

Product Datasheet

Lysine (K)-specific Demethylase 4A/KDM4A/JMJD2A Antibody - BSA Free NBP1-49602

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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Updated 4/13/2025 v.20.1

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NBP1-49602

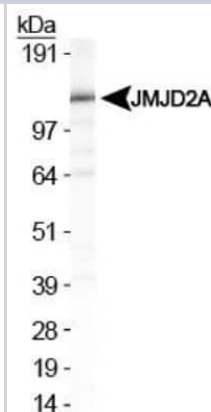
Lysine (K)-specific Demethylase 4A/KDM4A/JMJD2A Antibody - BSA Free

Product Information	
Unit Size	0.1 ml
Concentration	0.53 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
Product Description	
Host	Rabbit
Gene ID	9682
Gene Symbol	KDM4A
Species	Human, Mouse
Immunogen	A genomic peptide made to an internal region of the human JMJD2A protein (within residues 320-460). [Swiss-Prot O75164]
Notes	Manufactured by Genomic Antibody Technology™. GAT FAQs
Product Application Details	
Applications	Western Blot, Simple Western, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin
Recommended Dilutions	Western Blot 1:1000 - 1:2000, Simple Western 1:500, Immunohistochemistry 1:50, Immunocytochemistry/ Immunofluorescence 1:100, Immunohistochemistry-Paraffin 1:50
Application Notes	<p>This JMJD2A antibody is useful for Immunohistochemistry on paraffin tissues, Immunocytochemistry/Immunofluorescence and Western blot where a band is seen ~119 kDa. Prior to immunostaining paraffin tissues, antigen retrieval with sodium citrate buffer (pH 6.0) is recommended.</p> <p>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 HeLa lysate 0.5 mg/mL, separated by Size, antibody dilution of 1:500, apparent MW was 146 kDa. Separated by Size-Wes, Sally Sue/Peggy Sue.</p>

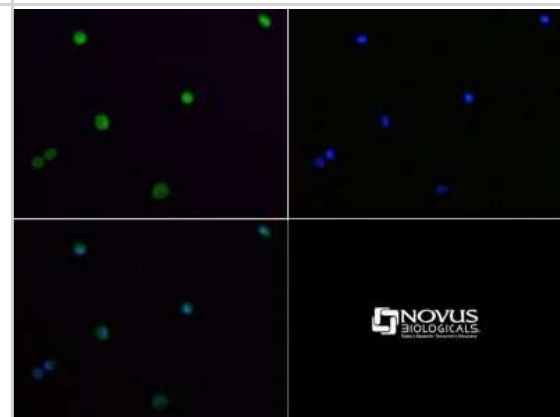


Images

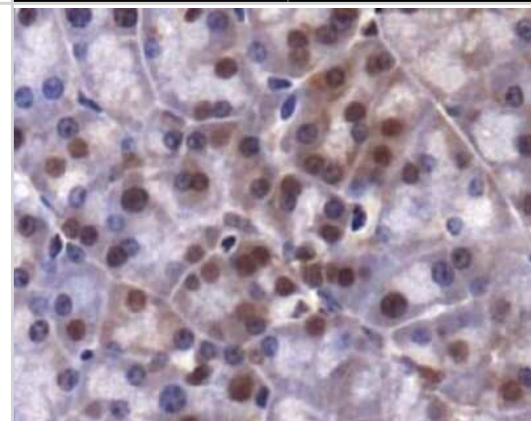
Western Blot: Lysine (K)-specific Demethylase 4A/KDM4A/JMJD2A Antibody [NBP1-49602] - Analysis of JMJD2A in HeLa nuclear extracts.



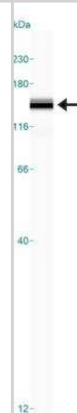
Immunocytochemistry/Immunofluorescence: Lysine (K)-specific Demethylase 4A/KDM4A/JMJD2A Antibody [NBP1-49602] - Antibody was tested at 1:50 in HeLa cells with FITC (green). Nuclei (Blue) were counterstained with Dapi (blue).



Immunohistochemistry: Lysine (K)-specific Demethylase 4A/KDM4A/JMJD2A Antibody [NBP1-49602] - Staining of JMJD2A in paraffin embedded mouse pancreas.



Simple Western: Lysine (K)-specific Demethylase 4A/KDM4A/JMJD2A Antibody [NBP1-49602] - Simple Western lane view shows a specific band for JMJD2A in 0.5 mg/ml of HeLa lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.



Procedures

Western Blot protocol for JMJD2A Antibody (NBP1-49602)

Lysine (K)-specific Demethylase 4A/KDM4A/JMJD2A 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 the rabbit anti-JMJD2A primary antibody (NBP1-49602) 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 diluted rabbit-IgG 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 JMJD2A Antibody (NBP1-49602)

Lysine (K)-specific Demethylase 4A/KDM4A/JMJD2A 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 JMJD2A Antibody (NBP1-49602)

Lysine (K)-specific Demethylase 4A/KDM4A/JMJD2A 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 NBP1-49602

NB800-PC9	HeLa Nuclear Cell Lysate
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.

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