

Product Datasheet

NFkB1/NFkB p105 Antibody - BSA Free NBP1-77395

Unit Size: 0.1 ml

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

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NBP1-77395

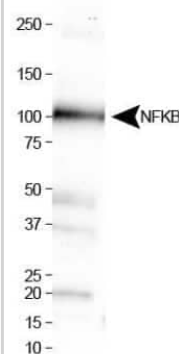
NFkB1/NFkB p105 Antibody - BSA Free

Product Information	
Unit Size	0.1 ml
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	0.05% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	PBS and 30% Glycerol
Target Molecular Weight	105 kDa
Product Description	
Host	Rabbit
Gene ID	4790
Gene Symbol	NFKB1
Species	Human, Mouse
Immunogen	A genomic peptide made to an N-terminal region of the human NFkB p105/p50 protein (within residues 400-590). [Swiss-Prot P19838]
Notes	Manufactured by Genomic Antibody Technology™. GAT FAQs
Product Application Details	
Applications	Western Blot, Simple Western, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin
Recommended Dilutions	Western Blot 1:1000, Simple Western 1:20, Immunohistochemistry 1:250, Immunocytochemistry/ Immunofluorescence 1:100, Immunohistochemistry-Paraffin 1:250, Immunohistochemistry-Frozen reported in scientific literature (PMID 26889045)
Application Notes	In Western blot, a band is seen approx. 110 kDa. Prior to immunostaining paraffin tissues, antigen retrieval with sodium citrate buffer (pH 6.0) is recommended. In Simple Western only 10 - 15 uL of the recommended dilution is used per data point. See Simple Western Antibody Database for Simple Western validation: Tested in HepG2 lysate 0.5 mg/mL, separated by Size, antibody dilution of 1:20, apparent MW was 46 kDa. Separated by Size-Wes, Sally Sue/Peggy Sue. The observed molecular weight of the protein may vary from the listed predicted molecular weight due to post translational modifications, post translation cleavages, relative charges, and other experimental factors.

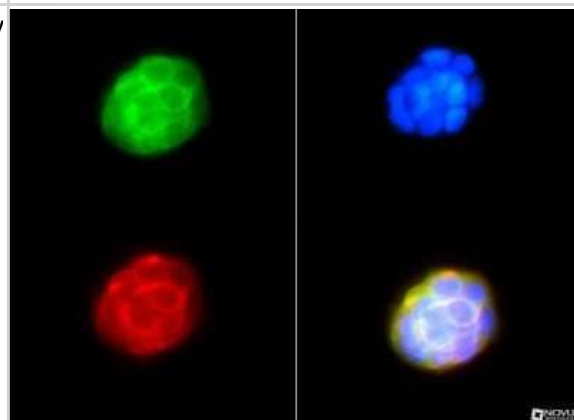


Images

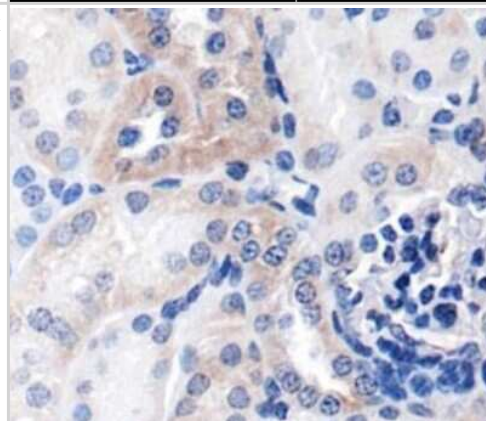
Western Blot: NFkB1/NFkB p105 Antibody [NBP1-77395] - WB analysis of NFkB in HepG2 whole cell lysate.



Immunocytochemistry/Immunofluorescence: NFkB1/NFkB p105 Antibody [NBP1-77395] - Antibody was tested in HeLa cells with FITC (green). Nuclei and actin were counterstained with Dapi (blue) and Phalloidin (red).



Immunohistochemistry: NFkB1/NFkB p105 Antibody [NBP1-77395] - IHC analysis of NFkB in mouse kidney using DAB with hematoxylin counterstain.



Simple Western: NFkB1/NFkB p105 Antibody [NBP1-77395] - Simple Western lane view shows a specific band for NFkB p105 in 0.5 mg/ml of HepG2 lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.



