Product Datasheet

OCT4 Antibody - BSA Free
NB100-2379

Unit Size: 0.1 ml

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

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Reviews: 2 Publications: 26

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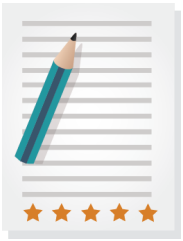
Updated 10/23/2024 v.20.1



NB100-2379

OCT4 Antibody - BSA Free

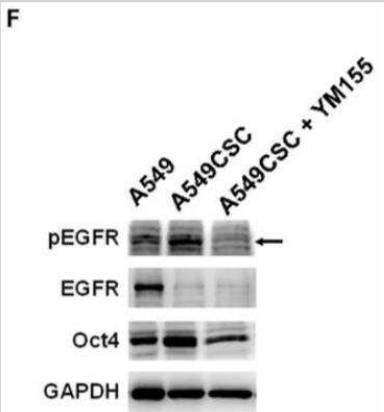
Product Information	
Unit Size	0.1 ml
Concentration	1.0 mg/ml
Storage	Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.05% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	PBS
Target Molecular Weight	40 kDa

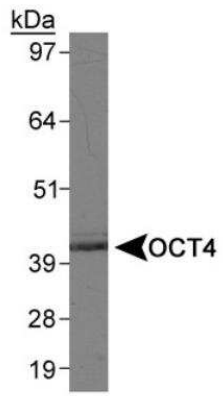
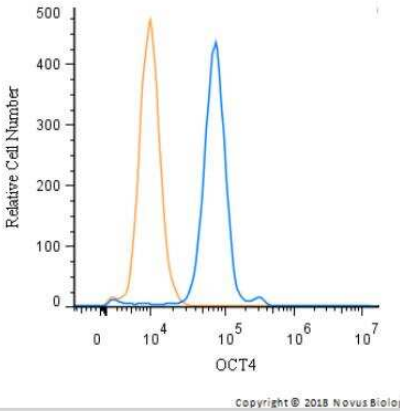
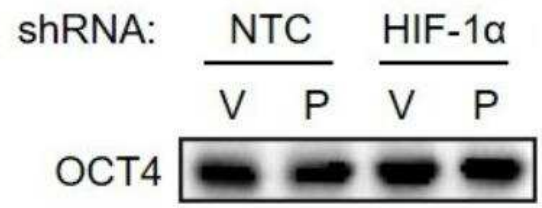
Product Description	
Host	Rabbit
Gene ID	5460
Gene Symbol	POU5F1
Species	Human, Mouse, Rat, Bovine, Primate
Reactivity Notes	Immunogen displays the following percentage of sequence identity for non-tested species: Feline (93%), Porcine (93%). Customer feedback suggests that this antibody does not give good results in IHC-P on feline tissues. Bovine reactivity reported in scientific literature (PMID: 23054358).
Marker	Embryonic Stem Cell Marker
Immunogen	A synthetic peptide made to an internal portion of the human OCT4 protein (between residues 100-200) [UniProt# Q01860]

Product Application Details	
Applications	Western Blot, Flow Cytometry, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin
Recommended Dilutions	Western Blot reported by customer review, Flow Cytometry 1:50-1:200, Immunohistochemistry 1:200. Use reported in scientific literature (PMID 30726734), Immunocytochemistry/ Immunofluorescence 1:50-1:200, Immunohistochemistry-Paraffin 1:200
Application Notes	In Western blot, a band is seen at ~40 kDa.

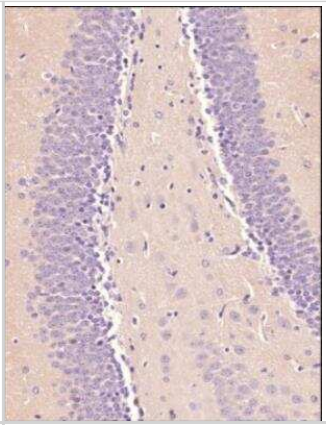
Images

Western Blot: OCT4 Antibody [NB100-2379] - YM155 significantly reduced tumorsphere formation and inhibited EGFR autophosphorylation and G9a expression. YM155 is reported to suppress cancer stemness; therefore, we used this compound to investigate the cellular mechanism of tumorsphere formation. To compare the cytotoxic capacity of YM155 against tumorsphere formation and parental cell lines, YM155 was applied to HCC827 and A549 cells in the stemness cultured or integrated medium. (F) Moreover, YM155 inhibited EGFR autophosphorylation in A549-derived tumorspheres and reduced Oct4 expression. Image collected and cropped by CiteAb from the following publication (//doi.org/10.1371/journal.pone.0182149) licensed under a CC-BY license.

