Datasheet for ABIN2786494

anti-SPNS2 antibody (N-Term)

1 Validation

1 Image

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Overview

Quantity:	100 µL
Target:	SPNS2
Binding Specificity:	N-Term
Reactivity:	Human, Mouse, Rat, Pig, Rabbit
Host:	Rabbit
Clonality:	Polyclonal
Conjugate:	This SPNS2 antibody is un-conjugated
Application:	Western Blotting (WB)

Product Details

Immunogen:	The immunogen is a synthetic peptide directed towards the N terminal region of human SPNS2
Sequence:	PPGTPGTPGC AATAKGPGAQ QPKPASLGRG RGAAAAILSL GNVLNYLDRY
Predicted Reactivity:	Human: 100%, Mouse: 85%, Pig: 100%, Rabbit: 92%, Rat: 93%
Characteristics:	This is a rabbit polyclonal antibody against SPNS2. It was validated on Western Blot using a cell lysate as a positive control.
Purification:	Affinity Purified

Target Details

Target:	SPNS2
Alternative Name:	SPNS2 (SPNS2 Products)

Target Details

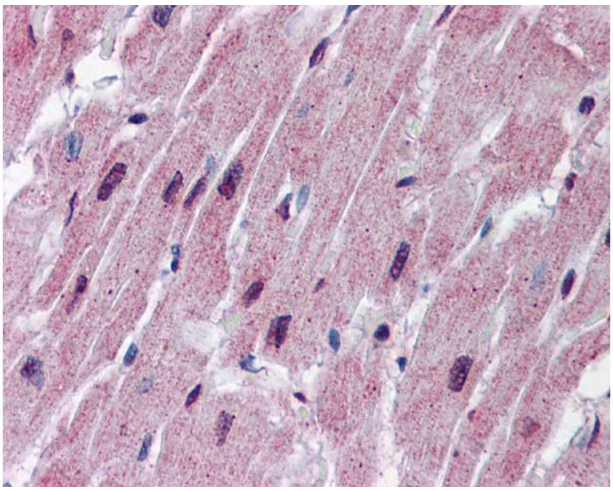
Background:	SPNS2 is the sphingolipid transporter required for migration of myocardial precursors. SPNS2 transports sphingosine 1-phosphate (S1P), a secreted lipid mediator that plays critical roles in cardiovascular, immunological, and neural development and function. SPNS2 mediates the export of S1P from cells in the extraembryonic yolk syncytial layer (YSL), thereby regulating myocardial precursor migration. Alias Symbols: - Protein Size: 549
Molecular Weight:	60 kDa
Gene ID:	124976
NCBI Accession:	NM_001124758, NP_001118230
UniProt:	Q8IVW8

Application Details

Application Notes:	Optimal working dilutions should be determined experimentally by the investigator.
Comment:	Antigen size: 549 AA
Restrictions:	For Research Use only

Handling

Format:	Liquid
Concentration:	Lot specific
Buffer:	Liquid. Purified antibody supplied in 1x PBS buffer with 0.09 % (w/v) sodium azide and 2 % sucrose.
Preservative:	Sodium azide
Precaution of Use:	This product contains Sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.
Handling Advice:	Avoid repeated freeze-thaw cycles.
Storage:	-20 °C
Storage Comment:	For short term use, store at 2-8°C up to 1 week. For long term storage, store at -20°C in small aliquots to prevent freeze-thaw cycles.



Immunohistochemistry
Image 1.

Validation report #101829 for Western Blotting (WB)

Successfully validated (Western Blotting (WB))

by Puissant lab, Institute of Hematology, Inserm U944, CNRS UMR 7212

Report Number: 101829

Date: Feb 01 2018

Target:	SPNS2
Lot Number:	QC66273-42972
Method validated:	Western Blotting (WB)
Positive Control:	Baf-3 cells/ MLL-AF9 murine primary cell model
Negative Control:	Baf-3 or MLL-AF9 murine primary cell model silenced for SPNS2 gene with 2 different shRNA
Notes:	Passed. ABIN2786494 specifically recognizes SPNS2 in murine Baf-3 cells or MLL-AF9 primary cell model.
Primary Antibody:	ABIN2786494
Secondary Antibody:	mouse anti-rabbit HRP conjugated antibody (GE Healthcare, NA9340V)
Protocol:	• Grow Baf-3 or MLL-AF9 cells in RPMI 1640 1x (Gibco, 21875-059) supplemented with 10% FBS (Sigma-Aldrich, F6178, lot 16G148) and 1x Penicillin/Streptomycin (Gibco, 15140-122), at 37°C and 5% CO_2 in 10ml on a flask to 70% confluency. • Transduce $3x10^6$ cells with 35µl of concentrated retrovirus, spin-infection (3h at 37°C to 1200 rpm). • Grow cells for 3d. • Induce and select cells. • Grow cells for 5d. • Sort and collect cells (doxycycline). • Lyse $7x10^5$ cells in 25µl per well in cold SDS Lysis Buffer (Cell Signaling). • Determine total protein content of the lysates using Bradford assay (Bio-Rad, 5000006). • Denature 20µg of total protein for 7min at 95°C in 6µl Laemmli SDS sample buffer and subsequently separate them on a denaturing NuPAGE 4-12% Bis-Tris Protein Gels (Invitrogen,NP0321BOX) in an ELECTROPHORESIS CHAMBER for 2h at 150V. • Transfer proteins onto PVDF membrane (Millipore) with a Western blotting system for 1h 40min at 300mA. • Block the membrane with blocking buffer (2.75% gelatine, 30% BSA, 10mM Tris, NaCl, EDTA, tween 20) for 1h at RT. • Incubation with rabbit anti-SPNS2 antibody (antibodies-online, ABIN2786494, lot QC66273-42972) diluted 1:500 in blocking buffer for 16h at 4°C. • Wash membrane 3x for 10min with tween TBS buffer.

- Incubation with secondary mouse anti-rabbit HRP conjugated antibody (GE Healthcare, NA9340V) diluted 1:5000 in blocking buffer for 1h at RT.
- Wash membrane 3x for 10min with TBST buffer.
- Reveal protein bands using Pierce ECL Western Blotting Substrate (ThermoFisher Scientific, 32106) on a Li-Cor imaging machine.

Experimental Notes:

- The SPNS2 antibody ABIN2786494 reveals a protein of the expected molecular weight for SPNS2 in lysates of murine Baf-3 cells or MLL-AF9 primary cell model.
- ABIN2786494 does show some unspecific binding but the SPNS2 band itself is clear and only visible in the positive but not the negative controls.

Image for Validation report #101829

Validation image no. 1 for anti-Spinster Homolog 2 (SPNS2) (N-Term) antibody (ABIN2786494)

Western blot analysis of SPNS2 expression in mouse Baf-3 (left) and MLL-AF9 cells (right) using ABIN2786494. Cells were transduced either with a control shRNA (control shRNA) or one of two SPNS2 shRNA (shSPNS2_1 and shSPNS2_5).