

# Product Datasheet

## NK1R Antibody - BSA Free NB300-119

Unit Size: 0.1 ml

Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.

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**NB300-119**

NK1R Antibody - BSA Free

Product Information	
<b>Unit Size</b>	0.1 ml
<b>Concentration</b>	1.0 mg/ml
<b>Storage</b>	Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.
<b>Clonality</b>	Polyclonal
<b>Preservative</b>	0.02% Sodium Azide
<b>Isotype</b>	IgG
<b>Purity</b>	Immunogen affinity purified
<b>Buffer</b>	PBS
<b>Target Molecular Weight</b>	46 kDa

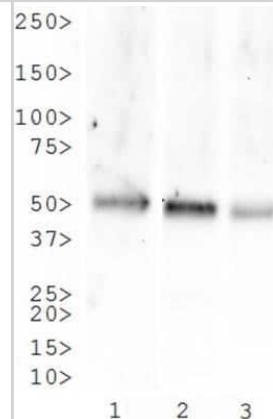
Product Description	
<b>Description</b>	Novus Biologicals Rabbit NK1R Antibody - BSA Free (NB300-119) is a polyclonal antibody validated for use in IHC, WB, Flow and ICC/IF. Anti-NK1R Antibody: Cited in 43 publications. All Novus Biologicals antibodies are covered by our 100% guarantee.
<b>Host</b>	Rabbit
<b>Gene ID</b>	6869
<b>Gene Symbol</b>	TACR1
<b>Species</b>	Human, Mouse, Rat
<b>Reactivity Notes</b>	Mouse reactivity reported from a verified customer review.
<b>Immunogen</b>	A peptide derived from the N-terminus region between amino acids 1-50 of human Neurokinin 1 Receptor. [UniProt# P25103]

Product Application Details	
<b>Applications</b>	Western Blot, Immunohistochemistry-Paraffin, Flow Cytometry, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen
<b>Recommended Dilutions</b>	Western Blot 1-2 ug/ml, Flow Cytometry 1:10 - 1:1000, Immunohistochemistry 10 ug/ml, Immunocytochemistry/ Immunofluorescence 1:100, Immunohistochemistry-Paraffin 10 ug/ml, Immunohistochemistry-Frozen reported in scientific literature (PMID 17604979)
<b>Application Notes</b>	The observed molecular weight of the protein may vary from the listed predicted molecular weight due to post translational modifications, post translation cleavages, relative charges, and other experimental factors.

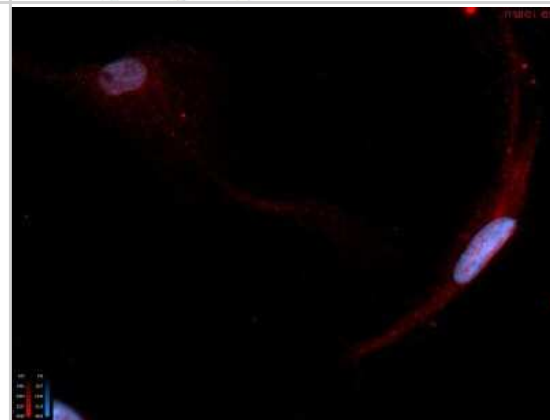


## Images

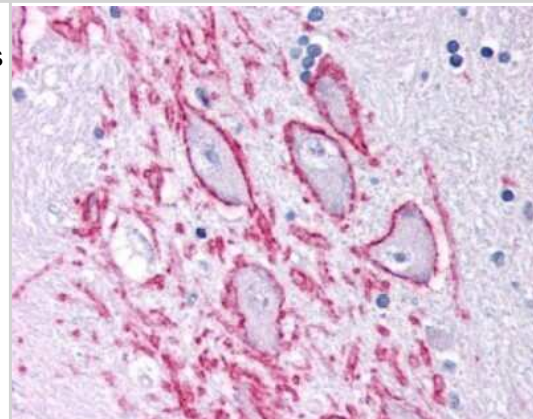
Western Blot: NK1R Antibody [NB300-119] - Neurokinin 1 Receptor Antibody [NB300-119] - Analysis of Neurokinin 1 Receptor in 1: human brain lysate, 2: rat brain lysate and 3: monkey brain lysate.



