abcam

ab277534

# Anti-NSUN2/SAKI antibody [EPR24140-93] - BSA and Azide free

Recombinant | RabMAb

## Key facts

| | |
|---|---|
| **Isotype** | IgG |
| **Host species** | Rabbit |
| **Storage buffer** | Constituents: PBS |
| **Form** | Liquid |
| **Clonality** | Monoclonal |
| **Immunogen** | The exact immunogen used to generate this antibody is proprietary information. |
| **Carrier free** | Yes |
| **Clone number** | EPR24140-93 |
| **Purification technique** | Affinity purification Protein A |
| **Concentration** | 0.982 mg/mL The concentration of this product may be batch-dependent<br>Batch concentration finder → |

## Reactivity data

## ICC/IF

### Tested

| | |
|---|---|
| **Species** | Human, Mouse |
| **Dilution info** | - |

| | |
|---|---|
| **Notes** | - |

### Expected

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

## IP

### Tested

| | |
|---|---|
| **Species** | Human, Mouse |
| **Dilution info** | - |
| **Notes** | - |

### Expected

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

## WB

### Tested

| | |
|---|---|
| **Species** | Human, Mouse, Rat |
| **Dilution info** | - |
| **Notes** | - |

## Flow Cyt (Intra)

### Tested

| | |
|---|---|
| **Species** | Human, Mouse |
| **Dilution info** | - |
| **Notes** | - |

### Expected

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

## IHC-P

### Tested

| | |
|---|---|
| **Species** | Human, Mouse, Rat |
| **Dilution info** | - |
| **Notes** | - |

# Target data

See full target information NSUN2

**Function**

RNA cytosine C(5)-methyltransferase that methylates cytosine to 5-methylcytosine (m5C) in various RNAs, such as tRNAs, mRNAs and some long non-coding RNAs (lncRNAs) (PubMed:17071714, PubMed:22995836, PubMed:31199786, PubMed:31358969). Involved in various processes, such as epidermal stem cell differentiation, testis differentiation and maternal to zygotic transition during early development: acts by increasing protein synthesis; cytosine C(5)-methylation promoting tRNA stability and preventing mRNA decay (PubMed:31199786). Methylates cytosine to 5-methylcytosine (m5C) at positions 34 and 48 of intron-containing tRNA(Leu)(CAA) precursors, and at positions 48, 49 and 50 of tRNA(Gly)(GCC) precursors (PubMed:17071714, PubMed:22995836, PubMed:31199786). tRNA methylation is required generation of RNA fragments derived from tRNAs (tRFs) (PubMed:31199786). Also mediates C(5)-methylation of mitochondrial tRNAs (PubMed:31276587). Catalyzes cytosine C(5)-methylation of mRNAs, leading to stabilize them and prevent mRNA decay: mRNA stabilization involves YBX1 that specifically recognizes and binds m5C-modified transcripts (PubMed:22395603, PubMed:31358969, PubMed:34556860). Cytosine C(5)-methylation of mRNAs also regulates mRNA export: methylated transcripts are specifically recognized by THOC4/ALYREF, which mediates mRNA nucleo-cytoplasmic shuttling (PubMed:28418038). Also mediates cytosine C(5)-methylation of non-coding RNAs, such as vault RNAs (vtRNAs), promoting their processing into regulatory small RNAs (PubMed:23871666). Cytosine C(5)-methylation of vtRNA VTRNA1.1 promotes its processing into small-vault RNA4 (svRNA4) and regulates epidermal differentiation (PubMed:31186410). May act downstream of Myc to regulate epidermal cell growth and proliferation (By similarity). Required for proper spindle assembly and chromosome segregation, independently of its methyltransferase activity (PubMed:19596847).

## Storage

| | |
|---|---|
| **Shipped at conditions** | Blue Ice |
| **Appropriate short-term storage conditions** | +4°C |
| **Appropriate long-term storage conditions** | +4°C |

## Notes

ab277534 is the carrier-free version of ab259941.

