Product Datasheet

ALKBH5 Antibody - BSA Free NBP2-78817SS

Unit Size: 0.025 mg

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

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NBP2-78817SS

ALKBH5 Antibody - BSA Free

Product Information	
Unit Size	0.025 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
Purity	Immunogen affinity purified
Buffer	PBS
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Product Description	
Description	Novus Biologicals Rabbit ALKBH5 Antibody - BSA Free (NBP2-78817) is a polyclonal antibody validated for use in IHC, WB and ICC/IF. All Novus Biologicals antibodies are covered by our 100% guarantee.
Host	Rabbit
Gene ID	54890
Gene Symbol	ALKBH5
Species	Human, Mouse, Rat
Immunogen	Partial recombinant protein made to an N-terminal portion of the human ALKB5H protein (amino acids 1-394) [UniProt 54890].

Product Application Details	
Applications	Western Blot, Immunohistochemistry-Paraffin, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry
Recommended Dilutions	Western Blot 1 - 2 ug/ml, Immunohistochemistry 1:200 - 1: 400, Immunocytochemistry/ Immunofluorescence 2 - 4 ug/ml, Immunohistochemistry-Paraffin 1:200 - 1: 400

Images

Immunocytochemistry/Immunofluorescence: ALKBH5 Antibody [NBP2-78817] - 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 anti-ALKBH5 Antibody NBP2-78817 at 1 ug/ml for 60 minutes at room temperature 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.



Procedures

Western Blot Protocol for ALKBH5 Antibody (NBP2-78817)

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 ALKBH5 Antibody (NBP2-78817)

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 ALKBH5 Antibody (NBP2-78817) 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 NBP2-78817SS

HAF008 Goat anti-Rabbit IgG Secondary Antibody [HRP]

NB7160 Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]

NBP1-82188PEP ALKBH5 Recombinant Protein Antigen

NBL1-11567 H4/j Overexpression Lysate

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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