

Product Datasheet

CYP46A1 Antibody NB400-140

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 10/23/2024 v.20.1

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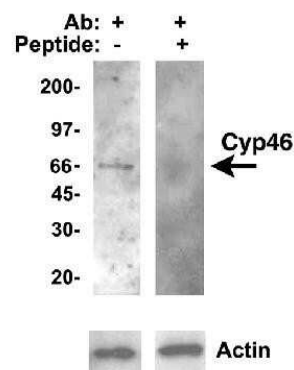
NB400-140**CYP46A1 Antibody**

Product Information	
Unit Size	0.1 ml
Concentration	This product is unpurified. The exact concentration of antibody is not quantifiable.
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	No Preservative
Isotype	IgG
Purity	Unpurified
Buffer	Whole antisera
Product Description	
Host	Rabbit
Gene ID	10858
Gene Symbol	CYP46A1
Species	Human, Rabbit, Mouse (Negative)
Reactivity Notes	Does not react with mouse.
Immunogen	A synthetic peptide corresponding to residues between 200-250 of human CYP46A1. [UniProt# Q9Y6A2]
Product Application Details	
Applications	Western Blot, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin
Recommended Dilutions	Western Blot 1:1000, Immunohistochemistry 1:200, Immunocytochemistry/ Immunofluorescence 1:500, Immunohistochemistry-Paraffin 1:200
Application Notes	This CYP46A1 antibody is useful for Immunocytochemistry/Immunofluorescence, Western blot and Immunohistochemistry paraffin embedded sections. In ICC/IF endoplasmic reticulum staining was observed.

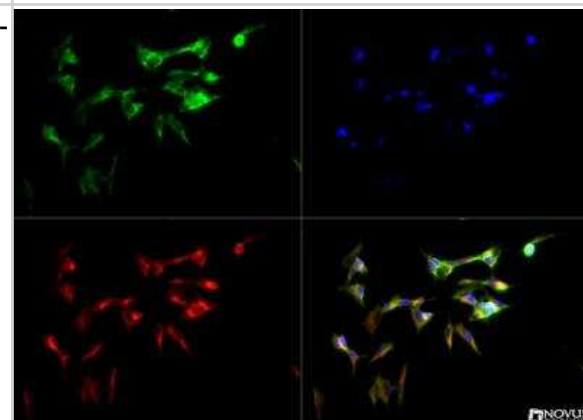


Images

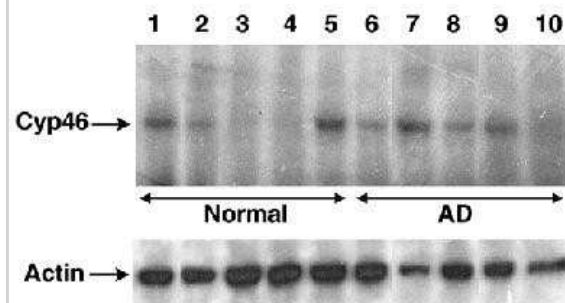
Western Blot: CYP46A1 Antibody [NB400-140] - Blocking experiment using NB 400-140 in HEK293 cells overexpressing CYP46A1.



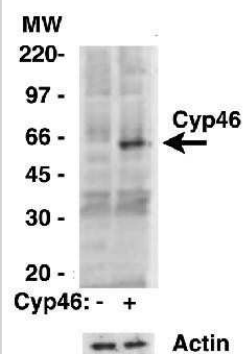
Immunocytochemistry/Immunofluorescence: CYP46A1 Antibody [NB400-140] - CYP46A1 antibody was tested in SH-SY5Y cells with DyLight 488 (green). Nuclei and alpha-tubulin were counterstained with DAPI (blue) and DyLight 550 (red).



Western Blot: CYP46A1 Antibody [NB400-140] - Detection of Cyp46 in human brain lysates.



Western Blot: CYP46A1 Antibody [NB400-140] - Analysis of Cyp-46 in HEK293 cells overexpressing CYP46A1 or transfected with an empty vector using NB400-140.



Publications

Zhang J, Zhang F, Wu J et al. Glutamate affects cholesterol homeostasis within the brain via the up-regulation of CYP46A1 and ApoE Toxicology 2020-01-22 [PMID: 31981724] (WB, Human)

Brown III, J et al. Differential Expression of Cholesterol Hydroxylase in Alzheimer's Disease. The Journal of Biological Chemistry 279(33): 34674-34681. 2004-01-01 [PMID: 15148325] (ICC/IF, WB, IF/IHC, Human)



Procedures

Immunocytochemistry/Immunofluorescence protocol for CYP46A1 Antibody (NB400-140)

CYP46A1 Antibody:

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.

Western Blot protocol for CYP46A1 Antibody (NB400-140)

CYP46A1 Antibody:

Western Blot Protocol

1. Perform SDS-PAGE (4-12% MOPS) on samples to be analyzed, loading 25 ug of total protein per lane.
 2. Transfer proteins to Nitrocellulose according to the instructions provided by the manufacturer of the transfer apparatus.
 3. Rinse membrane with dH₂O and then stain the blot using Ponceau S for 1-2 minutes to access the transfer of proteins onto the nitrocellulose membrane. Rinse the blot in water to remove excess stain and mark the lane locations and locations of molecular weight markers using a pencil.
 4. Rinse the blot in TBS for approximately 5 minutes.
 5. Block the membrane using 5% NFDM + 1% BSA in TBS + Tween, 1 hour at RT.
 6. Rinse the membrane in dH₂O and then wash the membrane in wash buffer [TBS + 0.1% Tween] 3 times for 10 minutes each.
 7. Dilute the rabbit anti-CYP46A1 primary antibody (NB400-140) in blocking buffer and incubate 1 hour at room temperature.
 8. Rinse the membrane in dH₂O and then wash the membrane in wash buffer [TBS + 0.1% Tween] 3 times for 10 minutes each.
 9. Apply the 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 [TBS + 0.1% Tween] 3 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 (Pierce ECL).
- Note: Tween-20 can be added to the blocking or antibody dilution buffer at a final concentration of 0.05-0.2%, provided it does not interfere with antibody-antigen binding.



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Products Related to NB400-140

NBL1-09694	CYP46A1 Overexpression 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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