

Product Datasheet

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Overview

Antigen	SEMA3E
Immunogen	Purified recombinant fragment of Mouse, SEMA3E, corresponding to AA: 26 - 575.
Host/isotype	Rabbit/IgG
Clonality	Recombinant monoclonal
Clone name	IPI-mSEM3E.36
RRID	AB_3740897
IPI ID	TAB0014348-013
Specificity	SEMA3E; Does not cross-react with other SEMAs.
Species reactivity	mouse
Amount	100 µg
Concentration	1 mg/mL
Purification	Expressed in HEK293T cells and affinity purified using Protein A
Storage buffer	PBS, pH 7.4
Shipping	Shipped on blue ice at +4C
Storage	Store at +4C for up to 3 months. For long-term storage, aliquot and store at -20C. Avoid multiple freeze/thaw cycles.

IPI Tested Applications[‡]

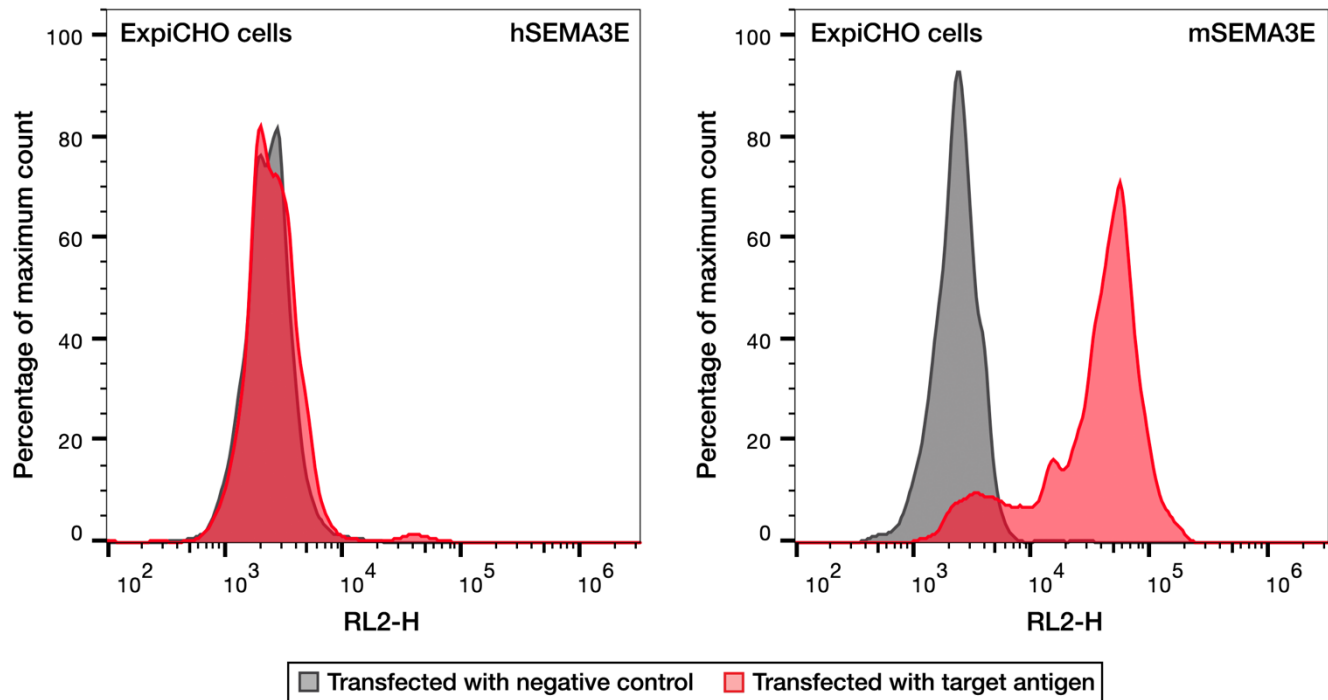
Application	Tested concentration	Result	Reference
Flow	0.66-100 µg/mL	Positive	https://doi.org/10.57733/addgene.aqoodk
IF – Binding	1 µg/mL (1:1000)	Positive	https://doi.org/10.57733/addgene.frmt60
IF – Specificity	1 µg/mL (1:1000)	Positive	https://doi.org/10.57733/addgene.4m50tw

[‡] Not suitable for WB application.

