# Product Datasheet

## alpha Tubulin Antibody (YOL1/34) - BSA Free
## NB100-1639SS

Unit Size: 0.05 mg

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

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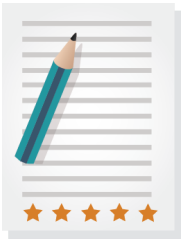
Updated 11/11/2025 v.20.1





# NB100-1639SS

alpha Tubulin Antibody (YOL1/34) - BSA Free

## Product Information

| | |
|---|---|
| **Unit Size** | 0.05 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** | YOL1/34 |
| **Preservative** | 0.02% Sodium Azide |
| **Isotype** | IgG2a |
| **Purity** | Protein G purified |
| **Buffer** | PBS |
| **Target Molecular Weight** | 50 kDa |

## Product Description

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|---|---|
| **Description** | Novus Biologicals Rat alpha Tubulin Antibody (YOL1/34) - BSA Free (NB100-1639) is a monoclonal antibody validated for use in IHC, WB, ELISA, Flow and ICC/IF. Anti-alpha Tubulin Antibody: Cited in 29 publications. All Novus Biologicals antibodies are covered by our 100% guarantee. |
| **Host** | Rat |
| **Gene ID** | 7846 |
| **Gene Symbol** | TUBA1A |
| **Species** | Human, Mouse, Rat, Drosophila, Fungi, Yeast |
| **Reactivity Notes** | Cross reacts with Arabidopsis, Saccharomyces, Platyzoma, Ashbya, and Naegleria. Based on sequence similarity, is expected to react with: Birds and Mammals. |
| **Marker** | Microtubule Marker |
| **Specificity/Sensitivity** | This alpha Tubulin Antibody (YOL1/34) detects an epitope on alpha tubulin between the amino acids 414-422. It has a higher affinity for fixed microtubules (formaldehyde or glutaraldehyde) than native microtubules when performing IHC. |
| **Immunogen** | This alpha Tubulin Antibody (YOL1/34) was made from yeast tubulin. |

## Product Application Details

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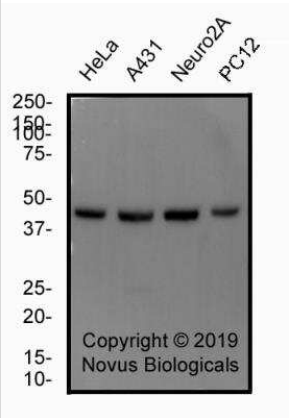
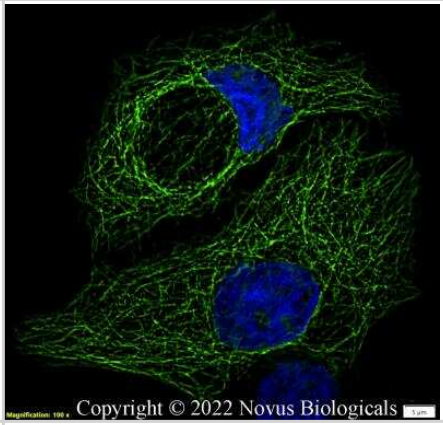
| **Applications** | Western Blot, ELISA, Flow Cytometry, Flow (Intracellular), Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Radioimmunoassay |
| **Recommended Dilutions** | Western Blot 1:100-1:2000, Flow Cytometry, ELISA 10 ug/ml, Immunohistochemistry 1:10-1:500, Immunocytochemistry/ Immunofluorescence 1:10-1:500, Immunohistochemistry-Frozen 1:10-1:500, Radioimmunoassay 1:100-1:2000, Flow (Intracellular) |

## Images

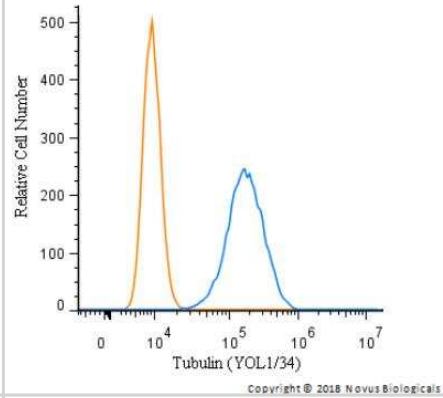
Western Blot: alpha Tubulin Antibody (YOL1/34) [NB100-1639] - Total protein from human HeLa and A431, mouse 3T3 and rat PC12 cell lines was separated on a 12% gel by SDS-PAGE, transferred to PVDF membrane and blocked in 5% non-fat milk in TBST. The membrane was probed with 2.0 ug/ml anti-alpha Tubulin in block buffer and detected with an anti-mouse HRP secondary antibody using West Pico PLUS chemiluminescence detection reagent. Alpha tubulin molecular weight: 50 kDa.



