Monoclonal anti-Human NIS Antibody

Product Name: Monoclonal anti-Human NIS Antibody
Cat. Number: REA011
Lot Number: REA-IM12
Unit Size: 100 µL
Concentration: 1.2 mg/mL
Storage Buffer: 2M Tris, 0.2M glycine (pH, 7.4)
Isotype: Mouse monoclonal, Affinity-purified
Reactivity: Human sodium iodide symporter

Product Description

Monoclonal antibody was raised against peptide NEDLLFFLGQKE LE corresponding to residues 598–621 of the human sodium iodide symporter (hNIS)[1].

This antibody recognizes 14 residues at the carboxy-terminus of hNIS. Detects both the native and denatured forms of hNIS. This antibody does not detect mouse NIS.

Storage Instructions

This antibody can be stored short term (1-2 weeks) at 2-8°C. For longer term storage, aliquot and store at or below -20°C. Avoid repeated freeze/thaw cycles.

Applications

	Dilution	Notes
Western blotting	1:1,000 - 1:5,000	*see below
Flow Cytometry	1:500	†Permeabilization required
Immunohistochemistry[1]	1:5,000-10,000	†Antigen retrieval required (paraffin)

*For western blotting of NIS proteins, it is recommended that samples be heated at 37°C for 30 minutes prior to loading for SDS-PAGE (do not boil).

†This antibody recognizes the cytosolic C-terminus of hNIS. Therefore, samples must be permeabilized prior to incubation with anti-human NIS antibody for immunohistochemistry and flow cytometry.

Recommended Controls

Cells expressing human NIS should be used as a positive control. For western blotting, lysates should be prepared from cells transduced with lentivirus encoding human NIS (Imanis #LV001 or LV002) or stably expressing high-levels of human NIS (Imanis #CL001). Normal human thyroid tissue can be used as a positive control for immunohistochemistry.

Recommended Protocol: Protein Extraction

To prepare membrane protein fractions, harvest and homogenize cells at 4°C in homogenizing buffer (10 mM Tris-HCl, pH7.5, 5 mM NaCl, 1 mM EDTA, 0.25 M sucrose, and 1X protease inhibitor). Clarify lysates at 700 x *g* for 10 min (4°C). Centrifuge the recovered supernatant at 200,000 x *g* for 1 h (4°C). Resuspend the pellet in homogenizing buffer and store at -70°C.

To prepare total protein fractions, lyse cells in RIPA buffer containing 1X protease inhibitors. Incubate on ice for 30 min then clarify at 8,000 x *g* for 15 min (4°C). Store at -70°C.

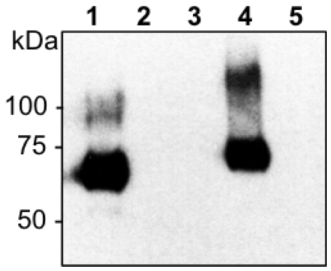
Product Citations:

[1] Tazebay et al. Nature Medicine. 2000. 8:871-878.

Certificate of Analysis

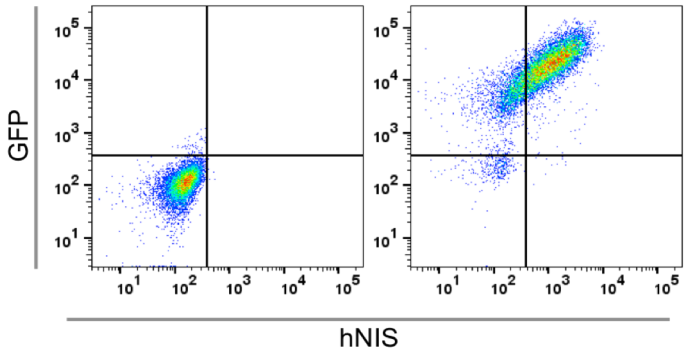
Testing performed by Imanis Life Sciences.

Western blotting



Membrane protein (10 µg; lanes 1-2) or total protein (50 µg; lanes 3-5) was subjected to SDS-PAGE and immunoblotting using monoclonal anti-hNIS antibody (1:3000) and HRP-conjugated anti-mouse secondary antibody (1:5000). Samples: humans NIS (lanes 1 and 4); murine NIS (lanes 2 and 5); negative control lysate (lane 3). Various glycosylated forms of human NIS are detected.

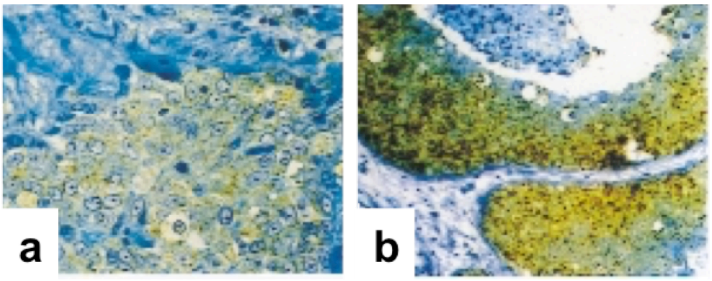
Flow cytometry



(**Left**) Parental HeLaH1 cells or (**Right**) HeLaH1 cells stably expressing hNIS linked to enhanced green fluorescent protein (GFP) were fixed, permeabilized, and stained with monoclonal anti-hNIS antibody (1:500) followed by an Alexa Fluor 594-conjugated anti-mouse secondary antibody (1:1000). Samples were analyzed by flow cytometry.

Immunohistochemistry

Detection of hNIS by immunohistochemistry was performed by Tazebay et al. 2000[1].



Tissue sections were deparafinized. Slides were subjected to antigen retreival using 10% citrate buffer. **a**, NIS expression of normal ductal–lobular units in the vicinity of breast cancer assessed with monoclonal antibody against NIS is shown above (magnification, X160). **b,** Ductal carcinoma stained with monoclonal antibody against NIS (magnification, x66).

Quality Control by: TS
Quality Assurance by: JKM
Effective Date: 30 Jan 2017

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