fustin JM, Doi M, Yamaguchi Y, Hida H, Nishimura S, Yoshida M, Isagawa T, Morioka MS, Kakeya H, Manabe I, Okamura H, et al.
Cell (2013) 1554: 793-806. . **IP**

MAPCap allows high-resolution detection and differential expression analysis of transcription start sites.
Bhardwaj V, Semplicio G, Erdogdu NU, Manke T, Akhtar A
Nature communications (2019) 101: 3219. . **IP; tested species: drosophila**

Microprocessor mediates transcriptional termination of long noncoding RNA transcripts hosting microRNAs.
Dhir A, Dhir S, Proudfoot NJ, Jopling CL
Nature structural & molecular biology (2015) 224: 319-27. . **IP**

XRN1 stalling in the 5' UTR of Hepatitis C virus and Bovine Viral Diarrhea virus is associated with dysregulated host mRNA stability.
Moon SL, Blackinton JG, Anderson JR, Dozier MK, Dodd BJ, Keene JD, Wilusz CJ, Bradrick SS, Wilusz J
PLoS pathogens (2015) 113: e1004708. . **IP**

Noncoding RNAs and LRRFIP1 regulate TNF expression.
Shi L, Song L, Fitzgerald M, Maurer K, Bagashev A, Sullivan KE
Journal of immunology (Baltimore, Md. : 1950) (2014) 1927: 3057-67. . **IP; tested species: human**

Stress-induced lncRNAs evade nuclear degradation and enter the translational machinery.
Galipon J, Miki A, Oda A, Inada T, Ohta K
Genes to cells : devoted to molecular & cellular mechanisms (2013) 185: 353-68. . **IP; tested species: fission yeast**

The eIF4E-binding protein Eap1p functions in Vts1p-mediated transcript decay.
Rendl LM, Bieman MA, Vari HK, Smibert CA
PloS one (2012) 710: e47121. . **IP**

Identification of a cytoplasmic complex that adds a cap onto 5'-monophosphate RNA.
Otsuka Y, Kedersha NL, Schoenberg DR
Molecular and cellular biology (2009) 298: 2155-67. . **IP**

Induced ncRNAs allosterically modify RNA-binding proteins in cis to inhibit transcription.
Wang X, Arai S, Song X, Reichart D, Du K, Pascual G, Tempst P, Rosenfeld MG, Glass CK, Kurokawa R
Nature (2008) 4547200: 126-30. . **IP**

The interaction between cap-binding complex and RNA export factor is required for intronless mRNA export.
Nojima T, Hirose T, Kimura H, Hagiwara M
The Journal of biological chemistry (2007) 28221: 15645-51. . **IP**

Access the online factsheet including applicable protocols at https://sysy.com/product/201001 or scan the QR-code.

FAQ - How should I store my antibody?

Shipping Conditions

- All SYSY antibodies and control proteins/peptides are shipped lyophilized (vacuum freeze-dried). In this form, they remain stable 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. **Do not freeze lyophilized antibodies**. Temperatures below 0°C may impair performance.
- **Fluorescence-labeled antibodies** should be reconstituted immediately upon receipt. Long-term storage of lyophilized fluorophore-conjugates may cause aggregation.
- **Control peptides** should be stored at -20°C before reconstitution.

Long Term Storage after Reconstitution (General Considerations)

- **Do not use frost-free ("no-frost") freezers.** These units periodically warm to remove ice buildup, causing freeze–thaw cycles that can damage antibodies.
- Store vials in areas with minimal temperature fluctuation - preferably toward the back of the freezer, not on the door.
- Aliquot reconstituted antibodies and store at –20°C to –80°C.
- Avoid very small aliquots (<20 µL), as evaporation and adsorption to tube surfaces can reduce antibody concentration and activity.
- Use the smallest practical storage vial to minimize surface area.
- Adding glycerol to a final concentration of 50% prevents freezing at -20°C, allowing storage in liquid form and effectively avoiding freeze–thaw cycles.

Product Specific Hints for Storage

Control proteins / peptides

- Store at -20°C to -80°C

Monoclonal Antibodies

- **Ascites and hybridoma supernatant:** Store at -20°C to -80°C
  Prolonged storage at 4°C is not recommended, as proteases present in ascites may degrade antibodies.
- **Purified IgG:** Store at -20°C to -80°C
  Adding a carrier protein (e.g., BSA) enhances long-term stability.
  Many SYSY antibodies already contain carrier proteins - refer to the respective datasheet for details.

Polyclonal Antibodies

- **Crude antisera:** Can be stored at 4°C with antimicrobials added, but -20°C to -80°C is preferred
- **Affinity-purified antibodies:** Less stable than antisera; store at -20°C to -80°C
  Adding a carrier protein such as BSA improves long-term stability.
  Most SYSY antibodies already contain carrier proteins - refer to the respective datasheet for details.

Fluorescence-labeled Antibodies

- Store as a liquid with 1:1 (v/v) glycerol at -20°C, and protect from light exposure

Avoid repeated freeze-thaw cycles for all antibodies!

FAQ - How should I reconstitute my antibody?

Reconstitution

- All purified SYSY antibodies are lyophilized from PBS. To reconstitute the antibody in PBS, add the volume of deionized water specified in the corresponding datasheet. If a larger final volume is desired, first add the recommended amount of water, then adjust with PBS and, if needed, add a stabilizing carrier protein (e.g., BSA) to a final concentration of 2%. Some SYSY antibodies already contain albumin; please take this into account before adding additional carrier protein.

For complete reconstitution, carefully remove the vial cap. After adding water, briefly vortex the solution. To collect the liquid at the bottom of the vial, place the vial inside a 50 ml centrifuge tube padded with paper and centrifuge briefly.

- If desired, small amounts of azide or thimerosal may be added to prevent microbial growth. This is particularly recommended when storing an aliquot at 4°C.
- After reconstitution of fluorescence-labeled antibodies, add glycerol 1:1 (v/v) to achieve a final concentration of 50%. This prevents freezing at –20°C and keeps the antibody in liquid form, effectively avoiding freeze–thaw cycles.
- Glycerol may also be added to unlabeled primary antibodies as a general measure to prevent freeze–thaw damage.
- For further guidance, please refer to our **storage tips** and recommendations for reconstituted antibodies, control peptides, and control proteins.