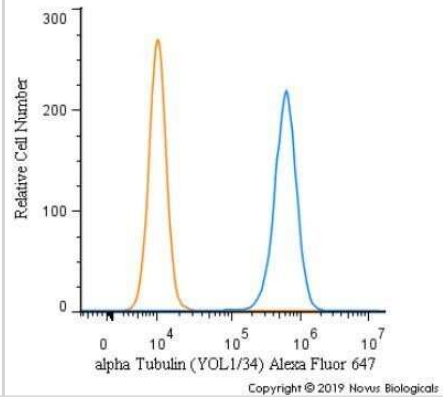

Immunocytochemistry/Immunofluorescence: alpha Tubulin Antibody (YOL1/34) - BSA Free [NB100-1639] - A431 cells were fixed in 4% paraformaldehyde for 10 minutes and permeabilized in 0.05% Triton X-100 in PBS for 5 minutes. The cells were incubated with alpha Tubulin Antibody [YOL1/34] (NB100-1639) at 1ug/ml overnight at 4C and detected with an anti-rat 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.

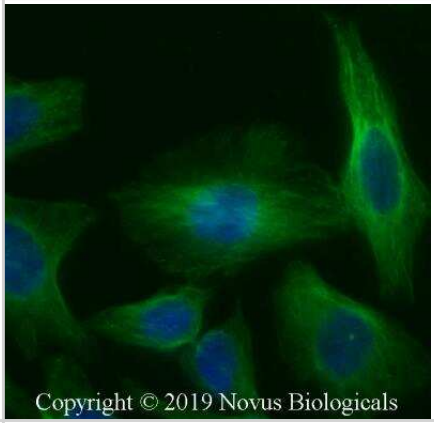
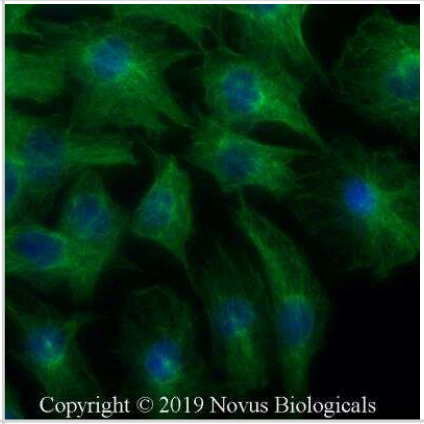
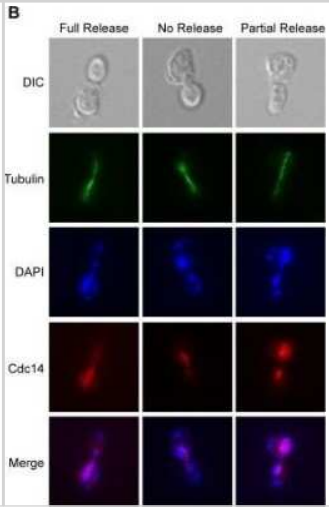



Flow (Intracellular): alpha Tubulin Antibody (YOL1/34) [NB100-1639] - An intracellular stain was performed on HeLa cells with Tubulin [YOL1/34] Antibody NB100-1639 (blue) and a matched isotype control (orange). Cells were fixed with 4% PFA and then permeabilized with 0.1% saponin. Cells were incubated in an antibody dilution of 1.0 ug/mL for 30 minutes at room temperature, followed by Rat IgG (H+L) Cross-Adsorbed Secondary Antibody, Dylight 488.