Publications

Sikorski K, Mehta A et al. A high-throughput pipeline for validation of antibodies. Nat Methods 2018-01-11 [PMID: 30377371] (Human)

Details:

Antibody validation based on denaturing gel electrophoresis of biotinylated cell lysates (PAGE) followed by mass spectrometry (MS) and antibody array analysis (MAP).

Erdogan S, Turkekul K, Dibirdik I et al. Midkine downregulation increases the efficacy of quercetin on prostate cancer stem cell survival and migration through PI3K/AKT and MAPK/ERK pathway Biomed. Pharmacother. 2018-08-21 [PMID: 30142541] (WB, Human)

Li J, Luco AL, Ochiatti B et al. Tumoral Vitamin D Synthesis by CYP27B1 1-alpha-Hydroxylase Delays Mammary Tumor Progression in the PyMT-MMTV Mouse Model and Its Action Involves NF-kB Modulation. Endocrinology. 2016-04-27 [PMID: 27119753] (ICC/IF, WB, Mouse)

Xu Y, Romero R, Miller D et al. An M1-like Macrophage Polarization in Decidual Tissue during Spontaneous Preterm Labor That Is Attenuated by Rosiglitazone Treatment J. Immunol. 2016-02-17 [PMID: 26889045] (IHC-Fr, Human)

Jaworski S, Sawosz E, Kutwin M et al. In vitro and in vivo effects of graphene oxide and reduced graphene oxide on glioblastoma. Int J Nanomedicine. 2015-03-11 [PMID: 25759581] (WB, Human)



Procedures

Western Blot protocol specific for NFkB1 antibody (NBP1-77395)

NFkB1/NFkB p105 Antibody:

Western Blot Protocol

1. Perform SDS-PAGE on samples to be analyzed, loading 40 ug of total protein per lane.
2. Transfer proteins to membrane according to the instructions provided by the manufacturer of the membrane and transfer apparatus.
3. Stain according to standard Ponceau S procedure (or similar product) to assess transfer success, and mark molecular weight standards where appropriate.
4. Rinse the blot.
5. Block the membrane using standard blocking buffer for at least 1 hour.
6. Wash the membrane in wash buffer three times for 10 minutes each.
7. Dilute primary antibody in blocking buffer and incubate 1 hour at room temperature.
8. Wash the membrane in wash buffer three times for 10 minutes each.
9. Apply the diluted HRP conjugated secondary antibody in blocking buffer (as per manufacturers instructions) and incubate 1 hour at room temperature.
10. Wash the blot in wash buffer 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.

*Note: Tween-20 can be added to the blocking or antibody dilution buffer at a final concentration of 0.05-0.2%.

Immunohistochemistry-Paraffin protocol for NFkB p105 Antibody (NBP1-77395)

NFkB1/NFkB p105 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 4C.
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 NFkB p105 Antibody (NBP1-77395)

NFkB1/NFkB p105 Antibody:

Immunocytochemistry Protocol

Culture cells to appropriate density in 35 mm culture dishes or 6-well plates.

1. Remove culture medium and add 10% formalin to the dish. Fix at room temperature for 30 minutes.
2. Remove the formalin and add ice cold methanol. Incubate for 5-10 minutes.
3. Remove methanol and add washing solution (i.e. PBS). Be sure to not let the specimen dry out. Wash three times for 10 minutes.
4. To block nonspecific antibody binding incubate in 10% normal goat serum from 1 hour to overnight at room temperature.
5. Add primary antibody at appropriate dilution and incubate at room temperature from 2 hours to overnight at room temperature.
6. Remove primary antibody and replace with washing solution. Wash three times for 10 minutes.
7. Add secondary antibody at appropriate dilution. Incubate for 1 hour at room temperature.
8. Remove antibody and replace with wash solution, then wash for 10 minutes. Add Hoechst 33258 to wash solution at 1:25,000 and incubate for 10 minutes. Wash a third time for 10 minutes.
9. Cells can be viewed directly after washing. The plates can also be stored in PBS containing Azide covered in Parafilm (TM). Cells can also be cover-slipped using Fluoromount, with appropriate sealing.

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





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Products Related to NBP1-77395

HAF008	Goat anti-Rabbit IgG Secondary Antibody [HRP]
NB7160	Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]
NBP2-24891	Rabbit IgG Isotype Control
NBP1-77395B	NFkB1/NFkB p105 Antibody [Biotin]

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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