abcam

ab300489

Anti-NSD3 antibody [EPR25189-8]

Recombinant | RabMAb | KO Validated | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR25189-8
Purification technique	Affinity purification Protein A
Concentration	0.499 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

WB

Tested

Species	Human
Dilution info	1/1000 - 1/2000
Notes	-

Species	Mouse

Dilution info	1/1000 - 1/2000
Notes	-

Species	Rat
Dilution info	1/1000 - 1/2000
Notes	-

IHC-P

Tested

Species	Human
Dilution info	1/500
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Species	Mouse
Dilution info	1/500
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Species	Rat
Dilution info	1/500
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

ICC/IF

Tested

Species	Human
Dilution info	1/50
Notes	-

Species	Mouse
Dilution info	1/50
Notes	-

Expected

Species	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/500
Notes	-

Species	Mouse
Dilution info	1/500
Notes	-

Expected

Species	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

IP

Tested

Species	Human
Dilution info	1/30
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.

Notes	-

Target data

See full target information NSD3

Function	Histone methyltransferase. Preferentially dimethylates 'Lys-4' and 'Lys-27' of histone H3 forming H3K4me2 and H3K27me2. H3 'Lys-4' methylation represents a specific tag for epigenetic transcriptional activation, while 'Lys-27' is a mark for transcriptional repression.

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free batch production

For more information, read more on recombinant antibodies.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	NSD3 also known as WHSC1L1 is a protein involved in gene expression regulation through its role as a histone methyltransferase. It has a molecular mass of approximately 170 kDa. This protein specifically modifies lysine residues on histone tails which influences chromatin structure and gene transcription. NSD3 is broadly expressed in various tissues but higher levels appear in actively dividing cells such as those found in the brain and reproductive organs.
Biological function summary	NSD3 plays an important role in the regulation of chromatin state and is part of the larger SETD2 family of methyltransferases. It forms part of complexes that influence transcriptional activation and repression by altering chromatin accessibility. These complexes allow NSD3 to exert control over genes involved in cell proliferation and differentiation impacting cellular processes critical to development and tissue maintenance.
Pathways	NSD3 integrates into chromatin remodeling and gene expression pathways. It interacts with other methyltransferases and proteins like NSD1 and NSD2 to regulate transcriptional programs essential for cellular growth. It is part of pathways that modulate key processes such as the cell cycle and DNA damage response indicating its role in maintaining genomic stability and regulating cell fate decisions.
Associated diseases and disorders	NSD3 has implications in cancer development and progression. It is particularly linked to certain types of breast cancer where overexpression leads to uncontrolled cell growth. In these cases NSD3 influences oncogenic pathways and interacts with proteins such as BRD4 to drive tumorigenesis. Its dysregulation might also contribute to neurodevelopmental disorders by affecting proteins involved in neural development and connectivity.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

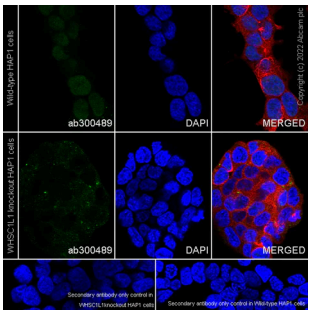
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

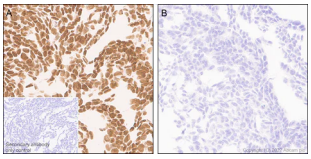
Full details and terms and conditions can be found here:
Terms & Conditions.

11 product images



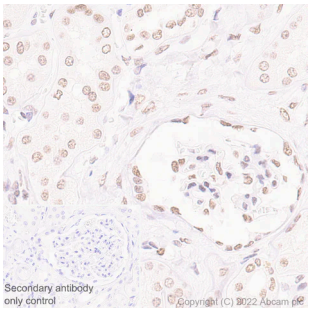
Immunocytochemistry/ Immunofluorescence - Anti-NSD3 antibody [EPR25189-8] (ab300489)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized WHSC1L1 KO HAP1 (WHSC1L1 knockout human chronic myelogenous leukemia near-haploid cell) cells labelling NSD3 with ab300489 at 1/500 (0.998 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 2 ug/ml dilution (Green). Confocal image showing nuclear staining in wild-type HAP1 cell line and no staining in WHSC1L1 knockout HAP1 cell line is observed. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 2.5 ug/ml dilution (Red). The Nuclear counterstain was DAPI (Blue). Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 2 ug/ml dilution.