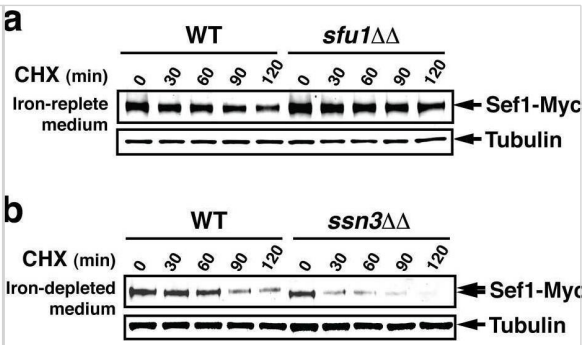



Flow Cytometry: alpha Tubulin Antibody (YOL1/34) [NB100-1639] - An intracellular stain was performed on HeLa cells with alpha Tubulin Antibody [YOL1/34] NB100-1639AF647 (blue) and a matched isotype control (orange). Cells were fixed with 4% PFA and then permeabilized with 0.1% saponin. Cells were incubated in an antibody dilution of 2.5 ug/mL for 30 minutes at room temperature. Both antibodies were conjugated to Alexa Fluor 647. Image using the Alexa Fluor 647 format of this antibody.



| | |
|---|---|
| Immunocytochemistry/Immunofluorescence: alpha Tubulin Antibody (YOL1/34) [NB100-1639] - HeLa cells were fixed for 10 minutes using 10% formalin and then permeabilized for 5 minutes using 1X PBS + 0.05% Triton-X100. The cells were incubated with anti-Tubulin Antibody [YOL1/34] at 5 ug/ml overnight at 4C and detected with an anti-rat Dylight 488 (Green) at a 1:500 dilution. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective. |  |
| Immunocytochemistry/Immunofluorescence: alpha Tubulin Antibody (YOL1/34) - BSA Free [NB100-1639] - alpha Tubulin Antibody (YOL1/34) [NB100-1639] - NIH3T3 cells were fixed for 10 minutes using 10% formalin and then permeabilized for 5 minutes using 1X PBS + 0.05% Triton-X100. The cells were incubated with anti-Tubulin Antibody [YOL1/34] at 5 ug/ml overnight at 4C and detected with an anti-rat Dylight 488 (Green) at a 1:500 dilution. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective. | |
| Immunocytochemistry/Immunofluorescence: alpha Tubulin Antibody (YOL1/34) [NB100-1639] - PC12 cells were fixed for 10 minutes using 10% formalin and then permeabilized for 5 minutes using 1X PBS + 0.05% Triton-X100. The cells were incubated with anti-Tubulin Antibody [YOL1/34] at 5 ug/ml overnight at 4C and detected with an anti-rat Dylight 488 (Green) at a 1:500 dilution. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective. |  |
| Immunocytochemistry/Immunofluorescence: alpha Tubulin Antibody (YOL1/34) [NB100-1639] - Mutants unable to ubiquitinate histone H2B-K123 display growth and Cdc14 release defects when combined with MEN mutants. Representative images from Cdc14 release assay. Cells are assayed for spindle morphology and Cdc14 release phenotype by indirect immunofluorescence with anti-tubulin or anti-HA, which recognizes the HA-tagged version of Cdc14 in all strains. Cells are visualized by nuclear staining with DAPI and by differential interference contrast (DIC) optics for comparison. No release refers to dense nucleolar staining of Cdc14-HA, full release refers to cells with uniform Cdc14-HA localization throughout the nucleus, while partial release refers to cells with both nuclear and residual nucleolar staining. Image collected and cropped by CiteAb from the following publication (https://dx.plos.org/10.1371/journal.pgen.1000588), licensed under a CC-BY license. |  |

Sfu1 and Ssn3 mediate opposing effects on Sef1 stability and virulence.a) Sef1 protein is stabilized in an sfu1ΔΔ mutant. Wild-type or sfu1ΔΔ strains containing Sef1-Myc were propagated in iron-replete medium containing 2 mg/ml cycloheximide. Samples were recovered at the indicated time points, and Sef1-Myc was visualized using monoclonal antibodies against the Myc epitope, followed by incubation with secondary antibodies that were coupled to infrared dyes and quantified using a Li-Cor instrument. Note that a single band corresponding to higher mobility (unphosphorylated) Sef1 is present in both strains. b) Sef1 protein is destabilized in the ssn3ΔΔ mutant. The experiment was performed as above, except that cells were propagated in iron-depleted medium containing cycloheximide. Note that phosphorylated Sef1 recovered from wild-type cells runs with slower mobility. c) Model for Sef1 regulation by Sfu1 and Ssn3 under iron-replete vs. iron-depleted conditions. Note that, even under iron-replete conditions when nuclear Sfu1 functions as a transcriptional repressor, a cytoplasmic pool of Sfu1 is available (FigureS4) that could participate in Sef1 sequestration. d) Overexpression of SFU1 leads to attenuated C. albicans virulence in a murine bloodstream infections model. * signifies p<0.02, log-rank test. E) Deletion of SSN3 leads to attenuated C. albicans virulence, and restoration of one copy of wild-type SSN3 complements the defect. * signifies p<0.0001. Image collected and cropped by CiteAb from the following open publication (https://pubmed.ncbi.nlm.nih.gov/23133381), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