**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.

**Conjugation ready**

Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

**Compatibility**

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

## Supplementary info

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

| | |
|---|---|
| **Activity summary** | NSUN2 also known as SAKI is a methyltransferase enzyme that adds a methyl group to cytosine within nucleic acids most often in RNA. It has a molecular mass of about 83 kDa. This protein is widely expressed in different tissues but shows high expression levels in the brain and reproductive organs. NSUN2 modifies RNA molecules impacting their function and stability which is essential in various cellular processes. |

| | |
|---|---|
| **Biological function summary** | NSUN2 participates in RNA metabolism and processing. By introducing m5C (5-methylcytosine) modifications to RNA NSUN2 influences RNA stability transport and translation. This protein is a part of larger complexes that regulate diverse cellular functions and maintains transcriptome integrity. Alterations in NSUN2 activity can affect cellular proliferation and differentiation highlighting its role in maintaining normal cell function. |
| **Pathways** | NSUN2 plays an integral role in the epitranscriptomic regulation pathway in particular RNA modification and gene expression regulation pathways. It acts alongside proteins like TRMT2A and TRMT61A which are involved in similar processes of RNA methylation and modification. These pathways help maintain a balance in gene expression profiles necessary for proper cellular function and response to external signals. |
| **Associated diseases and disorders** | Mutations or dysfunctions in NSUN2 are associated with mental retardation and cancer. In neurodevelopmental disorders altered NSUN2 activity disrupts normal brain development potentially linking it with proteins like FTSJ1 another RNA methyltransferase. Similarly in cancer NSUN2 dysregulation leads to aberrant cell proliferation and tumorigenesis often in connection with oncogenic pathways and other modified proteins. |

## 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**

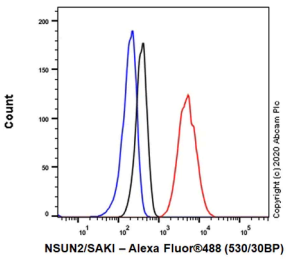
We do not recommend this combination. It is not covered by our product promise.

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.

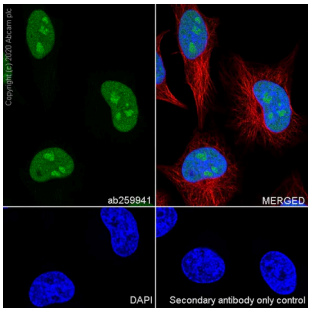
## 12 product images




### Flow Cytometry (Intracellular) - Anti-NSUN2/SAKI antibody [EPR24140-93] - BSA and Azide free (ab277534)

NSUN2/SAKI Flow Cytometry (Intracellular) staining using rabbit Anti-NSUN2/SAKI antibody

This data was developed using ab259941, the same antibody clone in a different buffer formulation.
Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol-permeabilized HeLa (human cervix adenocarcinoma epithelial cell) cells labelling NSUN2/SAKI with ab259941 at 1/500 dilution (Red) compared with a Rabbit monoclonal IgG (ab172730) isotype control (Black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor®488, ab150077) at 1/2000 dilution was used as the secondary antibody.

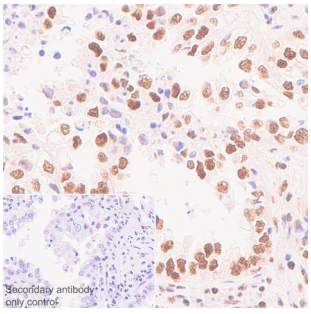



### Immunocytochemistry/ Immunofluorescence - Anti-NSUN2/SAKI antibody [EPR24140-93] - BSA and Azide free (ab277534)

NSUN2/SAKI Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-NSUN2/SAKI antibody

This data was developed using ab259941, the same antibody clone in a different buffer formulation.
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (human epithelial cell line from cervix adenocarcinoma) cells labelling NSUN2/SAKI with ab259941 at 1/1000 dilution (0.503 µg/ml), followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor®488) antibody at 1/1000 dilution (Green). Confocal image showing weak cytoplasmic and strong nuclear staining in HeLa cells. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.



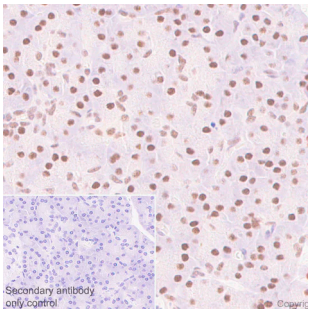

### Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NSUN2/SAKI antibody [EPR24140-93] - BSA and Azide free (ab277534)

NSUN2/SAKI Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-NSUN2/SAKI antibody

This data was developed using ab259941, the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue labeling NSUN2/SAKI with ab259941 at 1/200 dilution (2.515 µg/ml) followed by a ready to use LeicaDS9800 (Bond® Polymer Refine Detection). Nuclear staining on human colon carcinoma (PMID: 16713953). The section was incubated with ab259941 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).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.



