

Product Datasheet

IL-6 Antibody - Azide and BSA Free NB600-1131SS

Unit Size: 0.04 ml

Store at -20C short term. Aliquot and store at -80C long term. Avoid freeze-thaw cycles.

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NB600-1131SS

IL-6 Antibody - Azide and BSA Free

Product Information	
Unit Size	0.04 ml
Concentration	This product is unpurified. The exact concentration of antibody is not quantifiable.
Storage	Store at -20C short term. Aliquot and store at -80C long term. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	No Preservative
Isotype	IgG
Purity	Unpurified
Buffer	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2

Product Description	
Description	Store vial at -20C prior to opening. Aliquot contents and freeze at -20C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4C as an undiluted liquid. Dilute only prior to immediate use.
Host	Rabbit
Gene ID	3569
Gene Symbol	IL6
Species	Human, Mouse, Rat, Monkey
Reactivity Notes	Use in Rat reported in scientific literature (PMID:33651836).
Specificity/Sensitivity	Anti-IL-6 antiserum detects recombinant and native IL-6 present in body fluids and cell supernatants in various assays (ie. IL-1 stimulated IL-6 production from fibroblasts).
Immunogen	This whole rabbit serum was prepared by repeated immunizations with recombinant human IL-6 produced in E.coli. (Uniprot: P05231)

Product Application Details	
Applications	Western Blot, ELISA, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, Immunoprecipitation, Knockdown Validated
Recommended Dilutions	Western Blot 1:500-1:2000, ELISA 1:1000-1:5000, Immunohistochemistry 1:400-1:800, Immunocytochemistry/ Immunofluorescence 1:10 - 1:500, Immunoprecipitation 1:400-1:800, Immunohistochemistry-Paraffin 1:400 - 1:800, Immunohistochemistry-Frozen, Knockdown Validated

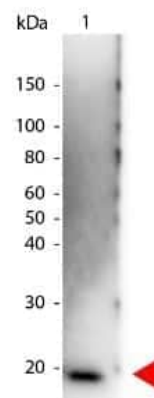


Application Notes

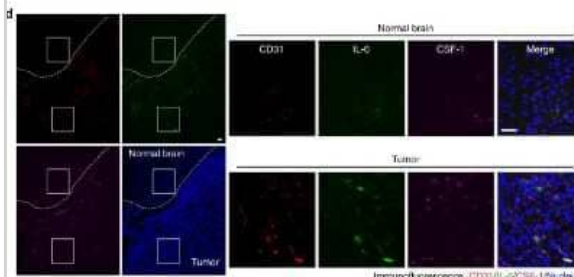
This product has been tested in western blot. This antibody is suitable for use in neutralizations, ELISA, radioimmunoassay, and immunoprecipitation. Reactivity in other immunoassays is unknown. Expect ~23.7kDa. In Western blot analysis of natural cell products or human body fluids, multiple bands of IL-6 will appear due to the variable amount of glycosylation on the molecule. For immunoblot use the supernatants or lysates of 2×10^6 endotoxin-stimulated human peripheral blood mononuclear cells (PBMC). PBMC are stimulated for 24 hours with 1% (v/v) human serum plus 10ng/mL E. coli LPS. For immunoprecipitation, pre-clearing with a non-specific rabbit IgG is helpful to reduce background. The antiserum is useful for neutralization of human IL-6 activity in bioassays. For neutralization, incubate the sample with a 1:400 dilution of the antiserum for at least 4 hours before being tested. A control of similarly diluted normal rabbit IgG (heat inactivated) is recommended. In neutralization experiments in vitro, this antibody does not result in enhanced activity of IL-6. However, because antibodies to IL-6 may act as a soluble receptor in vivo, some antibodies to IL-6 act as carriers and enhance IL-6 activity.

Images

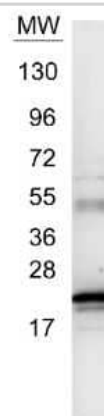
Western Blot: IL-6 Antibody [NB600-1131] - Western Blot: IL-6 Antibody [HRP] [NBP1-42762] - Lane 1: Human IL-6. Lane 2: none. Load: 50 ng per lane. Primary antibody: none. Secondary antibody: Peroxidase Human IL-6 secondary antibody at 1:1,000 for 60 min at RT. Block: incubated with blocking buffer for 30 min at RT. Predicted/Observed size: 20 kDa for Human IL-6. Other band(s): none. Image using the HRP form of this antibody.