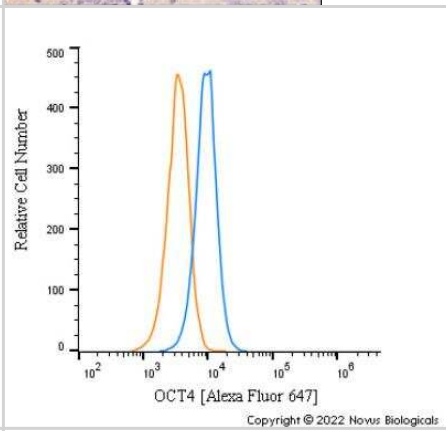


Western Blot: OCT4 Antibody [NB100-2379] - WB detection of OCT4 in mouse brain lysate using OCT4 antibody with ECL based detection. The antibody generated a specific band of OCT4 at the expected ~40kDa position.	
Flow Cytometry: OCT4 Antibody [NB100-2379] - An intracellular stain was performed on Jurkat with NB100-2379 and a matched isotype control. 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 Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody.	
Immunocytochemistry/Immunofluorescence: OCT4 Antibody [NB100-2379] - Ntera2 cells were fixed in 4% paraformaldehyde for 10 minutes and permeabilized in 0.5% Triton X-100 in PBS for 5 minutes. The cells were incubated with anti- NB100-2379 at 1 ug/ml overnight at 4C and detected with an anti-rabbit Dylight 488 (Green) at a 1:1000 dilution for 60 minutes. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 100X objective and digitally deconvolved.	
Western Blot: OCT4 Antibody [NB100-2379] - MDA-MB-231 subclones transfected with NTC or HIF-1 alpha shRNA vector were treated with vehicle (V) or paclitaxel (P) and Western blot assay was performed. WB image submitted by a verified customer review.	

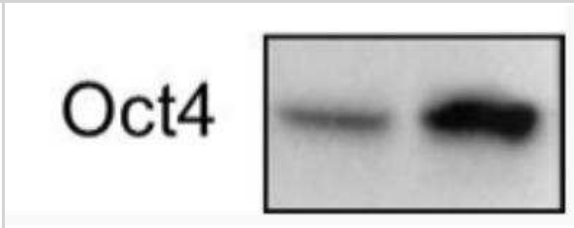
Immunohistochemistry-Paraffin: OCT4 Antibody [NB100-2379] - Analysis of a FFPE tissue section of mouse brain using 1:200 dilution of OCT4 antibody. The staining was developed using HRP labeled anti-rabbit secondary antibody and DAB reagent, and nuclei of cells were counter-stained with hematoxylin.



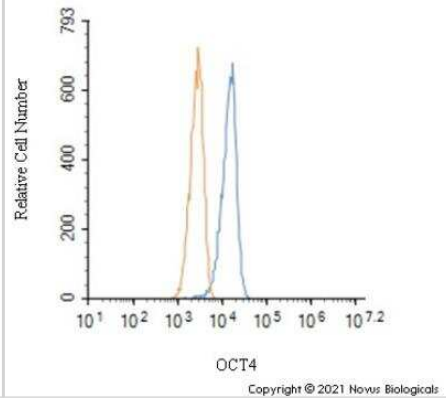
Flow Cytometry: OCT4 Antibody - BSA Free [NB100-2379] - An intracellular stain was performed on U2-OS cells with OCT4 NB100-2379AF647 (blue) and a matched isotype control NBP2-24891 (orange). Cells were fixed with 4% PFA and then permeabilized with 0.1% saponin. Cells were incubated in an antibody dilution of 2.5 ug/mL for 30 minutes at room temperature. Both antibodies were conjugated to Alexa Fluor 647.