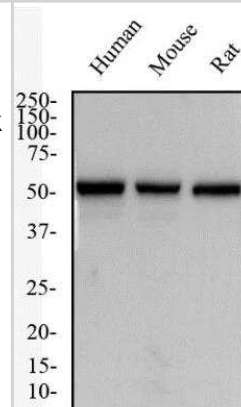
Immunocytochemistry/Immunofluorescence: NK1R Antibody [NB300-119] - Neurokinin 1 Receptor Antibody [NB300-119] - Immunostain of NK1R in rat enteric glial cells. Image from verified customer review.



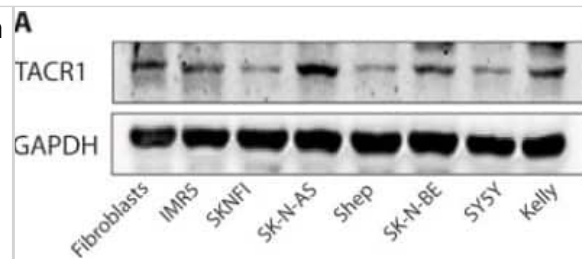
Immunohistochemistry: NK1R Antibody [NB300-119] - Neurokinin 1 Receptor Antibody [NB300-119] - Staining of human brain, basal nucleus of Meynert, neurons.



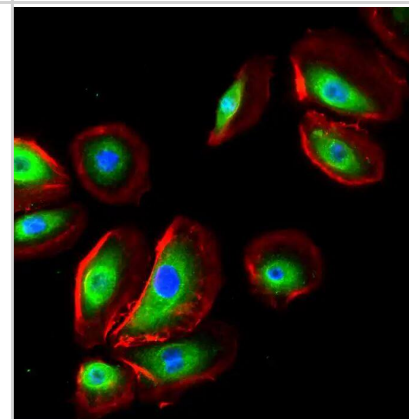
Western Blot: NK1R Antibody [NB300-119] - Total protein from human, mouse and rat brain was separated on a 12% gel by SDS-PAGE, transferred to PVDF membrane and blocked in 5% non-fat milk in TBST. The membrane was probed with 2.0 ug/mL anti-NK1R in 1% non-fat milk in TBST and detected with an anti-rabbit HRP secondary antibody using chemiluminescence.



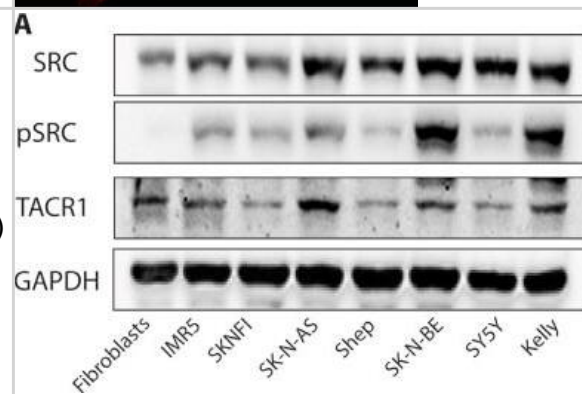
Western Blot: NK1R Antibody [NB300-119] - TACR1 and its downstream targets SRC and p-SRC are expressed in neuroblastoma cell lines. Endogenous TACR1 (NK1R), SRC and p-SRC expression in whole-cell extracts of neuroblastoma cell lines was visualized by western blotting. GAPDH was used as a loading control. Image collected and cropped by CiteAb from the following publication (<https://www.oncotarget.com/lookup/doi/10.18632/oncotarget.13440>), licensed under a CC-BY license.



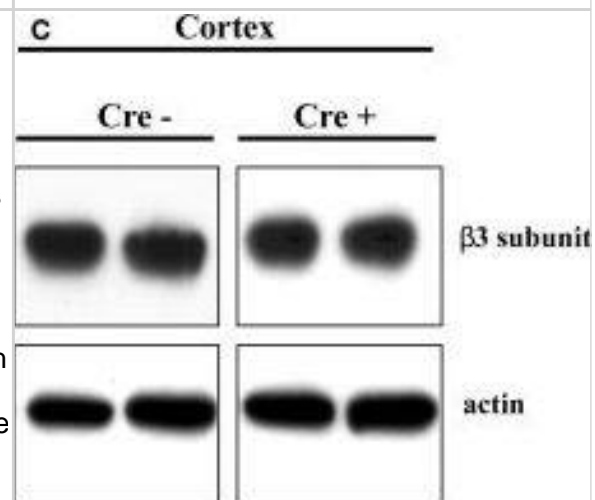
Immunocytochemistry/Immunofluorescence: NK1R Antibody [NB300-119] - Overnight incubation at 4C with NK1R antibody diluted 1:100 in PBS/BSA 1%. Red: Phalloidin, Blue: DAPI, Green: NK1R antibody. Image from verified customer review.