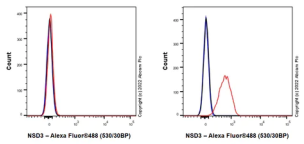
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NSD3 antibody [EPR25189-8] (ab300489)

Immunohistochemical analysis of paraffin-embedded A Wild-type HAP1 (H tissue labeling NSD3 with ab300489 at 1/500 (0.998 ug/ml) followed by a ready to use LeicaDS9800 (Bond☒ Polymer Refine Detection) was used. Positive staining on (A) wild-type HAP1 cell pellet, no staining on (B) WHSC1L1 knockout HAP1 cell pellet. The section was incubated with ab300489 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond☒ Polymer Refine Detection) was used.Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



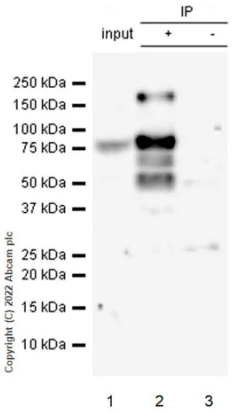
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NSD3 antibody [EPR25189-8] (ab300489)

Immunohistochemical analysis of paraffin-embedded Human kidney tissue labeling NSD3 with ab300489 at 1/500 (0.998 ug/ml) followed by a ready to use LeicaDS9800 (Bond☒ Polymer Refine Detection) was used. Nuclear staining on human kidney. The section was incubated with ab300489 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond☒ Polymer Refine Detection) was used.Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Flow Cytometry (Intracellular) - Anti-NSD3 antibody [EPR25189-8] (ab300489)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized Wild-type HAP1 (human chronic myelogenous leukemia near-haploid cell, Right) / NSD3 knockout HAP1(Left) cells labelling NSD3 with ab300489 at 1/500 dilution (0.1ug) (Red) (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/2000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-NSD3 antibody [EPR25189-8] (ab300489)

NSD3 was immunoprecipitated from 0.35 mg HAP1 (human chronic myelogenous leukemia near-haploid cell line) whole cell lysate 10 ug with ab300489 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab300489 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(ab131366) was used at 1/5000 dilution.
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

This blot was developed using a high sensitivity ECL substrate. The high-sensitivity ECL substrate used allows for the detection of proteins in the mid-femtogram range.

Lysates were freshly made and used immediately to minimize protein degradation.

Extra bands at 70 and 50 kDa observed in lane 2 may be caused by degradation during incubation.

All lanes:
Immunoprecipitation - Anti-NSD3 antibody [EPR25189-8] (ab300489) at 1/1000 dilution

Lane 1:
HAP1 (human chronic myelogenous leukemia near-haploid cell line) whole cell lysate at 10 µg

Lane 2:
ab300489 at 1/30 IP in HAP1 whole cell lysate

Lane 3:
Rabbit monoclonal IgG (ab172730) instead of ab300489 in HAP1 whole cell lysate at 10 µg

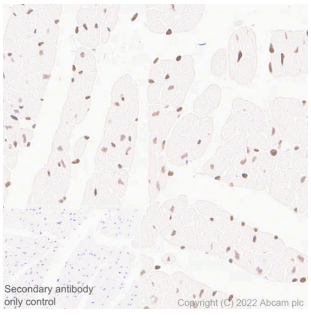
Secondary

All lanes:
Immunoprecipitation - VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/5000 dilution

Developed using the ECL technique.

Observed band size: 85 kDa, 180 kDa

Exposure time: 125s

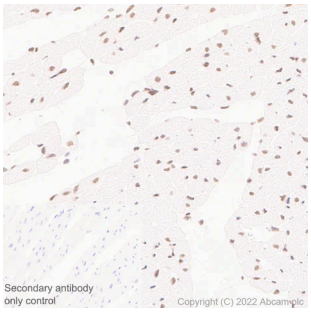


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NSD3 antibody [EPR25189-8] (ab300489)

NSD3 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Mouse cardiac muscle using rabbit Anti-NSD3 antibody