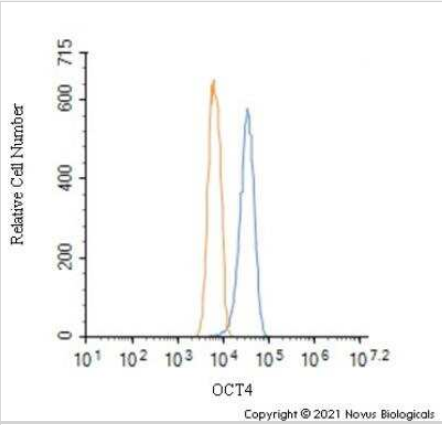
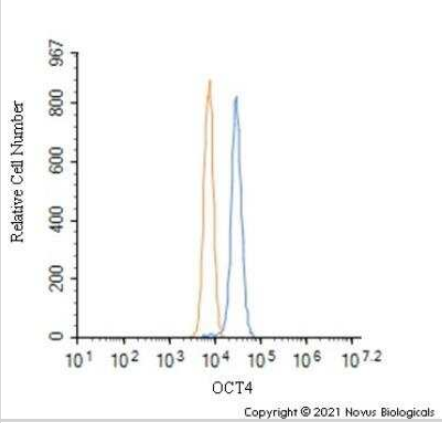
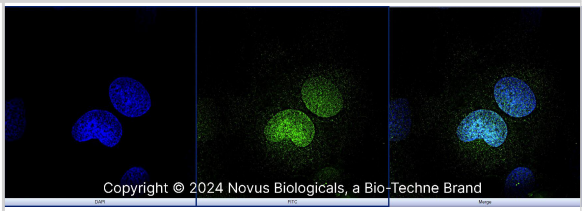
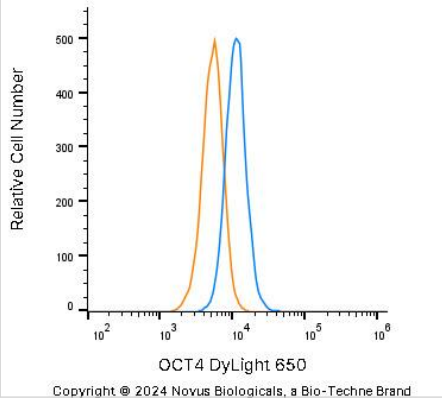


Western Blot: OCT4 Antibody [NB100-2379] - The expression of pluripotency factors Oct4 in human head and neck cancer cell lines.

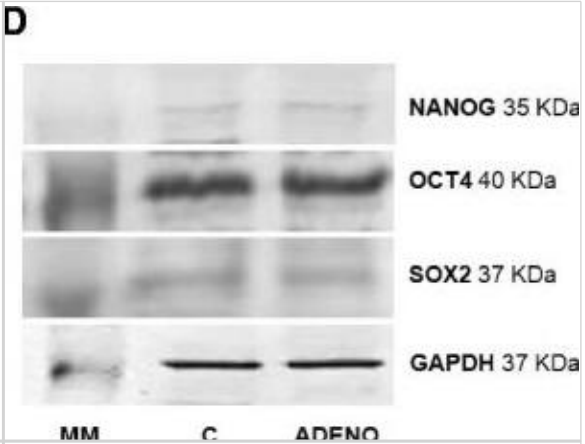


Flow Cytometry: OCT4 Antibody [NB100-2379] - An intracellular stain was performed on Jurkat cells with OCT4 Antibody NB100-2379 (blue) and a matched isotype control NBP2-24891 (orange). Cells were fixed with 4% PFA and then permeabilized with 0.1% saponin. Cells were incubated in an antibody dilution of 1.0 ug/mL for 30 minutes at room temperature, followed by Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody, Dylight 550 (SA5-10033, Thermo Fisher).