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Applications

Flow cytometry

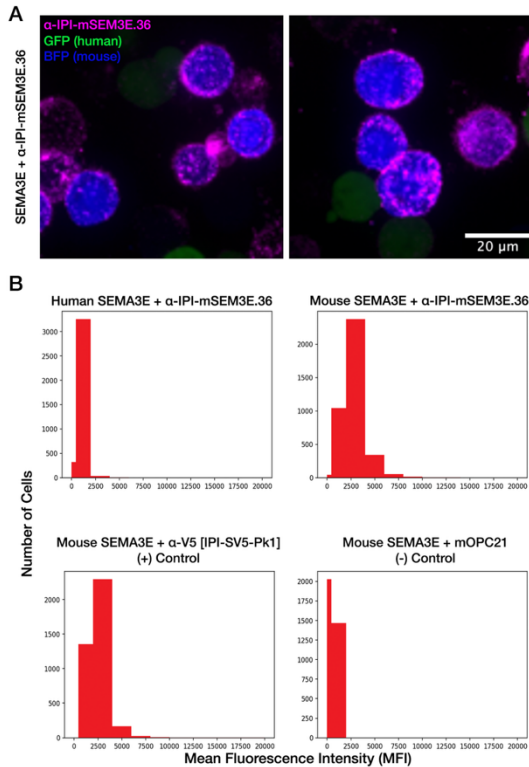


Anti-SEMA3E [IPI-mSEM3E.36] (Addgene #254649) recognizes mouse SEMA3E. Histogram from FACS analysis on ExpiCHO cells transfected with human or mouse SEMA3E (red), or B7H3 negative control (gray). Cells expressing human (left panel) or mouse (right panel) NRXN1b were labeled with Anti-SEMA3E [IPI-mSEM3E.36] and Alexa Fluor 647 F(ab')₂ goat anti-rabbit IgG Fc fragment (Jackson ImmunoResearch, 111-606-046). Labeled cells were analyzed with an Intellicyt iQue Screener Plus flow cytometer. Histograms were generated and normalized to mode using FlowJo™ v10.10. <https://doi.org/10.57733/addgene.aqoodk>

EC₅₀ (data not shown): A fourteen-point titration of antibody concentrations, ranging from 660 nM (0.1 mg/mL) to 4.42 pM with a 1:2.5 dilution factor, against human and mouse SEMA3E showed reactivity towards mouse SEMA3E with observed EC₅₀ values of 15.83 nM (mouse).

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Immunofluorescence (IF) – Species Reactivity

