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**: 1 : 1000 **ELISA**: yes
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 our antibodies and control proteins / peptides are shipped lyophilized (vacuum freeze-dried) 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 freeze-thaw cycles.
- Please refer to our **tips and hints for subsequent storage** of reconstituted antibodies and control peptides and proteins.