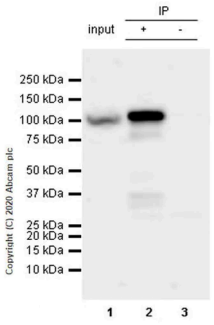

## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NSUN2/SAKI antibody [EPR24140-93] - BSA and Azide free (ab277534)

NSUN2/SAKI Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-NSUN2/SAKI antibody

This data was developed using ab259941, the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded human pancreas tissue labeling NSUN2/SAKI with ab259941 at 1/200 dilution (2.515 µg/ml) followed by a ready to use LeicaDS9800 (Bond® Polymer Refine Detection). Nuclear staining on human pancreas (PMID: 16713953). The section was incubated with ab259941 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).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.




## Immunoprecipitation - Anti-NSUN2/SAKI antibody [EPR24140-93] - BSA and Azide free (ab277534)

This data was developed using ab259941, the same antibody clone in a different buffer formulation.
NSUN2/SAKI was immunoprecipitated from 0.35 mg HEK-293 (human embryonic kidney epithelial cell) whole cell lysate with ab259941 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab259941 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP)(ab131366) was used at 1/5000 dilution.

**Lane 1:** HEK-293 (human embryonic kidney epithelial cell) whole cell lysate 10 ug

**Lane 2:** ab259941 IP in HEK-293 whole cell lysate

**Lane 3:** Rabbit monoclonal IgG (ab172730) instead of ab259941 in HEK-293 whole cell lysate

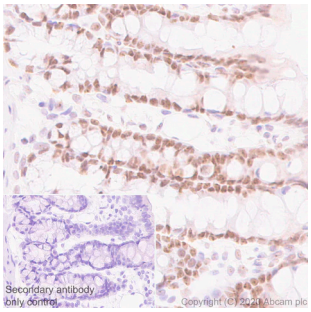
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 15 seconds.

All lanes:
Immunoprecipitation - Anti-NSUN2/SAKI antibody [EPR24140-93] (ab259941)

Predicted band size: 86 kDa

Observed band size: 100 kDa



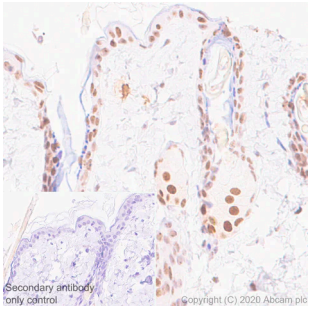

## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NSUN2/SAKI antibody [EPR24140-93] - BSA and Azide free (ab277534)

NSUN2/SAKI Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-NSUN2/SAKI antibody

This data was developed using ab259941, the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded rat colon tissue labeling NSUN2/SAKI with ab259941 at 1/200 dilution (2.515 µg/ml) followed by a ready to use LeicaDS9800 (Bond® Polymer Refine Detection). Nuclear staining on rat colon.The section was incubated with ab259941 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).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.



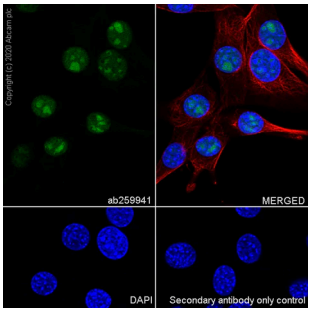

## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NSUN2/SAKI antibody [EPR24140-93] - BSA and Azide free (ab277534)

NSUN2/SAKI Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-NSUN2/SAKI antibody

This data was developed using ab259941, the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded mouse skin tissue labeling NSUN2/SAKI with ab259941 at 1/200 dilution (2.515 µg/ml) followed by a ready to use LeicaDS9800 (Bond® Polymer Refine Detection). Nuclear staining on mouse skin (PMID: 16713953). The section was incubated with ab259941 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).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.



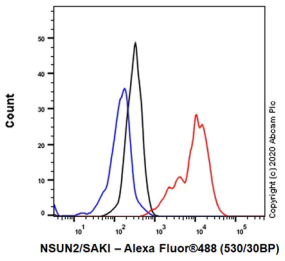

## Immunocytochemistry/ Immunofluorescence - Anti-NSUN2/SAKI antibody [EPR24140-93] - BSA and Azide free (ab277534)

NSUN2/SAKI Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-NSUN2/SAKI antibody

This data was developed using ab259941, the same antibody clone in a different buffer formulation.
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized NIH/3T3 (mouse embryonic fibroblast cell line) cells labelling NSUN2/SAKI with ab259941 at 1/1000 (0.503 µg/ml) dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor®488) antibody at 1/1000 dilution (Green). Confocal image showing weak cytoplasmic and strong nuclear

staining in NIH/3T3 cells. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor®594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).