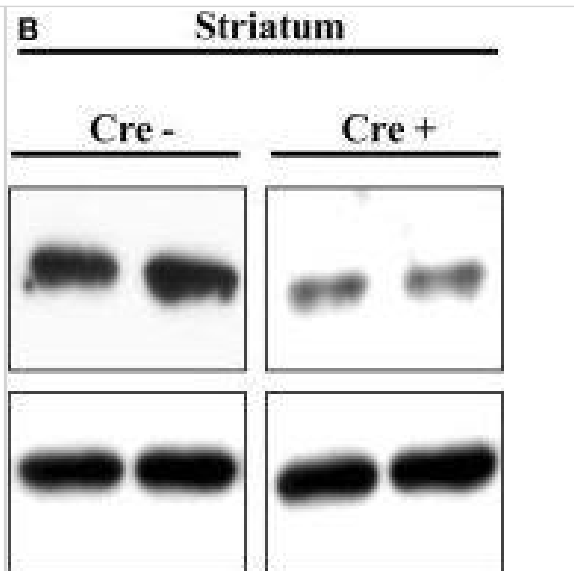
Western Blot: NK1R Antibody - BSA Free [NB300-119] - TACR1 & its downstream targets SRC & p-SRC are expressed in neuroblastoma cell lines (A) Endogenous TACR1, SRC & p-SRC expression in whole-cell extracts of neuroblastoma cell lines was visualized by western blotting. GAPDH was used as a loading control. (B) Quantitative RT-PCR of TACR1 mRNA in a panel of neuroblastoma cell lines ( $n = 3$ , error bars indicate standard deviation). (C–E) Quantification of SRC (C), p-SRC (D) & TACR1 (E) protein expression using densitometry analysis of western immunoblots ( $n = 3$ , error bars indicate standard deviation). Image collected & cropped by CiteAb from the following publication (<https://www.oncotarget.com/lookup/doi/10.18632/oncotarget.13440>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



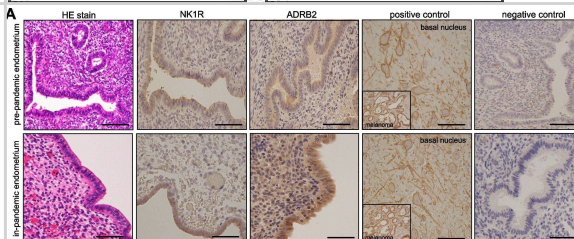
Western Blot: NK1R Antibody - BSA Free [NB300-119] - Striatal  $\beta 3$  subunit protein reduced conditional KO mice. (A) Fluorescent images demonstrating colocalization between endogenous tdTomato fluorescence & DARPP-32 expression. Approximately half of the DARPP-32+ cells expressed endogenous tdTomato, indicative of Cre expression. Scale bar is 25  $\mu\text{m}$ . (B) Representative western blot analysis of the GABA<sub>A</sub> receptor  $\beta 3$  subunit from individual 30-day-old mice revealed reduced amounts of  $\beta 3$  subunit protein in striatum (B) from Cre positive ( $\beta 3^f/fDrd2$ ) animals compared to Cre negative ( $\beta 3^f/+$  &  $\beta 3^f/f$ ) control animals. The amount of  $\beta 3$  protein in cortex (C) did not differ between genotypes. Blots were reprobed for  $\beta$ -actin. (D) Summary graph of western blot analysis demonstrating a significant reduction in  $\beta 3$  protein in striatum, but not cortex. Data are expressed as percent change in band intensity relative to Cre negative controls following normalization to actin. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/21847370>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Western Blot: NK1R Antibody - BSA Free [NB300-119] - Striatal  $\beta 3$  subunit protein reduced conditional KO mice. (A) Fluorescent images demonstrating colocalization between endogenous tdTomato fluorescence & DARPP-32 expression. Approximately half of the DARPP-32+ cells expressed endogenous tdTomato, indicative of Cre expression. Scale bar is 25  $\mu\text{m}$ . (B) Representative western blot analysis of the GABAA receptor  $\beta 3$  subunit from individual 30-day-old mice revealed reduced amounts of  $\beta 3$  subunit protein in striatum (B) from Cre positive ( $\beta 3\text{f}/\text{fDrd}2$ ) animals compared to Cre negative ( $\beta 3\text{f}/+$  &  $\beta 3\text{f}/\text{f}$ ) control animals. The amount of  $\beta 3$  protein in cortex (C) did not differ between genotypes. Blots were reprobed for  $\beta$ -actin. (D) Summary graph of western blot analysis demonstrating a significant reduction in  $\beta 3$  protein in striatum, but not cortex. Data are expressed as percent change in band intensity relative to Cre negative controls following normalization to actin. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/21847370>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



