Product Datasheet

Lysozyme Antibody - BSA Free
NBP2-61118

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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Publications: 7

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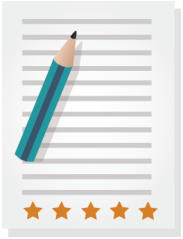
Updated 4/13/2025 v.20.1



NBP2-61118

Lysozyme Antibody - 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	Polyclonal
Preservative	0.02% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	PBS

Product Description

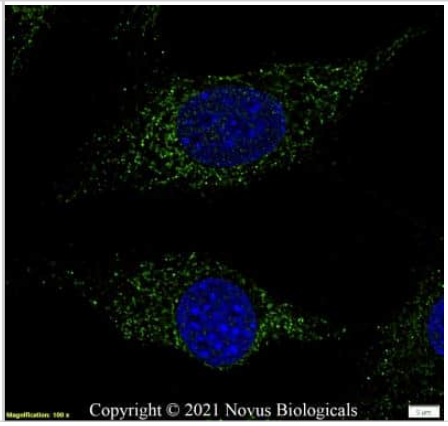
Host	Rabbit
Gene ID	4069
Gene Symbol	LYZ
Species	Human, Mouse
Immunogen	Full length recombinant human Lysozyme protein.

Product Application Details

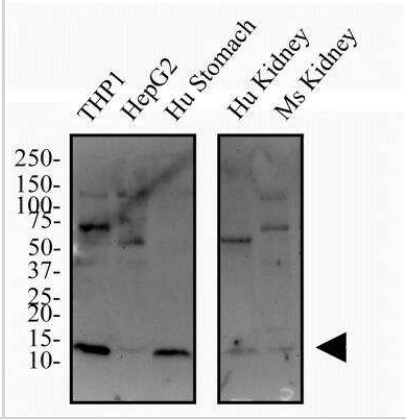
Applications	Western Blot, Simple Western, Flow Cytometry, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin
Recommended Dilutions	Western Blot 2 ug/ml, Simple Western, Flow Cytometry, Immunohistochemistry 1:100 - 1:250, Immunocytochemistry/ Immunofluorescence 2 ug/ml, Immunohistochemistry-Paraffin 1:100 - 1:250

Images

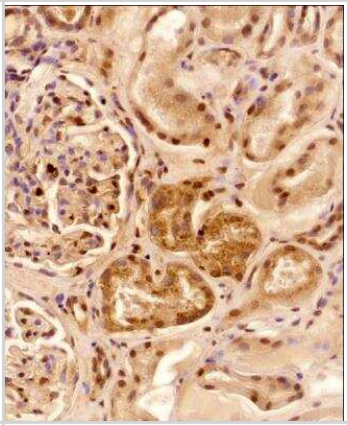
Immunocytochemistry/Immunofluorescence: Lysozyme Antibody [NBP2-61118] - NIH3T3 cells were fixed and permeabilized for 10 minutes using -20C MeOH. The cells were incubated with anti-Lysozyme NBP2-61118 at 2 ug/ml overnight at 4C and detected with an anti-rabbit Dylight 488 (Green) at a 1:1000 dilution for 60 minutes. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 100X objective and digitally deconvolved.



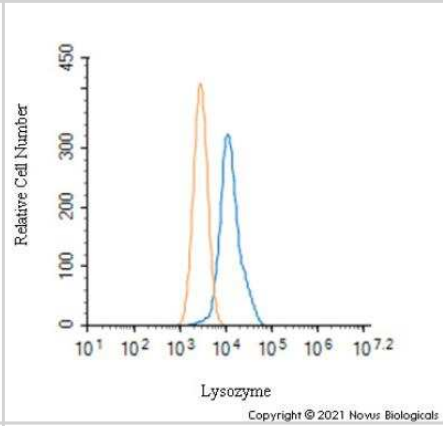
Western Blot: Lysozyme Antibody [NBP2-61118] - Total protein from human cell lines THP-1 and HepG2, human stomach and kidney as well as mouse kidney was separated on a 4-20% gel by SDS-PAGE, transferred to 0.2 um PVDF membrane and blocked in 5% non-fat milk in TBST. The membrane was probed with 2.0 ug/ml anti-Lysozyme in 5% non-fat milk in TBST and detected with an anti-rabbit HRP secondary antibody using chemiluminescence.