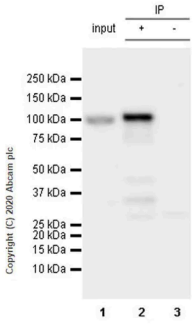
Secondary antibody only control: Secondary antibody is ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.




## Flow Cytometry (Intracellular) - Anti-NSUN2/SAKI antibody [EPR24140-93] - BSA and Azide free (ab277534)

NSUN2/SAKI Flow Cytometry (Intracellular) staining using rabbit Anti-NSUN2/SAKI antibody

This data was developed using ab259941, the same antibody clone in a different buffer formulation.
Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol-permeabilized ES-D3 (mouse embryonic multipotent stem cell) cells labelling NSUN2/SAKI with ab259941 at 1/50 dilution (Red) compared with a Rabbit monoclonal IgG (ab172730) isotype control (Black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor®488, ab150077) at 1/2000 dilution was used as the secondary antibody.




## Immunoprecipitation - Anti-NSUN2/SAKI antibody [EPR24140-93] - BSA and Azide free (ab277534)

This data was developed using ab259941, the same antibody clone in a different buffer formulation.
NSUN2/SAKI was immunoprecipitated from 0.35 mg mouse spleen tissue lysate with ab259941 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab259941 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP)(ab131366) was used at 1/5000 dilution.

**Lane 1:** Mouse spleen tissue lysate 10 ug

**Lane 2:** ab259941 IP in Mouse spleen tissue lysate

**Lane 3:** Rabbit monoclonal IgG (ab172730) instead of ab259941 in mouse spleen tissue lysate

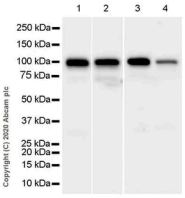
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 15 seconds.

All lanes:
Immunoprecipitation - Anti-NSUN2/SAKI antibody [EPR24140-93] (ab259941)

Predicted band size: 86 kDa

Observed band size: 100 kDa




## Western blot - Anti-NSUN2/SAKI antibody [EPR24140-93] - BSA and Azide free (ab277534)

NSUN2/SAKI Western blot staining using rabbit Anti-NSUN2/SAKI antibody

This data was developed using ab259941, the same antibody clone in a different buffer formulation.
Blocking and diluting buffer and concentration: 5% NFDM/TBST.

The observed MW is consistent with what has described in the literature (PMID: 23604283, 25233213, 27447970, 31487418).

Exposure times: Lane 1: 15 seconds

Lane 2-4: 48 seconds.

All lanes:
Western blot - Anti-NSUN2/SAKI antibody [EPR24140-93] (ab259941) at 1/1000 dilution

Lane 1:
HepG2 (human hepatocellar carcinoma epithelial cell) whole cell lysate at 20 µg

Lane 2:
HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Lane 3:
HEK-293 (human embryonic kidney epithelial cell) whole cell lysate at 20 µg

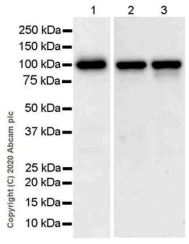
Lane 4:
A431 (human epidermoid carcinoma epithelial cell) whole cell lysate at 20 µg

Secondary

All lanes:
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (ab97051) at 1/50000 dilution

Predicted band size: 86 kDa

Observed band size: 100 kDa




## Western blot - Anti-NSUN2/SAKI antibody [EPR24140-93] - BSA and Azide free (ab277534)

NSUN2/SAKI Western blot staining using rabbit Anti-NSUN2/SAKI antibody

This data was developed using ab259941, the same antibody clone in a different buffer formulation.
Blocking and diluting buffer and concentration: 5% NFDM/TBST.

The observed MW is consistent with what has described in the literature (PMID: 27306184).

Exposure times: Lane 1: 48 seconds

Lane 2, 3: 3 minutes.

All lanes:
Western blot - Anti-NSUN2/SAKI antibody [EPR24140-93] (ab259941) at 1/1000 dilution

Lane 1:
ES-D3 (mouse embryonic mtipotent stem Cell) whole cell lysate at 20 µg

Lane 2:
Mouse spleen tissue lysate at 20 µg

Lane 3:
Rat cerebellum tissue lysate at 20 µg

Secondary

All lanes:
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (ab97051) at 1/50000 dilution

Predicted band size: 86 kDa

Observed band size: 100 kDa

---

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