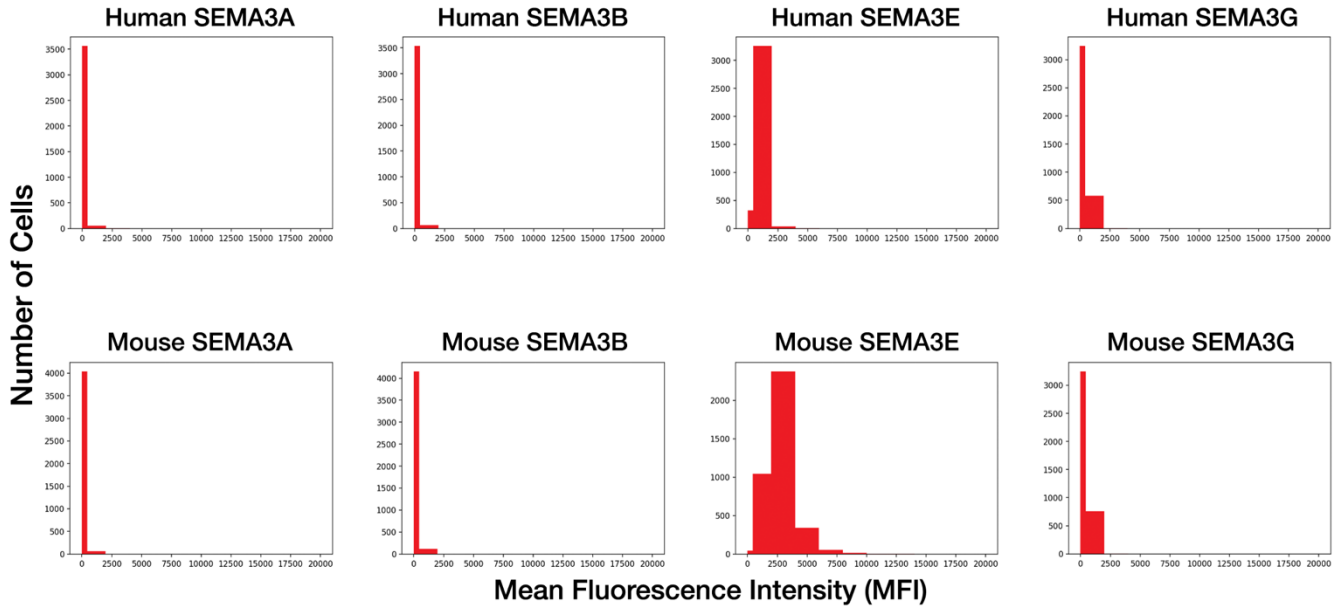


Anti-SEMA3E [IPI-mSEM3E.36] (Addgene#254649) shows binding to mouse SEM3E only. A) Immunofluorescence (IF) of ExpiCHO cells transfected with human (Green/GFP) and mouse (Blue/BFP) SEMA3E stained with IPI-mSEM3E.36 (magenta). Confocal images taken at 40X magnification on the ImageXpress confocal HT.ai microscope. Scale bar = 20 uM. B) Combined quantification of multiple images of the same transfected cells taken at 10X magnification. GFP- or BFP-positive cells were identified via the neural network CellPose, then the mean fluorescence intensity (MFI) of the far-red channel for each cell, representing IPI-mSEM3E.36 staining, was recorded. Each histogram displays the number of cells with MFIs ranging from below 100 (background fluorescence) to 20000. IPI-mSEM3E.36 staining of human and mouse SEMA3E is shown in the top row, and compared to a positive (left) and negative (right) control in the bottom row. For both panels, IPI-mSEM3E.36 was used at 1 ug/mL (1:1000 dilution).

<https://doi.org/10.57733/addgene.frmt60>

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Immunofluorescence (IF) – Target Specificity



		Family Crossreactivity							
		SEM3A		SEM3B		SEM3E		SEM3G	
		Hu	Mo	Hu	Mo	Hu	Mo	Hu	Mo
IPI-mSEM3E.36						++	++	+	+
	Strong	++							
	Weak	+							
	None								

Anti-SEMA3E [IPI-mSEM3E.36] (Addgene#254649) shows significant binding only to mouse SEM3E. Each graph depicts the combined quantification of multiple images of the same transfected cells taken at 10X magnification. GFP or BFP-positive cells were identified via the neural network CellPose, then the mean fluorescence intensity (MFI) of the far-red channel for each cell, representing IPI-mSEM3E.36 staining, was recorded. Each histogram displays the number of cells with MFIs ranging from below 100 (background fluorescence) to 20000. IPI-mSEM3E.36 staining of human and mouse variants of selected SEMA3 family members is compared on the top and bottom rows. To test family-wide cross-reactivity, IPI-mSEM3E.36 was used at 1 ug/mL (1:1000 dilution). <https://doi.org/10.57733/addgene.4m50tw>

Antibody Details

Antibody design and production

Human variable domains for the heavy and light chain of the FAB fragment used in yeast display were grafted onto the constant CH1, CH2 and CH3 domains of rabbit IgG. The chimera human/rabbit IgG1 construct was recombinantly expressed in Expi HEK293 cells, using pTipi2.1 as the expression vector. The antibody was purified by affinity chromatography using protein A (XYZ) and acid elution, followed by immediate buffer exchange using 1 x PBS buffer pH 7.4.

Sequence information

Heavy chain and light chain amino acid sequences are available upon request after purchase. [Contact us](#) to request.

Antibody Characterization

LC-MS: Intact mass analysis via LC-MS methods allows for confirmation antibody mass, and to identify any product-related variants such as glycosylation. Before conducting intact mass analysis via LC-MS, the antibody was reduced to its heavy chain (HC) and light chain (LC). This process allows for confirmation of the masses corresponding to the amino acid sequences of both chains.

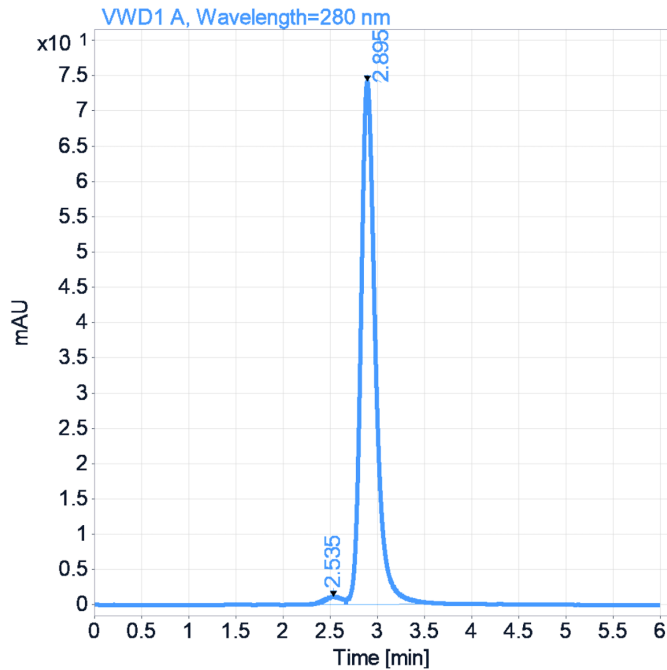
	HC MW (Da) <i>Calculated</i>	HC MW (Da) <i>Observed</i>	HC MW (Da) <i>Delta</i>	LC MW (Da) <i>Calculated</i>	LC MW (Da) <i>Observed</i>	LC MW (Da) <i>Delta</i>
IPI-mSEM3E.36	49840.23	49845.83	5.60	23817.44	23817.07	-0.37

Heavy Chain (HC) Mass Calculation: The calculated molecular weight (MW) of the HC is derived by adding the mass of the unmodified HC amino acid sequence to the mass of the predominant N-glycan form (G0F), which is 1444.5 Da. This calculation assumes that the intrachain disulfide bonds remain intact. For HCs with an N-terminal glutamine (Q), the mass of Q is converted to pyroglutamic acid (PyroGlu), resulting in a deduction of 17.03 Da from the total mass. Additionally, for HCs with a C-terminal lysine (K), the mass of K (128.09 Da) is also subtracted.

