Product Datasheet

MHC Class I Antibody (OX18) - BSA Free NB120-6405

Unit Size: 0.1 mg

Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.

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NB120-6405

MHC Class I Antibody (OX18) - BSA Free

Product Information	
Unit Size	0.1 mg
Concentration	1.0 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Clone	OX18
Preservative	0.02% Sodium Azide
Isotype	IgG1
Purity	Protein G purified
Buffer	PBS
Product Description	
Host	Mouse
Gene ID	3133
Gene Symbol	HLA-E
Species	Rat
Specificity/Sensitivity	Recognizes a monomorphic determinant of rat MHC Class I (RT1A), expressed by all rat strains. However, quantitative measurements suggest that not all of the class I molecules are recognised.
Immunogen	Rat spleen glycoproteins
Product Application Details	
Applications	ELISA, Electron Microscopy, Flow Cytometry, Flow (Intracellular), Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, Immunoprecipitation, Block/Neutralize, CyTOF-ready
Recommended Dilutions	Flow Cytometry 1:50-1:100, ELISA 1:100-1:2000. Use reported in scientific literature (PMID 2783579), Immunohistochemistry 1:10-1:500, Immunocytochemistry/ Immunofluorescence 1:10-1:500. Use reported in scientific literature (PMID 24678820), Immunoprecipitation 1:10-1:500, Immunohistochemistry-Paraffin 1:10-1:500, Immunohistochemistry-Frozen 1:10-1:500, Electron Microscopy reported in scientific literature (PMID 3044648), Flow (Intracellular), CyTOF-ready, Block/Neutralize reported in scientific literature (PMID 1698855)
Application Notes	The epitope recognized by this anti-rat RT1-A antibody may be sensitive to formaldehyde fixation and tissue processing. If possible, the use of acetone fixation for frozen sections is recommended. This antibody is CyTOF ready.



Images

Immunocytochemistry/Immunofluorescence: MHC Class I Antibody (OX18) [NB120-6405] - Major histocompatibility complex (MHC) I and II as well as Transporter associated with antigen presentation II (TAPII) were analyzed, using immunocytochemistry on rat Schwann cells (SCs). Corresponding merges are shown in the bottom rows. Treatment of SCs with IL-17 was performed at concentrations of 0.5 and 50 ng/mL. Graphs to the right show densitometry quantification. SCs showed expression of MHCI > TAPII > MHCII, which increased after IL-17 treatment. MHCI was mainly detected in the cytoplasm and the expression increased in a dose-dependent manner after IL-17 treatment, significant for 0.5 ng/mL and 50 ng/mL (**P <=0.01). Image collected and cropped by CiteAb from the following publication

(https://jneuroinflammation.biomedcentral.com/articles/10.1186/1742-2094-11-63), licensed under a CC-BY license.

Immunohistochemistry-Paraffin: MHC Class I Antibody (OX18) [NB120-6405] - Analysis of FFPE rat brain cerebellum using MHC Class I (OK18) antibody at 1:200 on a Bond Rx autostainer (Leica Biosystems). The assay involved 20 minutes of heat induced antigen retrieval (HIER) using 10mM sodium citrate buffer (pH 6.0) and endogenous peroxidase quenching with peroxide block. The sections were incubated with primary antibody for 30 minutes and Bond Polymer Refine Detection (Leica Biosystems) with DAB was used for signal development followed by counterstaining with hematoxylin. Whole slide scanning and capturing of representative images was performed using Aperio AT2 (Leica Biosystems). Endothelial staining was observed. Staining was performed by Histowiz.

Flow Cytometry: MHC Class I Antibody (OX18) [NB120-6405] - Analysis using the FITC conjugate of NB120-6405. Staining of rat spleen cells with mouse anti-rat RT1-A (OX18).

Immunocytochemistry/Immunofluorescence: MHC Class I Antibody (OX18) [NB120-6405] - PC-12 cells were fixed for 10 minutes using 10% formalin and then permeabilized for 5 minutes using 1X TBS + 0.5% Triton X-100. The cells were incubated with anti-MHC Class I (OX18) NB120-6405 at a 1:100 dilution overnight at 4C and detected with an anti-mouse DyLight 488 (Green) at a 1:500 dilution. Actin was detected with Phalloidin 568 (Red) at a 1:200 dilution. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective.









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Immunocytochemistry/Immunofluorescence: MHC Class I Antibody (OX18) [NB120-6405] - PC12 cells were fixed for 10 minutes using 4% PFA and then permeabilized for 5 minutes using 1X PBS + 0.05% Triton-X100. The cells were incubated with anti-MHC Class I Antibody (OX18) at 2 ug/ml overnight at 4C and detected with an anti-mouse Dylight 488 (Green) at a 1:500 dilution. Actin was detected with Phalloidin 568 (Red) at a 1:500 dilution. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective.







Flow (Intracellular): MHC Class I Antibody (OX18) [NB120-6405] - PC-12 cells were stained with MHC Class I NB120-6405 (Blue) and a matched isotype control NBP2-27287 (Orange). Cells were fixed with 4% PFA and then permeablized with 0.1% saponin. Cells were incubated in an antibody dilution of 1 ug/mL for 30 minutes at room temperature, followed by DyLight488-conjugated anti-mouse secondary antibody.

