SYSY Antibodies

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NSF

Cat.No. 123 011; Monoclonal mouse antibody, 100 µg purified IgG (lyophilized)

Data Sheet

Reconstitution/ Storage	100 µg purified IgG, lyophilized. For **reconstitution** add 100 µl H_2O 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	**WB**: 1 : 1000 (AP staining) **IP**: yes **ICC**: 1 : 500 **IHC**: not recommended **IHC-P (FFPE)**: 1 : 1000 **ELISA**: yes (see remarks)
Clone	83.11
Subtype	IgG1
Immunogen	Recombinant protein corresponding to AA 1 to 744 from rat NSF (UniProt Id: Q9QUL6)
Reactivity	Reacts with: rat (Q9QUL6), mouse (P46460). No signal: zebrafish. Other species not tested yet.
Remarks	**ELISA**: The ELISA-protocol for membrane proteins is required. Suitable as capture antibody for sandwich-ELISA. Please refer to the protocol for suitable detector antibodies.

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

Background

N-ethylamide **s**ensitive **f**usion protein **NSF** functions together with SNAPs (soluble NSF attachment proteins) and SNAREs (SNAP receptors) in vesicular transport.
NSF is a homotrimer whose polypeptide subunits are made up of three distinct domains: an amino - terminal domain (N) and two homologous ATP-binding domains (D1 and D2). NSF is an ATPase that dissociates SNARE complexes, such as the core complex composed of synaptobrevin/VAMP, syntaxin 1 and SNAP 25 under ATP hydrolysis. The ability of the D1 domain to hydrolyze ATP is required for NSF activity. The D2 domain is required for trimerization, but its ability to hydrolyze ATP is not absolutely required for NSF function.

Selected References for 123 011

LRRK2 G2019S kinase activity triggers neurotoxic NSF aggregation.
Pischedda F, Cirnaru MD, Ponzoni L, Sandre M, Biosa A, Carrion MP, Marin O, Morari M, Pan L, Greggio E, Bandopadhyay R, et al.
Brain : a journal of neurology (2021) 1445: 1509-1525. . **WB, ICC, IHC; tested species: mouse**

Autistic-Like Behavior and Impairment of Serotonin Transporter and AMPA Receptor Trafficking in N-Ethylmaleimide Sensitive Factor Gene-Deficient Mice.
Xie MJ, Iwata K, Ishikawa Y, Nomura Y, Tani T, Murata K, Fukazawa Y, Matsuzaki H
Frontiers in genetics (2021) 12: 748627. . **WB, IHC; KD verified; tested species: mouse**

Synapsin Condensates Recruit alpha-Synuclein.
Hoffmann C, Sansevrino R, Morabito G, Logan C, Vabulas RM, Ulusoy A, Ganzella M, Milovanovic D
Journal of molecular biology (2021) 43312: 166961. . **WB; tested species: rat**

Synaptic vesicle glycoprotein 2A (SV2A) regulates kindling epileptogenesis via GABAergic neurotransmission.
Tokudome K, Okumura T, Shimizu S, Mashimo T, Takizawa A, Serikawa T, Terada R, Ishihara S, Kunisawa N, Sasa M, Ohno Y, et al.
Scientific reports (2016) 6: 27420. . **WB**

CSPα knockout causes neurodegeneration by impairing SNAP-25 function.
Sharma M, Burré J, Bronk P, Zhang Y, Xu W, Südhof TC
The EMBO journal (2012) 314: 829-41. . **WB; tested species: mouse**

Endosomal sorting of readily releasable synaptic vesicles.
Hoopmann P, Punge A, Barysch SV, Westphal V, Bückers J, Opazo F, Bethani I, Lauterbach MA, Hell SW, Rizzoli SO
Proceedings of the National Academy of Sciences of the United States of America (2010) 10744: 19055-60. . **ICC**

Selected General References

Mechanisms of synaptic vesicle exocytosis.
Lin RC et al. Annu. Rev. Cell Dev. Biol. (2000) PubMed:11031229

Neurotransmitter release - four years of SNARE complexes.
Hanson PI et al. Curr. Opin. Neurobiol. (1997) PubMed:9232812

Structure and conformational changes in NSF and its membrane receptor complexes visualized by quick-freeze/deep-etch electron microscopy.
Hanson PI et al. Cell (1997) PubMed:9267032

The synaptic vesicle cycle: a cascade of protein-protein interactions.
Südhof TC et al. Nature (1995) PubMed:7791897

N-ethylmaleimide-sensitive fusion protein: a trimeric ATPase whose hydrolysis of ATP is required for membrane fusion.
Whiteheart SW et al. J. Cell Biol. (1994) PubMed:8051214

Access the online factsheet including applicable protocols at https://sysy.com/product/123011 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.