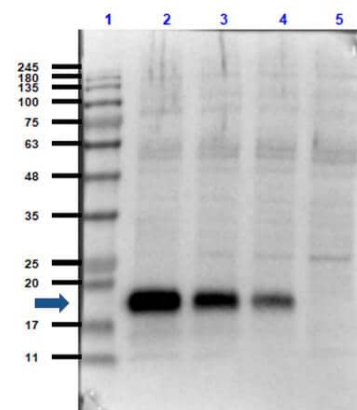
Immunohistochemistry: IL-6 Antibody [NB600-1131] - GBM ECs express IL-6. Mouse GBM was induced by orthotopic injection of GL26 glioma cells into wild-type mouse. The brain sections that include normal brains and tumors were stained with anti-CD31, anti-IL-6, and anti-CSF-1 antibodies. Representative immunofluorescence images are shown. Right, enlarged area in normal and tumor tissues. Bar represents 50? um. Zoom-in factor: 4 Image collected and cropped by CiteAb from the following publication (<https://www.nature.com/articles/s41467-018-03050-0>) licensed under a CC-BY license.



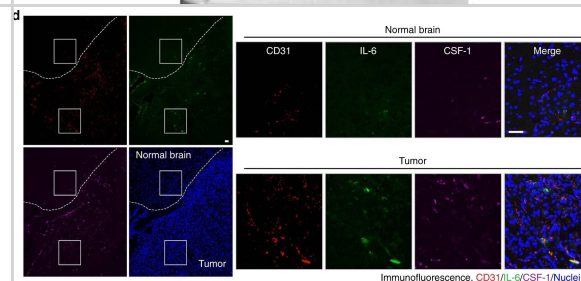
Western Blot: IL-6 Antibody [NB600-1131] - Protein was resolved on a 4-20% Tris-Glycine gel by SDS-PAGE and transferred onto nitrocellulose. The blot shows detection of a band 21 kDa in size corresponding to anti-IL6 antibody. Molecular weight markers are also shown (MW). After transfer, the membrane was blocked for 30 minutes with 1% BSA-TBST. Detection occurred using peroxidase conjugated anti-Rabbit IgG secondary antibody diluted 1:40,000 in blocking buffer for 30 min at RT followed by reaction with FemtoMax chemiluminescent substrate.



Western Blot of IL-6 Antibody. Lane 1: Opal Prestained Molecular Weight Marker



Immunocytochemistry/ Immunofluorescence: IL-6 Antibody [NB600-1131] - GBM ECs express IL-6. a Human brain ECs were treated with glioma-CM for 24 h, & cell lysates were subjected to multiplex cytokine array analysis. Left, a representative blot. Right, quantified dot intensity of most significantly changed cytokines. b Human microvascular brain ECs were treated with glioma-CM that were harvested from different human glioma cells. Cell lysates were immunoblotted. c Human microvascular brain ECs & tumor-associated ECs isolated from different GBM patients were subjected to immunoblot analysis. d Mouse GBM was induced by orthotopic injection of GL26 glioma cells into wild-type mouse. The brain sections that include normal brains & tumors were stained with anti-CD31, anti-IL-6, & anti-CSF-1 antibodies. Representative immunofluorescence images are shown. Right, enlarged area in normal & tumor tissues. Bar represents 50 μ m. Zoom-in factor: 4 Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/29422647>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Publications

Zamani A, EmamiAref P, Kubířková L, Haanová K et Al. Paclitaxel triggers molecular and cellular changes in the choroid plexus Front Pain Res (Lausanne) 2024-12-10 [PMID: 39654799]

Zhou, X;Zhu, Y;Gao, D;Li, M;Lin, L;Wang, Z;Du, H;Xu, Y;Liu, J;He, Y;Guo, Y;Wang, S;Qiao, S;Bao, Y;Liu, Y;Zhang, H; Matrilin-3 supports neuroprotection in ischemic stroke by suppressing astrocyte-mediated neuroinflammation Cell reports 2024-03-21 [PMID: 38520693]

Jeong HY, Park JS, Choi JW et Al. GRIM-19-mediated induction of mitochondrial STAT3 alleviates systemic sclerosis by inhibiting fibrosis and Th2/Th17 cells Exp Mol Med 2024-12-06 [PMID: 39643607]

Tran, TT;Lee, G;Huh, YH;Chung, KH;Lee, SY;Park, KH;Kwon, SH;Kook, MS;Chun, JS;Koh, JT;Ryu, JH; Disruption of cholesterol homeostasis triggers periodontal inflammation and alveolar bone loss Experimental & molecular medicine 2023-12-01 [PMID: 38036731]