Effects of anodal tDCS on protein expression. Representative immunohistochemical images of Ox18 + cells in the SVZ ipsilateral to anodal tDCS or sham stimulation. Staining for Ox18 (MHC I) in the ipsilateral SVZ revealed more Ox18 + cells in animals treated by anodal tDCS (left panel) compared to sham stimulation (right panel) by trend (scale bars = 100 µm). Results are displayed as mean ± SEM.

Major histocompatibility complex (MHC) I and II as well as Transporter associated with antigen presentation II (TAPII) were analyzed, using immunocytochemistry on rat Schwann cells (SCs). Corresponding merges are shown in the bottom rows. Treatment of SCs with IL-17 was performed at concentrations of 0.5 and 50 ng/mL. Graphs to the right show densitometry quantification. SCs showed expression of MHCI > TAPII > MHCII, which increased after IL-17 treatment. (A) MHCI was mainly detected in the cytoplasm and the expression increased in a dose-dependent manner after IL-17 treatment, significant for 0.5 ng/mL and 50 ng/mL (**P <=0.01). Image collected and cropped by CiteAb from the following publication

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Publications

Farahi S, Hosseini S, Ghanbarian H et al. The Use of Trichostatin A during Pluripotent Stem Cell Generation Does Not Affect MHC Expression Level Stem Cells International 2022-02-15 [PMID: 35371264] (FLOW)

Smith TA, Ghergherehchi CL, Mikesh M et al. Polyethylene glycol-fusion repair of sciatic allografts in female rats achieves immunotolerance via attenuated innate and adaptive responses J. Neurosci. Res. 2020-09-15 [PMID: 32931034]

Zhang X, de Oliveira Andrade F, Zhang H et al. Maternal obesity increases offspring\'s mammary cancer recurrence and impairs tumor immune response Endocr. Relat. Cancer 2020-06-01 [PMID: 32580156] (IF/IHC, Mouse)

Rabenstein M, Unverricht-Yeboah M, Keuters MH et al. Transcranial Current Stimulation Alters the Expression of Immune-Mediating Genes Front Cell Neurosci. 2019-10-25 [PMID: 31708742] (IHC-Fr, Rat)

Rada C, Lorenzi R, Powis SJ et al. Concerted evolution of class I genes in the major histocompatibility complex of murine rodents. Proc Natl Acad Sci U S A. 1990-03-01 [PMID: 2315309] (IP, Rat)

Jewtoukoff V, Lebar R, Bach MA. Oligodendrocyte-specific autoreactive T cells using an alpha/beta T-cell receptor kill their target without self restriction. Proc Natl Acad Sci U S A. 1989-04-01 [PMID: 2784860]

Schultzberg M, Olsson T, Samuelsson EB et al. Early major histocompatibility complex (MHC) class I antigen induction in hypothalamic supraoptic and paraventricular nuclei in trypanosome-infected rats. J Neuroimmunol. 1989-09-01 [PMID: 2681260] (IHC-Fr, Rat)

Matsumoto Y, Fujiwara M. In situ detection of class I and II major histocompatibility complex antigens in the rat central nervous system during experimental allergic encephalomyelitis. An immunohistochemical study. J Neuroimmunol. 1986-10-01 [PMID: 3489735]

Sedgwick JD, Hughes CC, Male DK, MacPhee IA, ter Meulen V. Antigen-specific damage to brain vascular endothelial cells mediated by encephalitogenic and nonencephalitogenic CD4+ T cell lines in vitro. J Immunol. 1990-10-15 [PMID: 1698855] (B/N)

Spencer SC, Fabre JW. Identification in rat liver and serum of water-soluble class I MHC molecules possibly homologous to the murine Q10 gene product. J Exp Med. 1987-06-01 [PMID: 3585249]

Hess AD, Horwitz L, Beschorner WE, Santos GW. Development of graft-vs.-host disease-like syndrome in cyclosporine-treated rats after syngeneic bone marrow transplantation. I. Development of cytotoxic T lymphocytes with apparent polyclonal anti-la specificity, including autoreactivity. J Exp Med. 1985-04-01 [PMID: 2580038]

Mayrhofer G, Schon-Hegrad MA. Ia antigens in rat kidney, with special reference to their expression in tubular epithelium. J Exp Med. 1983-06-01 [PMID: 6406641] (IHC-Fr, Rat)

More publications at http://www.novusbio.com/NB120-6405



Procedures

Immunocytochemistry/Immunofluorescence Protocol for MHC Class I Antibody (NB120-6405) 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 NB120-6405

HAF007	Goat anti-Mouse IgG Secondary Antibody [HRP]
NB720-B	Rabbit anti-Mouse IgG (H+L) Secondary Antibody [Biotin]
NBP1-97005-0.5mg	Mouse IgG1 Isotype Control (MG1)
NB120-6405UV	MHC Class I Antibody (OX18) [DyLight 350]

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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