Light Chain (LC) Mass Calculation: The calculated molecular weight (MW) of the LC is obtained from the mass of the unmodified LC amino acid sequence, assuming that the intrachain disulfide bonds are not reduced. For LCs with an N-terminal glutamine (Q), the mass of Q is converted to pyroglutamic acid (PyroGlu), leading to a deduction of 17.03 Da from the total mass. For LCs with a C-terminal lysine (K), the mass of K (128.09 Da) is subtracted as well.

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Size Exclusion Chromatography (SEC): 5 μ L of 1 ± 0.2 mg/mL of sample was analyzed with TSKgel® SuperSW mAb HTP (4 μ m) HPLC Column in 1X PBS, pH 7.4 as the mobile phase.



	RT (min)	Width (min)	Area	Height	Area %	Result
IPI-mSEM3E.36	2.90	0.18	795.72	74.09	98.27	Pass

Interpretation: SEC-QC pass. Main peak with greater than 90% purity and within retention time of 2.6 to 3.26 mins.

Antigen Details

Immunogen design:

cDNA of Mouse SEMA3E with C-terminal His-, and FLAG-tags was produced in transiently transfected Expi293F cells and purified from culture supernatant by Ni-NTA affinity purification followed by size-exclusion chromatography.

Immunogen sequences:

>Mouse SEMA3E (AA: 26 - 575):

ANPSYPRLRLSHKELLELNRTSIFQSPLGFLDLHTMLLDEYQERLFGGRDLVYSLNLERVSDGYREIYW
PSTAVKVEECIMKGDANECANYIRVLHHYNRTHLLTCATGAFDPHCAFIRVGGHHSEEPFHLESHRSER
GRGRCPFDPNSSFVSTLVGNELFAGLYSDYWGRDSAIFRSMGKLGHIRTEHDDERLLKEPKFVGSYMIP
DNEDRDDNKMYFFTEKALEAENNAHTIYTRVGRLCVNDMGGQRILVNKWSTFLKARLVCSVPGMNGID
TYFDELEDVFLPTRDPKPNPVIFFGLFNNTTSNIFRGHAVCVYHMSSIREAFNGPYAHKEGPEYHWSLYEGK
VPYPRPGSCASKVNGGKYGTTKDYPDDAIRFARMHPLMYQPIKPVHKKPILVKTGKYNLRQLAVDRVE
AEDGQYDVLFIGTDTGIVLKVITIYNQETEWMEEVILEELQIFKDPAPIISMEISSKRQQLYIGSASAVAQVRF
HHCDMYGSACADCCLARDPYCAWDGISCSTRYPTGAHAKSRFSRQDVRHGNAQQCFGQGGGGSG
GGGSDYKDDDDKSGGLNDIFEAQKIEWHEGSGHHHHHHHH

Sequence information:

HUGO: MGI:1340034
Uniprot: P70275
Refseq: NM_011348.2

Structural information:

Topology: Secreted extracellular protein
PDB IDs: -
AlphaFold: AF-P70275-F1

Expression profiles:

Human Protein Atlas: ENSMUSG00000063531

References

1. Z. Anderson, H. Li, T. Riedel, H. Zhu and D. Moshinsky. (2026). Flow Cytometry for Anti-SEMA3E [IPI-mSEM3E.36]. Addgene. <https://doi.org/10.57733/addgene.aqoodk>
2. A. Morano, T. Riedel, and D. Moshinsky. (2026). ICC/IF for Anti-SEMA3E [IPI-mSEM3E.36] in binding assay. Addgene. <https://doi.org/10.57733/addgene.frmt60>
3. A. Morano, T. Riedel, and D. Moshinsky. (2026). ICC/IF for Anti-SEMA3E [IPI-mSEM3E.36] in specificity assay. Addgene. <https://doi.org/10.57733/addgene.4m50tw>

How to cite this antibody:

Anti-SEMA3E [IPI-mSEM3E.36] - from Institute for Protein Innovation (IPI) (Addgene #254649; <http://n2t.net/addgene:254649>; RRID: AB_3740897).

If you publish research with this product, please [let us know](#) so that we can cite your paper.