Su-Jin Kim, Se Hui Lee, Binh Do Quang, Thanh-Tam Tran, Young-Gwon Kim, Jun Ko, Weon-Young Choi, Sun Young Lee, Je-Hwang Ryu Avenanthramide-C Shows Potential to Alleviate Gingival Inflammation and Alveolar Bone Loss in Experimental Periodontitis Molecules and Cells 2023-10-31 [PMID: 37641936]

Bai H, Liu X, Lin M et Al. Progressive senescence programs induce intrinsic vulnerability to aging-related female breast cancer Nat Commun 2024-06-17 [PMID: 38886378]

Jeong HY, Park JS, Woo JS et Al. SARS-CoV-2 spike protein accelerates systemic sclerosis by increasing inflammatory cytokines, Th17 cells, and fibrosis J Inflamm (Lond) 2023-12-21 [PMID: 38129904]

Moon J, Cho KH, Jhun J et Al. Small heterodimer partner-interacting leucine zipper protein suppresses pain and cartilage destruction in an osteoarthritis model by modulating the AMPK/STAT3 signaling pathway Arthritis Res Ther 2024-11-13 [PMID: 39533324]

T. Ollewagen, R.M. Benecke, C. Smith High species homology potentiates quantitative inflammation profiling in zebrafish using immunofluorescence Heliyon 2023-12-13 [PMID: 38187273]

KW Zhang, D Wang, H Cai, MQ Cao, YY Zhang, PY Zhuang, J Shen IL-6 plays a crucial role in epithelial-mesenchymal transition and pro-metastasis induced by sorafenib in liver cancer Oncology reports, 2021-01-07;0(0):. 2021-01-07 [PMID: 33432366]

Ashraf-Uz-Zaman M, Ji G, Tidwell D et al. Evaluation of Urea-Based Inhibitors of the Dopamine Transporter Using the Experimental Autoimmune Encephalomyelitis Model of Multiple Sclerosis ACS chemical neuroscience 2022-01-19 [PMID: 34978174]

Park JS, Yang S, Song D et al. A newly developed PLD1 inhibitor ameliorates rheumatoid arthritis by regulating pathogenic T and B cells and inhibiting osteoclast differentiation Immunology letters 2023-09-16 [PMID: 37722567] (IHC, Mouse)

More publications at <http://www.novusbio.com/NB600-1131>

Procedures

Immunohistochemistry-Paraffin Protocol Specific for NB600-1131: IL6 Antibody (NB600-1131)

Materials

- 1) 1 Phosphate buffered saline (pH 7.6): NaCl 137mmol/L, KCl 2.7mmol/L, Na₂HPO₄ 4.3mmol/L, KH₂PO₄ 1.4 mmol/L
- 2) Citrate buffer, 0.01 M, pH6.0, Sodium Citrate 3g, Citric acid 0.4g
- 3) 3% Hydrogen peroxide
- 4) Primary antibody
- 5) Blocking serum (normal serum)
- 6) Biotinylated secondary antibody
- 7) DAB staining kit

Methods

1. Dewax and hydration of slides using xylene and EtOH:

Dry slides for 20 min in a 60 C oven

Add Xylene, 2 x 10 min

100%, 95%, 80%, and 70% EtOH, 5 min each EtOH concentration

Rinse in PBS, 5'

- 2 Antigen retrieval method (only for paraffin slides)

- 1a. High-pressure antigen retrieval procedure (recommended method)

Place slides in a glass slide holder (ensure that the slide holder is completely filled with slides, slides without sections if necessary, to ensure even heating. The entire slide holder is immersed in 1000 ml of Citrate buffer (0.01M, pH6.0) within a pressure cooker

Once steam is produced, and ONLY when steam is visible, from the pressure cooker (usually 15-20 min), the required high-pressure will have been reached, and slides will be incubated for 2 min.

Turn off heat, and allow buffer and slides to cool to room temperature

Slides are then rinsed in PBS for 5 minutes

2. Add 3% hydrogen peroxide solution, 10'at RT, then PBS, 3X5'

3. Normal blocking serum, 20'at RT

4. Incubate with Primary Ab, 4C overnight or 1.5 hours at 37C

5. Rinse with PBS, 3 X 5' each rinse

6. Add Biotin-conjugated second antibody, 10'at RT

7. Rinse with PBS, 3 X 5' each rinse

8. Add Streptavidin-Peroxidase, 10'at RT

9. Rinse with PBS, 3 X 5' each rinse

10. Staining with DAB solution, 2-5'under microscope

11. Stop the reaction by washing in tap water

12. Counterstain in Haematoxylin for 3-5 minutes

13. 75%, 80%, 95% and 100% ethanol, 5x2', xylene 2 x 10'



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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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