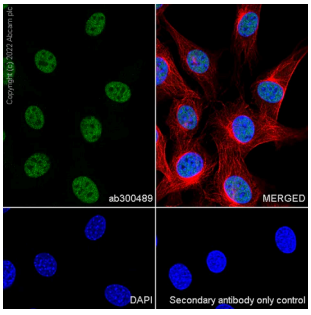
Immunohistochemical analysis of paraffin-embedded Mouse cardiac muscle tissue labeling NSD3 with ab300489 at 1/500 (0.998 ug/ml) followed by a ready to use LeicaDS9800 (Bond☒ Polymer

Refine Detection) was used. Nuclear staining on mouse cardiac muscle. The section was incubated with ab300489 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond☒ Polymer Refine Detection) was used.Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



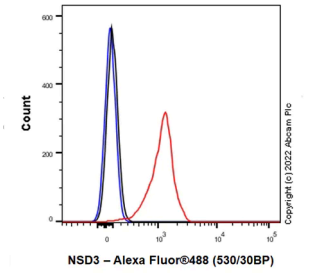
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NSD3 antibody [EPR25189-8] (ab300489)

Immunohistochemical analysis of paraffin-embedded Rat cardiac muscle tissue labeling NSD3 with ab300489 at 1/500 (0.998 ug/ml) followed by a ready to use LeicaDS9800 (Bond☒ Polymer Refine Detection) was used. Nuclear staining on rat cardiac muscle. The section was incubated with ab300489 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond☒ Polymer Refine Detection) was used.Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



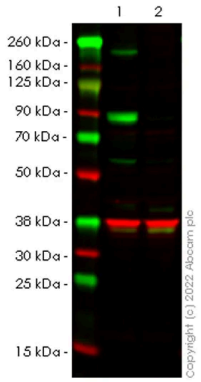
Immunocytochemistry/ Immunofluorescence - Anti-NSD3 antibody [EPR25189-8] (ab300489)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized NIH/3T3 (mouse embryonic fibroblast) cells labelling NSD3 with ab300489 at 1/50 (9.98 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 2 ug/ml dilution (Green). Confocal image showing nuclear staining in NIH/3T3 cell line is observed. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 2.5 ug/ml dilution (Red). The Nuclear counterstain was DAPI (Blue). Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 2 ug/ml dilution.



Flow Cytometry (Intracellular) - Anti-NSD3 antibody [EPR25189-8] (ab300489)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized NIH/3T3 (mouse embryonic fibroblast) cells labelling NSD3 with ab300489 at 1/500 dilution (0.1ug) (Red) (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/2000 dilution was used as the secondary antibody.



Western blot - Anti-NSD3 antibody [EPR25189-8] (ab300489)

Blocking and diluting buffer and concentration: Intercept® (TBS) Blocking Buffer diluted with an equal volume of 0.1% TBS
All lanes:
Western blot - Anti-NSD3 antibody [EPR25189-8] (ab300489) at 1/2000 dilution

Lane 1:
Wild-type HAP1 (human chronic myelogenous leukemia near-haploid cell), whole cell lysate

Lane 2:
WHSC1L1 knockout HAP1 whole cell lysate

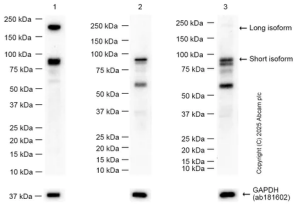
Secondary

Lanes 1 - 2:
Western blot - Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/20000 dilution

Lanes 1 - 2:
Western blot - Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) at 1/20000 dilution

Performed under reducing conditions.

Observed band size: 85 kDa, 180 kDa



Western blot - Anti-NSD3 antibody [EPR25189-8] (ab300489)

NSD3 Western blot staining using rabbit Anti-NSD3 antibody

In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) staining at 1/1000000 dilution.

Lysates were freshly made and used for Western blotting immediately to minimize protein degradation.

All lanes:
Western blot - Anti-NSD3 antibody [EPR25189-8] (ab300489) at 1/1000 dilution

Lane 1:
293T (human embryonic kidney epithelial cell), whole cell lysate at 20 µg

Lane 2:
NIH/3T3 (mouse embryonic fibroblast), whole cell lysate at 20 µg

Lane 3:
PC-12 (rat adrenal gland pheochromocytoma), whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 85 kDa, 180 kDa

Observed band size: 85 kDa, 180 kDa

Exposure time: 180s

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.