## Publications


Huang X, Chen G, Wu L et al. Coordinated regulation of pH alkalinization by two transcription factors promotes fungal commensalism and pathogenicity. Nature Communications 2025-08-22 [PMID: 40846842]

Ren J, Hannun Y Orm2 promotes nitrogen-induced sphingolipid production and endocytosis via Orm1 phosphorylation. Journal of Lipid Research 2025-10-08 [PMID: 41072797]

Murayama Y, Samora CP, Kurokawa Y, Iwasaki H et Al. Establishment of DNA-DNA Interactions by the Cohesin Ring Cell 2018-01-24 [PMID: 29358048]

Ren J, Rieger R, Pereira de Sa N et al. Orm proteins control ceramide synthesis and endocytosis via LCB-mediated Ypk1 regulation. Journal of lipid research 2024-10-25 [PMID: 39490931]

Peter T. A. Linders, Eveline C. F. Gerretsen, Angel Ashikov, Mari-Anne Vals, Rinse de Boer, Natalia H. Revelo, Richard Arts, Melissa Baerenfaenger, Fokje Zijlstra, Karin Huijben, Kimiyo Raymond, Kai Muru, Olga Fjodorova, Sander Pajusalu, Katrin Õunap, Martin ter Beest, Dirk Lefeber, Geert van den Bogaart Congenital disorder of glycosylation caused by starting site-specific variant in syntaxin-5 Nature Communications 2021-10-28 [PMID: 34711829]

Harry Warner, Giulia Franciosa, Guus van der Borg, Britt Coenen, Felix Faas, Claire Koenig, Rinse de Boer, René Classens, Sjors Maassen, Maksim V Baranov, Shweta Mahajan, Deepti Dabral, Frans Bianchi, Niek van Hilten, Herre Jelger Risselada, Wouter H Roos, Jesper Velgaard Olsen, Laia Querol Cano, Geert van den Bogaart Atypical cofilin signaling drives dendritic cell migration through the extracellular matrix via nuclear deformation. Cell reports 2024-04-01 [PMID: 38416638]

Wang PH, Washburn RS, Mariuzza DL et al. Reciprocal transmission of activating and inhibitory signals and cell fate in regenerating T cells Cell reports 2023-09-26 [PMID: 37756164] (ICC/IF, Mouse)

Acharya BR, Fang JS, Jeffery ED et al. Connexin 37 sequestering of activated-ERK in the cytoplasm promotes p27-mediated endothelial cell cycle arrest Life science alliance 2023-08-01 [PMID: 37197981]

Stempels FC, Jiang M, Warner HM et al. Giant worm-shaped ESCRT scaffolds surround actin-independent integrin clusters The Journal of cell biology 2023-07-03 [PMID: 37200023] (ICC/IF, Human)

Wang Y, Mao Y, Chen X et al. Homeostatic control of an iron repressor in a GI tract resident bioRxiv 2023-01-31 [PMID: 37227051] (WB)

Feng C, Kou L, Yin P, Jing Y Excessive activation of IL 33/ST2 in cancer associated fibroblasts promotes invasion and metastasis in ovarian cancer Oncology Letters 2022-03-17 [PMID: 35399326] (WB, Human)

Stempels FC, Janssens MH, Ter Beest M et al. Novel and conventional inhibitors of canonical autophagy differently affect LC3-associated phagocytosis FEBS letters 2022-01-10 [PMID: 35007347] (WB, Human)


More publications at http://www.novusbio.com/NB100-1639

## Procedures

**Immunocytochemistry/Immunofluorescence protocol for alpha Tubulin Antibody (NB100-1639)**

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.

**Western Blot protocol for alpha Tubulin Antibody (NB100-1639)**

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 alpha Tubulin Antibody (NB100-1639)**

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.