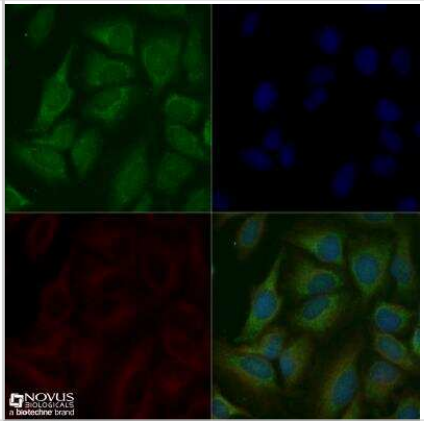
Immunohistochemistry-Paraffin: Lysozyme Antibody [NBP2-61118] - Analysis of a FFPE tissue section of human kidney with Lysozyme antibody at 1:100 dilution. The staining was developed with HRP-DAB detection method and the counterstaining was performed using hematoxylin. This Lysozyme antibody generated an expected cytoplasmic staining in all the cells with strongest signal in tubular epithelial cells. Some tubules showed more of a punctate staining pattern (vesicular Lysozyme) while all of the tubules depicted a diffused signal (secreted Lysozyme). A subset of cells, especially those from glomeruli, showed nuclear positivity also.

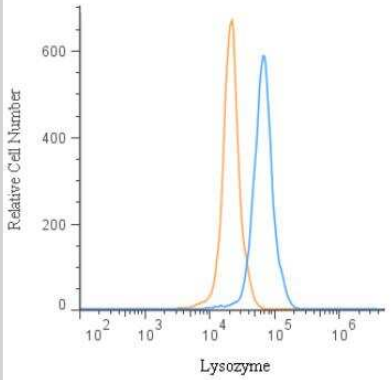
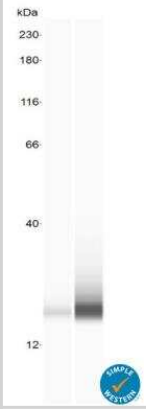


Flow Cytometry: Lysozyme Antibody [NBP2-61118] - An intracellular stain was performed on HepG2 cells with NBP2-61118 (blue) and a matched isotype control NBP2-24891 (orange). Cells were fixed with 4% PFA and then permeabilized with 0.1% saponin. Cells were incubated in an antibody dilution of 1.0 ug/mL for 30 minutes at room temperature, followed by Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody, Dylight 550 (SA5-10033, Thermo Fisher).



Immunocytochemistry/Immunofluorescence: Lysozyme Antibody [NBP2-61118] - HeLa cells were fixed and permeabilized for 10 minutes using -20C MeOH. The cells were incubated with anti-Lysozyme at 2 ug/ml overnight at 4C and detected with an anti-rabbit DyLight 488 (Green) at a 1:500 dilution. Alpha tubulin (DM1A) NB100-690 was used as a co-stain at a 1:1000 dilution and detected with an anti-mouse DyLight 550 (Red) at a 1:500 dilution. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective.



Flow Cytometry: Lysozyme Antibody [NBP2-61118] - An intracellular stain was performed on THP-1 Cells with NBP2-61118 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 2.5 ug/mL for 30 minutes at room temperature, followed by Rabbit IgG APC-conjugated Secondary Antibody (R&D Systems, F0111).	
Simple Western: Lysozyme Antibody [NBP2-61118] - Lane view shows lysates of Ileum stem cells undifferentiated and day 5 differentiated, loaded at 0.2 mg/mL. A specific band was detected for Lysozyme at approximately 20 kDa (as indicated) using 20 ug/mL of Rabbit Anti-Lysozyme Polyclonal Antibody (NBP2-61118). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.	
Lysozyme was detected in immersion fixed HepG2 human hepatocellular carcinoma cell line using Rabbit anti-Lysozyme Affinity Purified Polyclonal Antibody conjugated to DyLight 650 (Catalog # NBP2-61118C) (light blue) at 10 µg/mL overnight at 4C. Cells were counterstained with DAPI (dark blue). Cells were imaged using a 100X objective and digitally deconvolved.	