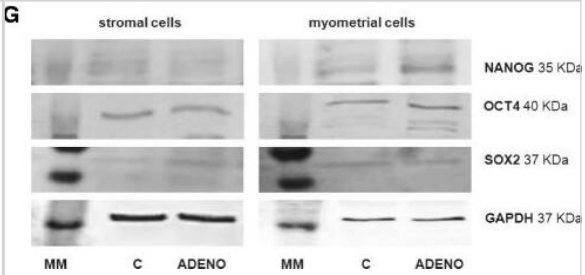


Flow Cytometry: OCT4 Antibody [NB100-2379] - An intracellular stain was performed on Neuro2a cells with OCT4 Antibody NB100-2379 (blue) and a matched isotype control NBP2-24891 (orange). Cells were fixed with 4% PFA and then permeabilized with 0.1% saponin. Cells were incubated in an antibody dilution of 1.0 ug/mL for 30 minutes at room temperature, followed by Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody, Dylight 550 (SA5-10033, Thermo Fisher).	
Flow Cytometry: OCT4 Antibody [NB100-2379] - An intracellular stain was performed on Ntera2 cells with OCT4 Antibody NB100-2379 (blue) and a matched isotype control NBP2-24891 (orange). Cells were fixed with 4% PFA and then permeabilized with 0.1% saponin. Cells were incubated in an antibody dilution of 1.0 ug/mL for 30 minutes at room temperature, followed by Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody, Dylight 550 (SA5-10033, Thermo Fisher).	
OCT4 was detected in immersion fixed U-2 OS human osteosarcoma cell line using Rabbit anti-OCT4 Affinity Purified Polyclonal Antibody conjugated to Alexa Fluor® 488 (Catalog # NB100-2379AF488) (green) at 10 µg/mL overnight at 4C. Cells were counterstained with DAPI (blue). Cells were imaged using a 100X objective and digitally deconvolved.	
An intracellular stain was performed on A431 human skin carcinoma cell line using Rabbit anti-OCT4 Affinity Purified Polyclonal Antibody conjugated to DyLight 650 (Catalog # NB100-2379C, blue histogram) or matched control antibody (orange histogram) at 2.5 µg/mL for 30 minutes at RT.	

Western Blot: OCT4 Antibody - BSA Free [NB100-2379] - Protein expression of NANOG a, OCT4 b & SOX2 c in bovine uterine tissues obtained from control cows & from cows with adenomyosis. Data were normalized against glyceraldehyde-3-phosphate dehydrogenase (GAPDH). Bars represent the mean ± SEM. There were no statistical differences between uterine normal & adenomyotic tissues (P > 0.05), as determined by Student's t-test. Representative blots for NANOG, OCT4, SOX2 & GAPDH are shown below the graphs d. MM – molecular weight marker, C – tissues obtained from control cows, ADENO – tissues obtained from cows with adenomyosis Image collected & cropped by CiteAb from the following publication (http://www.rbej.com/content/13/1/110), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Western Blot: OCT4 Antibody - BSA Free [NB100-2379] - Protein expression of pluripotency markers in uterine cells isolated from control cows & from cows with adenomyosis. NANOG a OCT4 b & SOX2 c protein expression in uterine stromal cells. NANOG d OCT4 e & SOX2 f protein expression in uterine myometrial cells. Data were normalized against glyceraldehyde-3-phosphate dehydrogenase (GAPDH). Bars represent the mean ± SEM. Asterisks indicate statistical differences between uterine normal & adenomyotic tissue (*P < 0.05; **P < 0.01), as determined by Student's t-test. Representative blots for NANOG, OCT4, SOX2 & GAPDH are shown below the graphs g. MM – molecular weight marker, C – cells obtained from control cows, ADENO – cells obtained from cows with adenomyosis Image collected & cropped by CiteAb from the following publication (http://www.rbej.com/content/13/1/110), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Publications

Haiquan Lu, Yajing Lyu, Linh Tran, Jie Lan, Yangyiran Xie, Yongkang Yang, Naveena L Murugan, Yueyang J Wang, Gregg L Semenza HIF-1 recruits NANOG as a coactivator for TERT gene transcription in hypoxic breast cancer stem cells. Cell reports 2022-02-10 [PMID: 34592152]

Siu WS, Ma H, Ko CH et al. Rat Plantar Fascia Stem/Progenitor Cells Showed Lower Expression of Ligament Markers and Higher Pro-Inflammatory Cytokines after Intensive Mechanical Loading or Interleukin-1? Treatment In Vitro Cells 2023-09-06 [PMID: 37759446] (Immunocytochemistry/ Immunofluorescence)