Staining:
1. Wash sections in deionized water three times for 5 minutes each.
2. Wash sections in wash buffer for 5 minutes.
3. Block each section with 100-400 ul blocking solution 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 biotinylated diluted secondary antibody. Incubate 30 minutes at room temperature.
7. Remove secondary antibody solution and wash sections three times with wash buffer for 5 minutes each.
8. Add 100-400 ul Streptavidin-HRP reagent to each section and incubate for 30 minutes at room temperature.
9. Wash sections three times in wash buffer for 5 minutes each.
10. Add 100-400 ul DAB substrate to each section and monitor staining closely.
11. As soon as the sections develop, immerse slides in deionized water.
12. Counterstain sections in hematoxylin.
13. Wash sections in deionized water two times for 5 minutes each.
14. Dehydrate sections.
15. Mount coverslips.

**Flow (Intracellular) protocol for alpha Tubulin Antibody (NB100-1639)**

Sample Preparation.
1. Grow cells to 60-85% confluency. Flow cytometry requires between 2 x 105 and 1 x 106 cells for optimal performance.
2. If cells are adherent, harvest gently by washing once with staining buffer and then scraping. Avoid using trypsin as this can disrupt certain epitopes of interest. If enzymatic harvest is required, use Accutase, Collagenase, or TrypLE Express for a less damaging option.
3. Reserve 100 uL for counting, then transfer cell volume into a 50 mL conical tube and centrifuge for 8 minutes at 400 RCF.
a. Count cells using a hemocytometer and a 1:1 trypan blue exclusion stain to determine cell viability before starting the flow protocol. If cells appear blue, do not proceed.
4. Re-suspend cells to a concentration of 1 x 106 cells/mL in staining buffer (NBP2-26247).
5. Aliquot out 100 uL samples in accordance with your experimental samples.

Tip: When cell surface and intracellular staining are required in the same sample, it is advisable that the cell surface staining be performed first since the fixation and permeablization steps might reduce the availability of surface antigens.

Intracellular Staining.
Tip: When performing intracellular staining, it is important to use appropriate fixation and permeabilization reagents based upon the target and its subcellular location. Generally, our Intracellular Flow Assay Kit (NBP2-29450) is a good place to start as it contains an optimized combination of reagents for intracellular staining as well as an inhibitor of intracellular protein transport (necessary if staining secreted proteins). Certain targets may require more gentle or transient permeabilization protocols such as the commonly employed methanol or saponin-based methods.
Protocol for Cytoplasmic Targets:
1. Fix the cells by adding 100 uL fixation solution (such as 4% PFA) to each sample for 10-15 minutes.
2. Permeabilize cells by adding 100 uL of a permeabization buffer to every 1 x 106 cells present in the sample. Mix well and incubate at room temperature for 15 minutes.
a. For cytoplasmic targets, use a gentle permeabilization solution such as 1X PBS + 0.5% Saponin or 1X PBS + 0.5% Tween-20.
b. To maintain the permeabilized state throughout your experiment, use staining buffer + 0.1% of the permeabilization reagent (i.e. 0.1% Tween-20 or 0.1% Saponin).
3. Following the 15 minute incubation, add 2 mL of the staining buffer + 0.1% permeabilizer to each sample.
4. Centrifuge for 1 minute at 400 RCF.
5. Discard supernatant and re-suspend in 100 uL of staining buffer + 0.1% permeabilizer.
6. Add appropriate amount of each antibody (eg. 1 test or 1 ug per sample, as experimentally determined).
7. Mix well and incubate at room temperature for 30 minutes- 1 hour. Gently mix samples every 10-15 minutes.
8. Following the primary/conjugate incubation, add 1-2 mL/sample of staining buffer +0.1% permeabilizer and centrifuge for 1 minute at 400 RCF.
9. Wash twice by re-suspending cells in staining buffer (2 mL for tubes or 200 uL for wells) and centrifuging at 400 RCF for 5 minutes. Discard supernatant.
10. Add appropriate amount of secondary antibody (as experimentally determined) to each sample.
11. Incubate at room temperature in dark for 20 minutes.
12. Add 1-2 mL of staining buffer and centrifuge at 400 RCF for 1 minute and discard supernatant.
13. Wash twice by re-suspending cells in staining buffer (2 mL for tubes or 200 uL for wells) and centrifuging at 400 RCF for 5 minutes. Discard supernatant.
14. Resuspend in an appropriate volume of staining buffer (usually 500 uL per sample) and proceed with analysis on your flow cytometer.

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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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