Publications

Tang L, Xie L, Zhou H et al. Perillaldehyde mitigates ionizing radiation-induced intestinal injury by inhibiting ferroptosis via the Nrf2 signaling pathway Research Square 2023-03-06 (IHC-P)

HAfliger J, Schwarzfischer M, Atrott K et al. Glycoprotein (GP)96 Is Essential for Maintaining Intestinal Epithelial Architecture by Supporting Its Self-Renewal Capacity Cellular and molecular gastroenterology and hepatology 2022-12-11 [PMID: 36516930] (IHC-P, Mouse)

Chen B, Li R, Kubota A et al. Identification of macrophages in normal and injured mouse tissues using reporter lines and antibodies Scientific reports 2022-03-16 [PMID: 35296717] (IHC-P, Mouse)

Bao L, Cui X, Wang X et al. Carbon Nanotubes Promote the Development of Intestinal Organoids through Regulating Extracellular Matrix Viscoelasticity and Intracellular Energy Metabolism ACS Nano 2021-10-26 [PMID: 34622660]

Details:
Citation using the PE format of this antibody.

Gobert AP, Latour YL, Asim M Et al. Protective Role of Spermidine in Colitis and Colon Carcinogenesis Gastroenterology 2021-11-10 [PMID: 34767785]

Conzelmann C, Grob R, Zou M et al. Salivary extracellular vesicles inhibit Zika virus but not SARS-CoV-2 infection Extracell Vesicles 2020-08-24 [PMID: 32939236] (WB)

Li Q, Nirala NK, Chen HJ et al The Misshapen subfamily of Ste20 kinases regulate proliferation in the aging mammalian intestinal epithelium. J Cell Physiol. 2019-12-01 [PMID: 31042012] (ICC/IF)

Details:
Citation using the Alexa Fluor 488 version of this antibody.

Procedures

Western Blot protocol for Lysozyme Antibody (NBP2-61118)
Lysozyme Antibody:
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 anti-Lysozyme 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 Lysozyme Antibody (NBP2-61118)
Lysozyme Antibody:
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.

Immunocytochemistry/ Immunofluorescence Protocol for Lysozyme Antibody (NBP2-61118)
Lysozyme Antibody:
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.

*The above information is only intended as a guide. The researcher should determine what protocol best meets their needs. Please follow safe laboratory procedures.

Flow (Intracellular) protocol for Lysozyme Antibody (NBP2-61118)

Lysozyme Antibody:
Protocol for Flow Cytometry Intracellular Staining
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 1 mL 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:
Optional: Perform cell surface staining as described in the previous section.
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 5 minutes at 400 RCF.
5. Discard supernatant and re-suspend in 1 mL of staining buffer + 0.1% permeabilizer.
6. Stain each sample at 1 uL/ 1 x 106 cells of primary antibody or 1-3 uL/ 1 x 106 cells for directly conjugated antibodies. Mix well and incubate at room temperature for 30 minutes- 1 hour. Gently mix samples every 10-15 minutes.
7. Following the primary/conjugate incubation, add 2 mL/sample of staining buffer +0.1% permeabilizer and centrifuge for 5 minutes at 400 RCF.
8. Remove supernatant and re-suspend each sample in 2 mL staining buffer + 0.1% permeabilizer, repeat wash for 5 minutes at 400 RCF.
9. If using a directly conjugated antibody, after the second wash, re-suspend cell pellet to a final volume of 500 uL per sample and proceed with flow analysis.

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Products Related to NBP2-61118

HAF008	Goat anti-Rabbit IgG Secondary Antibody [HRP]
NB7160	Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]
NBP2-24891	Rabbit IgG Isotype Control
P5238	Lysozyme Native Protein

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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