A An image of slides showing the hematoxyline eosin (HE)-stained and immunohistochemical analysis of neurokinin receptor 1 (NK1R)- and  $\beta 2$ -adrenergic receptor (ADRB2)-stained cells in the pre-pandemic (upper row) and in-pandemic endometria (lower row) with corresponding positive controls (basal nucleus, Meynert neurons of human brain tissue for NK1R and melanoma for ADRB2) and negative controls. The immunoreactivities of NK1R B and ADRB2 C are shown as measured by the immunoreactive score (IRS) in pre-pandemic and in-pandemic endometria. The procedure of IRS measurement is described in methods. Mann–Whitney U test indicated that while no significance difference in the endometrial expression of NK1R was observed between the groups, ADRB2 expression in the endometria was significantly higher in the in-pandemic group than that in pre-pandemic group ( $p = 0.015$ ) (C). The boxes represent the interquartile ranges and horizontal lines in the boxes represent median values. Scale bar = 50  $\mu\text{m}$  for each slide. Image collected and cropped by CiteAb from the following open publication (<https://pubmed.ncbi.nlm.nih.gov/37142998>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



## Publications

Ogawa, K;Khan, KN;Koshiba, A;Fujishita, A;Horiguchi, G;Teramukai, S;Itoh, K;Guo, SW;Mori, T; Association between tissue stress reaction and ACE2/TMPRSS2 expression in endometria of reproductive aged women before and during Covid-19 pandemic BMC women's health 2023-05-04 [PMID: 37142998] (Immunohistochemistry, Human)

Zhu E, Liu Y, Zhong M et al. Targeting NK-1R attenuates renal fibrosis via modulating inflammatory responses and cell fate in chronic kidney disease Frontiers in Immunology 2023-03-24 [PMID: 37033943] (Immunohistochemistry, Human)

Guan M, Ying S, Wang Y. Increased expression of transient receptor potential channels and neurogenic factors associates with cough severity in a guinea pig model BMC Pulmonary Medicine 2021-12-01 [PMID: 34078339] (Immunohistochemistry, Human)

Rodriguez E, Pei G, Kim ST et al. Substance P Antagonism as a Novel Therapeutic Option to Enhance Efficacy of Cisplatin in Triple Negative Breast Cancer and Protect PC12 Cells against Cisplatin-Induced Oxidative Stress and Apoptosis Cancers (Basel) 2021-07-31 [PMID: 34359773] (Flow Cytometry)

Zheng Y, Sang M, Liu F et al. Aprepitant inhibits the progression of esophageal squamous cancer by blocking the truncated neurokinin<sup>1</sup> receptor Oncology reports 2023-07-01 [PMID: 37203393] (WB, Human)

Restaino AC, Walz A, Vermeer SJ et al. Functional neuronal circuits promote disease progression in cancer Science advances 2023-05-10 [PMID: 37163587] (WB, IHC, Human)

### Details:

IHC Dilution: 1:500; WB Dilution: 1:500

Brzozowska M, Romaniewicz M, Calka J, Jana B Effects of Substance P and Neurokinin A on the Contractile Activity of Inflamed Porcine Uterus International Journal of Molecular Sciences 2022-10-29 [PMID: 36361972] (WB, Porcine)

Talbot S, Doyle B, et al. Vagal sensory neurons drive mucous cell metaplasia. J Allergy Clin Immunol 2020-06-01 [PMID: 31954778] (FLOW, Mouse)

Xu X, Cai X, Liu X, Guo SW Possible involvement of neuropeptide and neurotransmitter receptors in Adenomyosis Reproductive biology and endocrinology : RB&E 2021-02-19 [PMID: 33602248] (IF/IHC, Human)

Morelli Ae, Sumpter TI, Rojas-Canales Dm et Al. Neurokinin-1 Receptor Signaling Is Required for Efficient Ca<sup>2+</sup> Flux in T-Cell-Receptor-Activated T Cells Cell Rep 2020-03-10 [PMID: 32160549] (WB, Mouse)

Zhou Y, Wang M, Tong Y et al. miR-206 Promotes Cancer Progression by Targeting Full-Length Neurokinin-1 Receptor in Breast Cancer Technol. Cancer Res. Treat. 2019-01-01 [PMID: 31506061] (WB, IF/IHC, Human, Mouse)

