# SYSY HistoSure

Rudolf-Wissell-Str. 28a 37079 Göttingen, Germany Phone: +49 551-50556-0 Fax: +49 551-50556-384 E-mail: sales@sysy.com Web: sysy-histosure.com

# **iNOS** mouse specific

Cat.No. HS-484 017; Monoclonal rat antibody, 100 µg purified IgG (lyophilized)

## Data Sheet

Reconstitution/ Storage	100 $\mu$ g purified IgG, lyophilized. Albumin and azide were added for stabilization. For <b>reconstitution</b> add 100 $\mu$ l H <sub>2</sub> 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	WB: 1 : 1000 (AP staining) IP: not tested yet ICC: not tested yet IHC: not tested yet IHC-P: 1 : 200 up to 1 : 1000
Clone	SY-171D8E8
Subtype	IgG2b (λ light chain)
Immunogen	synthetic peptide corresponding to residues near the carboxy terminus of mouse iNOS (UniProt Id: P29477)
Reactivity	Reacts with: mouse (P29477). No signal: rat, human. Other species not tested yet.

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

### Background

Inducible nitric oxide synthase (iNOS) belongs together with endothelial NOS (eNOS) and neuronal NOS (nNOS) to the NOS family of enzymes catalyzing the production of nitric oxide (NO) from Larginine. In contrast to the constitutively expressed calcium-dependent eNOS and nNOS isoforms, iNOS is calcium-independent and usually activated in defense responses such as infection and inflammation (1). Expression of the iNOS gene is induced in vitro in macrophages polarized to the proinflammatory M1 state (2). NO-producing tumoricidal macrophages in the tumor microenvironment can modulate immune responses and promote apoptosis of tumor cells (2). In the brain, iNOS is normally not expressed except in chronic and acute inflammatory conditions. In ischemic brain injury, iNOS was found to be highly expressed in neutrophils and endothelium (3) and in M1-like proinflammatory microglia (4).

### **Selected General References**

NO control of mitochondrial function in normal and transformed cells. Tengan CH et al. Biochim Biophys Acta Bioenerg (2017) PubMed:28216426

Macrophage Reprogramming and Cancer Therapeutics: Role of iNOS-Derived NO. Kashfi K et al. Cells (2021) PubMed:34831416

Individual in vivo Profiles of Microglia Polarization After Stroke, Represented by the Genes iNOS and Ym1. Collmann FM et al. Front Immunol (2019) PubMed:31214190

Inducible nitric oxide synthase in neutrophils and endothelium contributes to ischemic brain injury in mice. Garcia-Bonilla L et al. J Immunol (2014) PubMed:25038255

Access the online factsheet including applicable protocols at <u>https://sysy-histosure.com/product/HS-484017</u> 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.