Tang S, Jones C, Dye J, Coward K Dissociation, enrichment, and the in vitro formation of gonocyte colonies from cryopreserved neonatal bovine testicular tissues Theriogenology 2023-07-22 [PMID: 37499372] (ICC/IF, Bovine)

Panda DK, Bai X, Zhang Y et al. SCF-SKP2 E3 ubiquitin ligase links mTORC1-ER stress-ISR with YAP activation in murine renal cystogenesis The Journal of clinical investigation 2022-11-03 [PMID: 36326820] (WB, Mouse)

Zhang R, Zhang X, Zhang W et al. Sohlh2 regulates the stemness and differentiation of colon cancer stem cells by downregulating LncRNA-H19 transcription Molecular cancer research : MCR 2022-10-26 [PMID: 36287177] (WB, Human)

Hekman KE, Koss KM, Ivancic DZ et al. Autophagy Enhances Longevity of Induced Pluripotent Stem Cell-Derived Endothelium via mTOR-Independent ULK1 Kinase Stem cells translational medicine 2022-09-29 [PMID: 36173887] (FLOW, Human)

Yang Y, Chen C, Zuo Q Et al. NARF is a hypoxia-induced coactivator for OCT4-mediated breast cancer stem cell specification Sci Adv 2022-12-09 [PMID: 36490339] (WB, Human)

Details:
Citation using the DyLight 405 version of this antibody.

Zhang R, Liu L, Wang F et al. AKAP8L enhances the stemness and chemoresistance of gastric cancer cells by stabilizing SCD1 mRNA Research Square 2022-08-25 [PMID: 36522343] (WB, Human)

BAAth M, JOnsson JM, Westbom Fremer S et al. MET Expression and Cancer Stem Cell Networks Impact Outcome in High-Grade Serous Ovarian Cancer Genes 2021-05-14 [PMID: 34069138] (WB, Human)

Yang J, Liu H, Sun H et al. Construction of induced pluripotent stem cell line (ZZUi0017-A) from the fibroblast cells of a female patient with CACNA1A mutation by unintegrated reprogramming approach Stem Cell Res 2020-08-04 [PMID: 32791484] (ICC/IF, Human)

Li W, Zhang N, Jin C et al. MUC1-C drives stemness in progression of colitis to colorectal cancer JCI Insight 2020-05-19 [PMID: 32427590] (KO, WB, Mouse)

Lu H, Xie Y, Tran L et al. Chemotherapy-induced S100A10 recruits KDM6A to facilitate OCT4-mediated breast cancer stemness J Clin Invest. 2020-05-19 [PMID: 32427586] (WB, KD, Human)

More publications at http://www.novusbio.com/NB100-2379

Procedures

Western Blot Protocol for OCT4 Antibody (NB100-2379)
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 manufacturers instructions.

Immunohistochemistry-Paraffin Protocol for OCT4 Antibody (NB100-2379)
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 all the time).

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 OCT4 Antibody (NB100-2379)
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.

Novus Biologicals USA
10730 E. Briarwood Avenue
Centennial, CO 80112
USA
Phone: 303.730.1950
Toll Free: 1.888.506.6887
Fax: 303.730.1966
nb-customerservice@bio-techne.com

Bio-Techne Canada
21 Canmotor Ave
Toronto, ON M8Z 4E6
Canada
Phone: 905.827.6400
Toll Free: 855.668.8722
Fax: 905.827.6402
canada.inquires@bio-techne.com

Bio-Techne Ltd
19 Barton Lane
Abingdon Science Park
Abingdon, OX14 3NB, United Kingdom
Phone: (44) (0) 1235 529449
Free Phone: 0800 37 34 15
Fax: (44) (0) 1235 533420
info.EMEA@bio-techne.com

General Contact Information
www.novusbio.com
Technical Support: nb-technical@bio-techne.com
Orders: nb-customerservice@bio-techne.com
General: novus@novusbio.com

Products Related to NB100-2379

NB800-PC1	HeLa Whole Cell Lysate
NB100-2379PEP	OCT4 Antibody Blocking Peptide
HAF008	Goat anti-Rabbit IgG Secondary Antibody [HRP]
NB7160	Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]
NBP2-24891	Rabbit IgG Isotype Control

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.

For more information on our 100% guarantee, please visit www.novusbio.com/guarantee

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