Wang Dong, Tawfik Vivianne L, Corder Gregory et al. Functional Divergence of Delta and Mu Opioid Receptor Organization in CNS Pain Circuits. Genome Medicine 2018-04-04 [PMID: 29576387] (IF/IHC, Mouse)

More publications at <http://www.novusbio.com/NB300-119>



## Procedures

### Western Blot Protocol for Neurokinin 1 Receptor Antibody (NB300-119)

#### Western Blot Protocol

1. Perform SDS-PAGE on samples to be analyzed, loading 10-25 ug of total protein per lane.
2. Transfer proteins to PVDF membrane according to the instructions provided by the manufacturer of the membrane and transfer apparatus.
3. Stain the membrane with Ponceau S (or similar product) to assess transfer success, and mark molecular weight standards where appropriate.
4. Rinse the blot TBS -0.05% Tween 20 (TBST).
5. Block the membrane in 5% Non-fat milk in TBST (blocking buffer) for at least 1 hour.
6. Wash the membrane in TBST three times for 10 minutes each.
7. Dilute primary antibody in blocking buffer and incubate overnight at 4C with gentle rocking.
8. Wash the membrane in TBST three times for 10 minutes each.
9. Incubate the membrane in diluted HRP conjugated secondary antibody in blocking buffer (as per manufacturer's instructions) for 1 hour at room temperature.
10. Wash the blot in TBST three times for 10 minutes each (this step can be repeated as required to reduce background).
11. Apply the detection reagent of choice in accordance with the manufacturer's instructions.

### Immunohistochemistry-Paraffin Protocol for NK1R Antibody (NB300-119)

#### Immunohistochemistry-Paraffin Embedded Sections

##### Antigen Unmasking:

Bring slides to a boil in 10 mM sodium citrate buffer (pH 6.0) then maintain at a sub-boiling temperature for 10 minutes. Cool slides on bench-top for 30 minutes (keep slides in the sodium citrate buffer at all times).

##### Staining:

1. Wash sections in deionized water three times for 5 minutes each.
2. Wash sections in PBS for 5 minutes.
3. Block each section with 100-400 ul blocking solution (1% BSA in PBS) for 1 hour at room temperature.
4. Remove blocking solution and add 100-400 ul diluted primary antibody. Incubate overnight at 4 C.
5. Remove antibody solution and wash sections in wash buffer three times for 5 minutes each.
6. Add 100-400 ul HRP polymer conjugated secondary antibody. Incubate 30 minutes at room temperature.
7. Wash sections three times in wash buffer for 5 minutes each.
8. Add 100-400 ul DAB substrate to each section and monitor staining closely.
9. As soon as the sections develop, immerse slides in deionized water.
10. Counterstain sections in hematoxylin.
11. Wash sections in deionized water two times for 5 minutes each.
12. Dehydrate sections.
13. Mount coverslips.



**Immunocytochemistry/ Immunofluorescence Protocol for NK1R Antibody (NB300-119)**

## Immunocytochemistry Protocol

Culture cells to appropriate density in 35 mm culture dishes or 6-well plates.

1. Remove culture medium and wash the cells briefly in PBS. Add 10% formalin to the dish and fix at room temperature for 10 minutes.
2. Remove the formalin and wash the cells in PBS.
3. Permeablize the cells with 0.1% Triton X100 or other suitable detergent for 10 min.
4. Remove the permeablization buffer and wash three times for 10 minutes each in PBS. Be sure to not let the specimen dry out.
5. To block nonspecific antibody binding, incubate in 10% normal goat serum from 1 hour to overnight at room temperature.
6. Add primary antibody at appropriate dilution and incubate overnight at 4C.
7. Remove primary antibody and replace with PBS. Wash three times for 10 minutes each.
8. Add secondary antibody at appropriate dilution. Incubate for 1 hour at room temperature.
9. Remove secondary antibody and replace with PBS. Wash three times for 10 minutes each.
10. Counter stain DNA with DAPI if required.





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### **Products Related to NB300-119**

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NBL1-16672	NK1R Overexpression Lysate
NB300-119PEP	NK1R Antibody Blocking Peptide
NBP2-33376H	Blue Marker Antibody (6F4-F6) [HRP]
HAF008	Goat anti-Rabbit IgG Secondary Antibody [HRP]
NB7160	Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]
NBP2-24891	Rabbit IgG Isotype Control

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### **Limitations**

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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